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LIII
CONGRESO
INTERNACIONAL
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DE LA HEMATOLOGÍA
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Nota

El contenido de este Suplemento se transcribe tal como se recibió por parte de sus autores, razón por la que los editores no son responsables de las fallas u omisiones de ningún tipo.

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HEMATOPOIESIS

ORAL PRESENTATION

A1063

IN VITRO EFFECTS OF STROMAL CELLS EXPRESSING DIFFERENT LEVELS OF JAGGED-1 AND DELTA-1 ON THE GROWTH OF PRIMITIVE AND INTERMEDIATE CD34+ CELL SUBSETS FROM HUMAN CORD BLOOD. Fernández-Sánchez V*, Pelayo R**, Flores-Guzmán P*, Flores-Figueroa E***, Hector Mayani H*. *Hematopoietic Stem Cells Laboratory, Oncology Research Unit, Oncology Hospital, National Medical Center, IMSS, Mexico City, Mexico; **Lymphopoiesis Laboratory, Oncology Research Unit, Oncology Hospital, National Medical Center, IMSS, Mexico City, Mexico; ***Hematopoietic Microenvironment Laboratory, Oncology Research Unit, Oncology Hospital, National Medical Center, IMSS, Mexico City, Mexico.

Introduction: Within the hematopoietic system, Notch signaling has been shown to play key roles, regulating the development of multiple cell types. The Notch ligands, Delta1 and Jagged1, have been shown to maintain and expand primitive human hematopoietic cells capable of repopulating the marrow of NOD-SCID mice. Moreover, Notch ligands seem to be equally effective at modulating HSC growth regardless of the way they are presented to Notch receptors; indeed, soluble and cell-associated forms, as well as Notch ligands immobilized within a fibronectin matrix, have been shown to efficiently stimulate HSC self-renewal and expansion. **Objective:** In trying to contribute to our knowledge

on the role of Notch and its ligands within the human hematopoietic system, we have assessed the effects of the OP9 stroma cell line – naturally expressing Jagged-1 – transduced with either the Delta-1 gene (OP9-DL1 cells) or with vector alone (OP9-V), on the in vitro growth of two different hematopoietic cell populations. **Material and Methods:** Primitive (CD34+ CD38- Lin-) and intermediate (CD34+ CD38+ Lin-) CD34+ cell subsets from human cord blood were cultured in the presence of 7 stimulatory cytokines under four different conditions: cytokines alone (control); cytokines and mesenchymal stromal cells; cytokines and OP9-V cells; cytokines and OP9-DL1 cells. Proliferation and expansion were determined after 7 days of culture. **Results:** Culture of CD34+ CD38- Lin- cells in the presence of OP9-V or OP9-DL1 cells resulted in a significant increase in the production of new CD34+ CD38- Lin- cells (expansion), which expressed increased levels of Notch-1; in contrast, production of total nucleated cells (proliferation) was reduced, as compared to control conditions. In cultures of CD34+ CD38+ Lin- cells established in the presence of OP9-V or OP9-DL1 cells, expansion was similar to that observed in control conditions, whereas proliferation was also reduced. Interestingly, in these latter cultures we observed production of CD34+ CD38- Lin- cells. **Conclusions:** Our results indicate that, as compared to MSC, OP9 cells were more efficient at inducing self-renewal and/or de novo generation of primitive (CD34+ CD38- Lin-) cells, and suggest that such effects were due, at least in part, to the presence of Jagged-1 and DL1.

POSTERS

A1075

FRECUENCY OF MYELODISPLASIC SYNDROMES: SUB-TYPES ACCORDING THE WHO CRITERIA AND CLINICAL CHARACTERISTICS AND GLOBAL SURVIVAL IN PEDI-ATRIC PATIENTS. Jiménez-Hernández E, Contreras-Escobedo T, Arellano-Galindo J, Jaimes-Reyes EZ, Dueñas-González MT, Berges-García A, Bekker-Méndez C, Tiznado-García HM, Balam-Canul A, Mejía Aranguré JM. Hematología Pediátrica

UMAE, Centro Médico Nacional “La Raza” IMSS, Laboratorio De Investigación Hospital Infantil de México “Federico Gómez” SS, División de Laboratorios de Vigilancia E Investigación Epidemiológica UMAE, Centro Médico Nacional “La Raza” IMSS, Unidad de Investigación en Epidemiología Clínica, UMAE Hospital de Pediatría Centro Médico Nacional “Siglo XXI”, IMSS. Ciudad de México.

Introduction: Myelodysplastic syndromes diseases (MSD) are characterized by different level of alteration on the proliferation and differentiation of the hematopoietic stem cell, the presence

of cytopenia in peripheral blood (PB), bone marrow dysplasia (BMD) and predisposition of evolution to acute myeloid leukaemia (AML). Is an heterogeneous group of diseases with low incidence (0.5-1.8 cases per 100,000 inhabitants per year) and with a difficult clinical biological characterization. This is the main reason that there is not specific treatment. **Objective:** To know the frequency of the MSD, the subtypes according to the WHO, the clinical characteristics and global survival (GS) in a group of mexican pediatric patients. **Material and Methods:** A retrospective cohort study, descriptive and observational was done, patients less than 16 years old were included from Pediatric Haematology Service at the UMAE Centro Médico Nacional La Raza from January 2006 to March 2011. Statistical analysis. An analysis using descriptive statistical and Kaplan-Meier by SPSS software version 19 was done. **Results:** A total of 3250 patients admitted to the Pediatric Hematology Service at the UMAE Centro Médico Nacional la Raza in a period of five years, 31 out of them (0.95%) were diagnosed as MDS. 18/31(58.1%) males and 13/31 (41.9%) female, aged in an average of 7.5 years old (min 1 max 15 years old). Out of them: 17/31 (54.8%) with petechiae, 28/31 (90.3%) with thrombocytopenia, neutropenia in 25/31 (80.6%), monocitopenia in 24/31 (77.4%), and anaemia in 20/31 (64.5%). The most frequent subtype was the non-classifiable with 14/31 (45.2%) followed of refractory anaemia with excess of blasts-1 (RAEB-1) in 6/31 (19.4%), refractory cytopenia with multilineage dysplasia (RCMD) in 3/31 (9.7%) and refractory anaemia with excess of blasts-2 (RAEB-2) in 1/31 (3.2%). **Conclusions:** The frequency of MSD in our pediatric population was of 0.95% and the most frequent subtype was the non-classifiable. The global survival to five years was of 87.1%.

A1143

ROLE OF HELICOBACTER PYLORI INFECTION IN THE THERAPEUTIC RESPONSE OF PRIMARY IMMUNE THROMBOCYTOPENIA: EXPERIENCE FROM THE “INSTITUTO NACIONAL DE CIENCIAS MÉDICAS Y NUTRICIÓN SALVADOR ZUBIRÁN”. Scherling-Ocampo AA*, Agreda-Vásquez GP*. *Hematology Department, “Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán”, Mexico City, Mexico.

Introduction: Diagnosis of primary immune thrombocytopenia (PIT) is established once non-hematologic causes of thrombocytopenia are ruled out. Helicobacter pylori (HP) infection has been documented as a reversible cause of PIT and many possible mechanisms are considered. The prevalence of HP infection in Mexico ranges from 20 to 50 per cent, depending on the population age and location. Only few trials have explored the impact of HP eradication in the response to PIT treatment. Hence, this study assessed the role of HP eradication, as a strategy to modify the prevalence of a favorable response in patients with PIT. **Objective:** To determine the prevalence of therapeutic (complete or partial) response of PIT, before and after HP eradication treatment. **Material and Methods:** Design: analytic cross-sectional, retrospective and observational study.

Methods: All data were collected from the clinical notes from patients attended at the Hematology Department from the “Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán”, during the period from 2000-2010 according to the following selection criteria. Inclusion criteria: • Diagnosis of PIT, according the recently published International Consensus. • Positive test for HP infection, assessed by breath screening test. Elimination criteria: • Incomplete information regarding clinical follow up. • Evidence of any cause of immune thrombocytopenia (non-primary) during the clinical outcome. **Results:** Sample size was of 26 patients, 16 with HP infection and 10 without it. Table 1 shows baseline clinical characteristics and outcomes during follow-up. Table 2 describes the status of PIT response to immunosuppressor therapy before HP eradication treatment, in the HP-positive population. Only one patient improved the level of response (from PR to CR) after antibiotic scheme for HP. Patients with vs without HP infection have no difference regarding basal characteristics and outcome after immunosuppressor therapy. Regardless of the evidence that HP infection could cause secondary immune thrombocytopenia, its role in PIT is still controversial, since no significant benefit after eradication therapy was observed in this study. **Conclusions:** HP infection eradication had no clear role for PIT response. However, our sample was low, then more studies are required.

	Without HP infection (n=10)	With infection (n=16)	P
Age (mean ± SE ^o)	35.9 ± 3.81	37.2 ± 4.63	0.41
Gender (female/ male)	7/3	12/4	0.86
Lack of response to corticoids ¹	0/10	4/16	0.34
Initial platelet count (at diagnosis; mean ± SE)	17.8 ± 5.51	17.9 ± 6.28	0.49
Final platelet count (end of the follow-up; mean ± SE)	196.2 ± 22.69	185.3 ± 24.02	0.38
Significant ANA ² levels (yes/not; n(%))	1/9 (0.1)	1/15(0.06)	0.67
Number of treatment strategies ⁴ (mean ± SE)	2.62 ± 0.30	2.6 ± 0.25	0.13
Solenectomy (yes/not; n(%))	6/4 (0.6)	12/4 (0.75)	1.00
Significant APS ² antibodies levels (yes/not; n(%))	0/10 (0.1)	3/13 (0.18)	0.49
The reapeutic response			
CR ⁵	9/1 (0.9)	12/4 (0.75)	0.66
PR ⁶	1/9 (0.1)	3/13 (0.18)	1.00
Inmunosupresor dependence	1/9 (0.1)	4/12 (0.30)	0.66

A1169

PREVALENCE OF JAK2 V617F MUTATION IN NORTHEAST MEXICAN PATIENTS WITH MYELOPROLIFERATIVE NEOPLASMS. Mendoza-Ibarra SI, López-Silva LJ, Cancela-Murrieta CO, Gómez-Almaguer D, Salazar-Riojas R. Department of Hematology, Hospital Universitario, Universidad Autonoma de Nuevo Leon, Monterrey, Mexico.

Introduction: Myeloproliferative neoplasms (MPNs) are a collection of progenitor cell disorders that result in the inappropriate accumulation of myeloid cells in the blood and bone marrow. Classic Philadelphia chromosome negative MPNs include polycythemia vera, essential thrombocythemia, and primary myelofibrosis. A new mutation (V617F) affecting the JAK2 gene has been recently described as acquired in a large proportion of patients with MPNs and other myeloid disorders. This mutation changes a valine to a phenylalanine at residue 617 of JAK2. JAK2-V617F is present in about 95% of polycythemia vera patients and about 50% of essential thrombocythemia and primary myelofibrosis patients. **Objective:** The aim of this study was to determinate the prevalence of JAK2 V617F mutation in patients with MPNs. **Material and Methods:** A total of 123 blood samples were obtained from patients seen in the Hospital Universitario, U.A.N.L. during January 2008 - January 2012. DNA was extracted from ethylenediaminetetraacetic acid stabilized blood or full blood using automated DNA purification by MAXWELL® (Promega, Nacka, Sweden) according to manufacturer's instructions. Diagnoses of the patients were Polycythemia Vera, Primary Thrombocytosis, Myelofibrosis and Hypereosinophilic Syndrome mainly. Polymerase chain reaction amplification for the detection of JAK2 V617F using a commercial assay (Seplex JAK2 Genotyping Kit; Seegene, Seoul) (Figure 1). **Results:** JAK2 V617F mutation was detected in 34 of 123 (27.65%) samples with MPNs. Sixty-one (49.7%) was female and Fifty-five (44.7%) was male. The median age was 50.15, and the range age was 1 to 87 49 months. Table 1. Patient characteristics

All patients	JAK2 mutation positive	JAK2 mutation negative
Number n(%)	123	34 (27.65%) 89 (72.35%)
Female n(%)	61 (49.7%)	12 (35.3%) 49 (55.0%)
Male n(%)	55 (44.7%)	19 (55.9%) 36 (40.5%)
No date n(%)	7 (5.6%)	3 (8.8%) 4 (4.5%)
Age years (median, range)	50.15 (1-87)	49.98 (14-87) 50 (1-86)

Conclusion: JAK2 V617F mutation occurs in a high proportion of patients MPNs. In our laboratory we detected a prevalence of 27.65%.

A1183

INDUCTION OF APOPTOSIS IN LEUKEMIC CELL LINES BY EXTRACTS OF A CHAYOTE HYBRID (SECHIUM EDULE). Aguiñiga-Sánchez I*, Soto-Hernández M**, Cadena-Iñiguez J***, Ruíz-Posadas L**, Weiss-Steider B*, Tiburcio-Félix R*, Gómez-García G*, González-Ugarte A*, Santiago-Osorio E*.

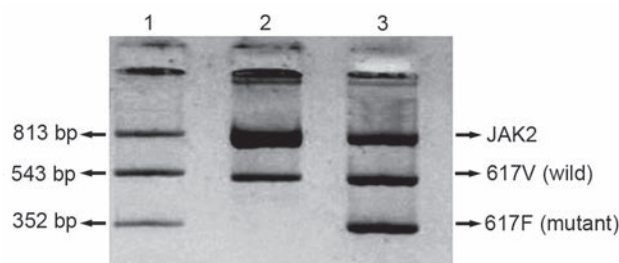


Figure 1. PCR Results. Lane 1, JAK2 Marker, Lane 2, Clinical simple wild, Lane 3, Clinical simple mutant.

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Introduction: Acute myeloid leukemia accounts for 12 % of cancer deaths. This neoplasm is more common in older adults and has a 5-year survival of less than 20 % despite novel therapeutic strategies. In the search for new treatment alternatives plants remain an important source for new drugs. Recent studies indicate that crude extracts of some plants have greater antitumor activity than purified molecules. Crude extracts from different varietal groups of *Sechium edule* (Jacq.) Sw. (commonly called chayote), have been reported to present antiproliferative activity on cell lines of solid tumors and leukemia in doses of mg.mL⁻¹. Recently, the Interdisciplinary Research Group of *Sechium edule* in México (GISeM) generated a hybrid from *Sechium edule* called and recorded as H-386-07-GISeM(TM) whose antiproliferative properties were to be determined. **Objective:** Determine antiproliferative properties of hybrid *Sechium edule*, H-386-07-GISeM(TM). **Material and Methods:** Mouse acute myeloid leukemia cell lines (P388 and J774) or normal bone marrow cells, were cultured in the presence of different concentrations of extracts H-386-07-GISeM(TM). In order to evaluate proliferative potential, cells were stained with crystal violet and the optical density was determined using a plate reader. Death by apoptosis was determined by detection of DNA fragmentation. **Results:** The data indicate a dose-dependent effect with an IC₅₀ of less than 1.3 ug.mL⁻¹ for the P388 and J774 leukemic cell lines, while greater than 2.5 ug.mL⁻¹ for normal mouse bone marrow cells. The viability of P388 was affected by IC₅₀ but not for J774 and normal cells. Interestingly apoptosis was induced only on the cell lines. **Conclusions:** These data suggest that the extract of fruits of the hybrid H-386-07- GISeMMR has a strong antiproliferative and apoptotic activity on leukemic lines but not in normal bone marrow cells. This work was supported in part by ICyTDF (PICSA 10-156), CONACyT scholarship 1 (247169).

A1187

SODIUM CASEINATE INDUCES PROLIFERATION OF BONE MARROW MONONUCLEAR CELLS, APOPTOSIS OF

WEHI-3 LEUKAEMIC CELLS AND INCREASED SURVIVAL IN A LEUKAEMIC MOUSE MODEL. Ledesma-Martínez E*, Pérez-Cordero C**, Córdova-Galaviz Y***, Sánchez-Tellez G*, Huerta-Yepez S**, Aguiñiga-Sánchez I****, Miranda-Peralta E***, Monroy-García A****, Weiss-Steider B*, Santiago-Osorio E*. *Facultad de Estudios Superiores Zaragoza UNAM, México DF, México; ** Hospital Infantil de México, Federico Gómez, México DF, México. **Hospital de Oncología, México DF, México ****IMSS, CMN SXXI, México DF, México.

Introduction: Acute myeloid leukaemia results from neoplastic transformation of haematopoietic stem cells. Though there have been recent advances in its treatment, mortality remains high. Here, we present evidence that sodium caseinate (CasNa), a salt of casein, the principal protein in milk, could possess important antileukaemic properties. **Objective:** Determine whether CasNa induce apoptosis in WEHI-3 and mononuclear bone marrow cells in vitro and induce antileukaemic effects in vivo. **Material and Methods:** CasNa was dissolved in PBS at a concentration of 100 mg/ml. WEHI-3 cells (750) or MNC/ml (1x10⁵) were cultured for 120 h with rmIL-3 and 0, 0.5, 1 or 2 mg/ml CasNa. In order to evaluate cell proliferation, cells were stained with crystal violet and the optical density was determined using a plate reader. Death by apoptosis was determined by detection of DNA fragmentation. To evaluate the antileukaemic activity of CasNa, we used two groups of 10 BALB/c mice injected intraperitoneally with 2.5x10⁴ WEHI-3/ml. After 48 h, one group was treated i.p. with 1 ml of CasNa (10 % in PBS p/v), another group only with 1 ml/PBS, this treatment was repeated every 48 h and the survival were recorded for more than 40 days. **Results:** The data showed that CasNa inhibits in vitro proliferation of mouse myelomonocytic leukaemic cell line WEHI-3, and induces the cells into apoptosis, however, under identical conditions, CasNa strongly promotes the proliferation of normal mouse mononuclear bone marrow cells. On the other hand we found that CasNa increases the survival of mice bearing WEHI-3-induced tumors, suggesting that this molecule is also capable of inhibiting the proliferation of those cells in vivo. The fact that CasNa inhibits the proliferation and induces apoptosis of leukaemic cells in vitro, but increases survival in vivo in a leukaemic mouse model, indicate that it may be useful in leukaemia therapy. **Conclusion:** The results of this study indicate that CasNa induces apoptosis of leukaemia cells without exerting a cytotoxic effect on normal haematopoietic cells, in addition promote the survival of leukaemic mice. This work was supported in part by Fondo SEP-CONACYT (104025), PAPIIT (IN225610), CONACYT scholarship 1,2,3,4(48959; 169059; 15750 and 247169).

A1188

CASNA MOBILIZES HEMATOPOIETIC PROGENITOR CELLS. Santiago-Osorio E*, Silvestre-Santana O*, Dominguez-Meledez V*, Hernández-Cristino J*, Pérez-Cordero C*1, Aguiñiga-Sánchez I*2, Gómez-García G*, Weiss-Steider B*, Moreno-Fierros L**. *Facultad de Estudios Superiores Zaragoza

UNAM, México DF, México; **Facultad de Estudios Superiores Iztacala UNAM, Estado de México, México.

Introduction: Under normal circumstances bone marrow cells reside in the marrow but they can be mobilized into the blood stream by certain cytokines like the granulocyte and macrophage growth factors (G and GM-CSF) or by molecules like AMD3100. It has already been established that sodium caseinate (CasNa), a sodium salt of casein, favours granulopoiesis through G and GM-CSF induction. **Objective:** This work was undertaken to evaluate if CasNa could mobilize mouse hematopoietic precursors into the blood stream. **Material and Methods:** A triplicate experiment containing two groups of mice were intraperitoneally injected with or without 1 mL of CasNa at a 10% dilution on PBS every 48 hours for 6 days. 24 hours after the last inoculation mononuclear blood cells were collected and the number of hematopoietic colony forming units (UFC) and precursors evaluated. **Results:** Our results showed a fivefold increase of UFC in blood as well as an increase in lin-c-kit+Sca-1+ cells. A similar result was obtained with mice treated with AMD3100 that were used as a positive control. **Conclusion:** Our results thus indicate that CasNa can mobilize hematopoietic cells in a similar way as AMD3100 does. This work was supported in part by PAPIIT (IN225610), CONACYT scholarship 1,2 (169059; 247169).

A1190

INDUCED PROLIFERATION OF BONE MARROW CELLS AND REDUCED THYMIC INDEX IN MICE TREATED WITH WEREQUE (IBERVILLEA SONORAE). Aguiñiga-Sánchez I, Pacheco-González J, Delgadillo-Gamboa J, Guzmán-Urieta E, González-Ugarte A, Hernández-Cervantes F, Ruíz-Ramos M, Weiss-Steider B, Castillejos-Cruz C, Mendoza-Nuñez V, Santiago-Osorio E. Facultad de Estudios Superiores Zaragoza UNAM, México DF, México.

Introduction: The wereque, *Ibervillea sonorae* Greene (Cucurbitaceae), is an endemic plant in northern Mexico widely used in traditional medicine as antirheumatic, antiinflammatory, analgesic, cardiotoxic and antidiabetic. This product is not legally considered a drug and is thus sold without prescription. In order to contribute to the study of the possible toxicity risks of this "traditional medicine" treatment, we evaluated its effect on mice. For this purpose mouse bone marrow cells, a group of cells known to be highly sensitive to the presence of toxic agents, blood, spleens, livers and thymuses were used to ensure phytopharmacologic beneficial effects and safety for consumers. **Objective:** Analyze the potential hypoglycemic and cytotoxic effect in mice that were intraperitoneally treated with an aqueous infusion from *Ibervillea sonorae*. **Material and Methods:** Six groups of 5 mice were treated intraperitoneally with 1 mL of an aqueous PBS solution containing 0, 36.7, 73.5 or 146 mg/mL from *Ibervillea sonorae* every 48 h for 7 days. Blood was collected to measure glucose levels, and bone marrow cells to evaluate the mitotic index, while their spleens, thymuses and

livers extracted to evaluate their size indexes. **Results:** Our results showed a decreased glucose levels in all the treated animals. The spleen and liver indexes were in general not altered, except in the spleen of mice treated with 36.7 mg/mL. Interestingly the mitotic index of bone marrow cells increased in a dose dependent way, while the thymic index decreased. These results could indicate that the consumption of *Ibervillea sonorae* has a hypoglycemic effect, and that is not toxic to bone marrow cells

but instead could even induce their proliferation. We consider that the fact that it also reduced the size of the thymus should be studied in more depth to evaluate the possible compromise of the immune response. **Conclusions:** Aqueous infusion from *Ibervillea sonorae* has a hypoglycemic effect, and is not toxic to bone marrow cells. This work was supported in part by Fondo SEP-CONACYT (104025), PAPIIT (IN225610), ICyTDF PICSA 10-156. CONACYT scholarship 1 (247169).

RED CELL PHYSIOLOGY AND DISORDERS

ORAL PRESENTATION

A1033

NATURAL KILLER ACTIVITY INDUCED BY IN VITRO IL-2 IN PATIENTS WITH BETA-THALASSEMIA MAJOR. Karakas Z*, Atasever-Arslan B**, Erdem-Kuruca S**, Erman B***. *Istanbul Faculty of Medicine, Department of Pediatrics, Istanbul University, Istanbul, Turkey; **Istanbul Faculty of Medicine, Department of Physiology, Istanbul University, Istanbul, Turkey; *** Faculty of Engineering and Natural Sciences, Biological Sciences & Bioengineering Program, Sabanci University, Istanbul, Turkey. *Present address: Faculty of Engineering and Natural Sciences, Biological Sciences & Bioengineering Program, Sabanci University, Istanbul, Turkey.

Introduction: Deficient natural killer (NK) activity is one of the various immunological abnormalities in patients with thalassemia major (TM). The function of NK cells is to recognize and kill virus infected cells and certain tumor cells. The mechanism of recognition is not exactly understood, but it involves both activating and inhibitory receptors. CD 16 and NKG2D are receptors correlated to two of the activation pathways of NK cells. These include direct cell-cell signalling via surface molecules, and indirect signaling by cytokines. The essential cytokine is interleukin (IL-2) which is regulator on NK cell functions. **Objective:** This study aims to evaluate the relationship between NK cytotoxicity, CD 16 and NKG2D receptors, and effects of in vitro IL-2 stimulation in patients with TM. **Material and Methods:** The study includes 26 patients with TM and 16 healthy subjects sex and age matched. Fourteen of the patients were splenectomized. NK cells are isolated from blood samples of all subjects by using of RosetteSep isolation kit. Before and after the IL2 incubation; CD16, NKG2D receptors levels of NK cells are examined in all subjects by flow cytometry and we analyzed cytolytic function of NK cells against to K562 cells. The cytotoxic activity of NK cells was assessed by MTT assay. Ratios of NK cells to K562 cells [Efactor:Target (E:T)] were 1:1, 10:1 and 20:1. **Results:** NK cytotoxicity of patients with TM was found to be lower at E:T ratios of 1:1 and 10:1 ($p<0.05$) than the NK cytotoxicity of healthy persons. Patients with above 18 years old had significantly lower as NK activity than under 18 years old. The sex differences and the splenectomy did not affect on NK activity

in patients. NK cytotoxicity was significantly increased by IL-2 stimulation in all patients at E:T ratio of 10:1 ($p<0.05$). As well, there were not any differences CD16 and NKG2D receptor levels of NK cells in between patients with TM and healthy persons; however, after the IL2 incubation, NKG2D receptor levels of IL2 induced NK cells significantly increased in healthy persons ($p<0.003$). Whereas NKG2D receptor levels of IL2 induced NK cells which only splenectomized TM patients have significantly increased ($p<0.005$). **Conclusions:** Our results demonstrated IL2 induced NK activity was significantly higher than unstimulated NK activity in all patients. According to these results, IL-2 may have potential therapeutic effect to improve the defective NK activity in patients with TM patients. This study was supported by Scientific Research Fund of Istanbul University.

A1165

EVALUATION OF MEAN SPHERED CORPUSCULAR VOLUME, LIKE A ROUTINE SCREENING TEST FOR PREDICTING HEREDITARY SPHEROCYTOSIS IN PEDIATRIC PATIENTS. Romero-Guzmán L, Mujica-Guzmán F, Mendoza-González F, Farfan-Becerril C, Jaloma-Avendaño R, Rodríguez-Sánchez A, Paredes-Aguilera R. National Institute of Pediatric, México DF, México

Introduction: Hereditary spherocytosis (HS) is a frequent inherited RBC disease in Caucasian populations, with a prevalence estimated between 1 in 2000 and 1 in 5000. Frequently the diagnosis of HS is based both on clinical and biological features. Nevertheless, diagnosis is not always easy to make because family history is missing in 25% cases, clinical features are not always evident and spherocytes can be observed in other cases of hemolytic anemia such as autoimmune hemolytic anemias (AIHA). It has been shown that the mean sphere corpuscular volume (MSCV), an artificial volume, is always lower than MCV in HS and also in some autoimmune hemolytic anemias (AIHA). **Objective:** Our purpose was to assess the reliability of MSCV in routine practice, and its relevance in screening for hereditary spherocytosis. **Material and Methods:** We included in the study 30 patients with different types of anemia, 30 patients with previously diagnosed Hereditary Spherocytosis by traditional method, of erythrocyte fragility, and 50 healthy children. We compared the Mean Corpuscular Volume (MCV) to the Mean Spherized Corpuscular Volume (MSCV), with the difference

obtained between the two, called delta value. Assessed during the reticulocyte count procedure under hypo-osmotic conditions; this analysis was performed on the LH 750 of Beckman Coulter. The MSCV became smaller than the MCV. **Results:** We observed that the determination of delta value had 87.8% specificity, 96.2% sensitivity, 86.7% positive predictive value, and 96.7% negative predictive value for Hereditary spherocytosis. Delta value (MCV-MSCV) > 13.71 fl is highly indicative of Hereditary Spherocytosis. The cutoff value was taken from 50 healthy children. **Conclusion:** We suggest that MSCV is as reliable parameter as routine screening test to identify Hereditary Spherocytosis. We obtained MCV-MSCV > 13.71 fl. The MSCV is a rapid method and low cost.

A1224

IRON STORE CHANGES IN A FAMILY'S OF PRESCHOOL CHILDREN IN MEXICO CITY, UNDER THE EFFECT OF HFE GENE MUTATIONS (H63D/C282Y). Baptista GHA, Rosenfeld MF, Trueba GR, Bouchan VP, Coeto BG. Instituto Nacional de Perinatología; Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. Ciudad de Mexico, Mexico.

Introduction: Iron deficiency (ID) and anemia (IDA) is influenced by economic, social and cultural determinants were the children and his family lives. A biological variable with special interest in iron metabolisms is the HFE gene mutations. The HFE mutations in very common in Caucasian population, and explains about 40% of changes in serum ferritin (SF). **Objectives:** To evaluate the association between iron store status of both parents and its

impact on their respective children, under the influence of HFE gene mutations. **Material and Method:** In a cross-sectional design, we evaluated nuclear families at the pre-school of Mexico City area. Informed consent was obtained from both parents and preceded to the taking of venous blood sample of nuclear family. We determined erythrocyte indices, using the methodology of impedance readings in triplicate. In serum samples were measured SF concentrations by ELFA methodology. We defined as cases group (IO) subjects with SF = 300 ug/L and normal iron store (NIS) with FS 20-299 ug /L and iron deficiency (ID) with FS <30 ug/L. In leukocyte DNA sample were amplified sequences specific for HFE gene mutations (H63D, C282Y, S65D). **Result:** We included 170 subjects from 44 families; 44 mothers and 43 parents and 44 first child, 33 second child and child three or four in six cases (total 83 children), with ID prevalence of 0.591, 0.047, 0.422, 0.353, and 0.833, respectively. Of the 26 women with ID, 17 children presented ID too, but without statistical association [OR 1.51 (IC 95 % 0.4 to 5.1)], even in the order of child. The IO occurred in ten cases, all male parents. The prevalence of HFE gene mutations in the all population was 16.4%, no differences in SF median values by effect of HFE mutations, neither C282Y nor H63D. However, for H63D and C282Y polymorphism showed differences between percentiles 75 for SF. **Conclusion:** The prevalence of IDA was not different from reported. No association between maternal or paternal iron stores, with their respective children. In the 3-4 son, showed a higher prevalence of IDA. The HFE gene mutations have no clinically relevant effect in our population. The difference in the prevalence of IO between father and mother seems to be related to sociocultural factors, but not related to HFE gene mutations.

POSTERS

A1005

COMPARISON OF DISCRIMINATIVE INDICES FOR IRON DEFICIENCY ANEMIA AND THALASSEMIA TRAIT IN A BRAZILIAN POPULATION. Matos JF***, Dusse LMS*, Stubbert RVB***, Ferreira MFR***, Faria JR****, Gomes KB*, Carvalho MG*. *Clinical and Toxicological Department-Faculty of Pharmacy-Federal University of Minas Gerais, Belo Horizonte, Brazil; **Federal Institute of Minas Gerais - Ouro Preto Campus, MG, Brazil; ***Laboratory of Clinical Pathology - Hospital Governador Israel Pinheiro (IPSEMG), Belo Horizonte, MG, Brazil; ****Department of Internal Medicine- Medical School - Federal University of Minas Gerais, Belo Horizonte, MG, Brazil.

Introduction. Considering that microcytic and hypochromic anemias have different pathogenesis, treatment and prognosis, it is mandatory the correct identification of them. With the purpose of discriminating iron deficiency anemia from thalassemia trait in a faster and simple way, several indices obtained from modern blood count analyzers were reported by literature. **Objective:** The purpose of this study was to evaluate the efficiency of seven in-

dices in discriminating patients with iron deficiency anemia from those with thalassemia trait, whose diagnosis has been previously confirmed by gold standard tests. **Material and Methods:** Power of discrimination of seven indices to differentiate between iron deficiency anemia and thalassemia trait such as Green and King Indice (GKI), RDW Indice (RDWI), Srivastava Indice (SRI), Mentzer Indice (MI), Shine and Lal Indice (SLI), Ehsani Indice (EI) and Sirdah Indice (SI) were evaluated. These indices were applied in 53 patients with thalassemia trait and in 289 with iron deficiency anemia confirmed by gold standard tests. Sensitivity, specificity, positive and negative predictive values, efficiency, area under ROC curve and Youden's indice were calculated. **Results:** The best diagnostic accuracy for differentiation between iron deficiency anemia and thalassemia trait was observed for GKI and RDWI indices. Conversely, Shine and Lal Indice has shown the less satisfactory performance in discriminating these two types of anemias. **Conclusions:** A comparative analysis has indicated the superiority of Green and King and RDW indices for discriminating between iron deficiency anemia and thalassemia trait.

A1012

ASYMPTOMATIC MALARIA IN ANGOLAN BLOOD DONORS: RELATIONSHIP WITH THE ABO BLOOD GROUP AND S-HEMOGLOBIN. Fernández Aguila JD *, Neto Matias A**, Rivero Jiménez RA ***. *Dr. Gustavo Aldereguía Lima University Hospital, Cienfuegos, Cuba. **Múltipil Clinic. Luanda, Angola. *** Hematology and Immunology Institute, Havana, Cuba.

Introduction: Malaria is the most disseminated infectious parasitic disease in the world. The genetic causes of patient's susceptibility to malaria parasites have been tried to be elucidated during several years, and a great polygenicity has been identified. The genetic variations of innate immune system and in the erythrocytes have been proposed as protecting factors against severe malaria. During blood donor screening, individuals with positive malaria test without symptoms of the disease can be found.

Objective: We investigated the possible relationship between asymptomatic malaria with genetic alterations of hemoglobin and frequency of ABO blood group. **Material and Methods:** A prospective study was conducted during period from January 1st to July 31st, 2010, in 1081 blood donor candidates in the "Clínica Múltipil", Luanda, Angola. Those who were identified 47 without symptoms of malaria but with a positive result for a rapid immunoassay test and a positive Giemsa stain smears for *Plasmodium falciparum*. Haemoglobin electrophoreses in agarose gel was performance. The proportion of S-Hb was compared with those of a control historic group of 591 blood donors, and the frequency of ABO blood groups with the rest of the donors in the same period. **Results:** AS-Hb genotype was found in 26 (55, 32%) of blood donor candidates, a proportion higher than in the control group ($p < 0.0001$), and the frequency of ABO blood group were similar between the compared groups. Nevertheless, when the frequency of carriers of S-Hb with asymptomatic malaria was compared in the different ABO group, in the A blood group was low (10%) while in those with B blood group 92, 3% of the candidates have the AS-Hb genotype. No explanations for this fact have been found. **Conclusions:** A better knowledge of the host gens polymorphism associated with parasite resistance to clinical malaria and/or lower parasitemia will offer new elements for future prophylactic or therapeutic interventions.

A1014

OCULAR MANIFESTATIONS IN ADULTS WITH SICKLE CELL DISEASE. Fernández-Aguila JD*, Villares-Alvarez I*, Ríos-Araujo B*, Aroche-Quintana M*, Fojaco Y*. *Dr. Gustavo Aldereguía Lima" University Hospital. Cienfuegos, Cuba.

Introduction: Sickle cell anemia is the most common hemoglobin hereditary disorder in Cuba. **Objective:** The objectives were aimed at assessing the frequency of ophthalmologic alterations related with the clinical and hematological parameters and to estimate their functional consequences. **Material and Methods:** A descriptive study was done with a transversal cut to assess

the ocular signs of sickle cell disease in 64 adults: 44 with SS genotype (HbSS), 15 with hemoglobin SC disease (HbSC), and 5 with SB thalassemia (SB thal), who were assisted at the "Dr. Gustavo Aldereguía Lima" University Hospital. The medical histories were checked to gather information and a complete ophthalmologic exam was done under basal conditions. Descriptive statistics were used for the data analysis (means comparisons and the Chi 2 test). **Results:** The comma-shaped fragment was most frequent alteration found. It was associated with the SS genotype ($p=0.002$) and with lower levels of the hemoglobin ($p<0.000$). Vessel tortuous was the most common finding gathered from the retina of all genotypes. A non-proliferate retinopathy was diagnosed in 88.6% of the patients with SS genotype, in 100% of those having HbSC and in 80% of those who presented SB thal. Five patients having HbSS (11.3%) and one having HbSC (6.6%) there were findings of both types of retinopathies. **Conclusions:** Despite the several lesions found, visual keenness was classified as a good one in 94.6% of the examined eyes.

A1017

A NEW DISCRIMINATING FORMULA BETWEEN IRON DEFICIENCY ANEMIA AND THALASSEMIA MINOR. Matos JF***, Dusse LMS*, Stubbert RVB***, Ferreira MFR***, Faria, JR****, Moreira RCN⁵; Gomes KB*; Carvalho MG*. *Departament of Clinical and Toxicological Analyses, Faculty of Pharmacy of the Federal University of Minas Gerais, Belo Horizonte, MG, Brazil; ** Federal Institute of Minas Gerais - Ouro Preto Campus, MG, Brazil; ***Laboratory of Clinical Pathology - Hospital Governador Israel Pinheiro (IPSEMG), Belo Horizonte, MG, Brazil; ****Department of Internal Medicine, Medical School of the Federal University of Minas Gerais, Belo Horizonte, MG, Brazil; ⁵CEFET, Belo Horizonte, MG, Brazil.

Introduction: Microcytic and hypochromic anemias such as iron deficiency anemia and thalassemia minors affect several people in worldwide. Therefore, differentiation of these microcytic anemias is of clinical importance. Currently, diagnosis of such anemias is defined by gold standard tests that based on iron metabolism and HbA2 dosage. However, methodologies for these tests are time consuming and expensive. Then, development of a more simple method for screening of microcytic and hypochromic anemias may be extremely useful in clinical routine labs, using data extracted from a simple blood test. **Objective:** The aim of this study was to develop a simple discriminative index for differentiating iron deficiency anemia from thalassemia minor. **Material and Methods:** To create this "New Index" hematological parameters values of 83 patients with iron deficiency anemia and 23 patients with thalassemia minor were subjected to Fisher Discriminant Analysis and also plotting the ROC curve (Receiver Operating Characteristic Curve). The "New Index" uses the parameters mean corpuscular hemoglobin concentration (MCHC) and erythrocyte counts (RBC) presenting the formula $1.91 \times \text{RBC} + 0.44 \times \text{MCHC}$ and cutoff 23,85. **Results:** If the value found for a given patient is < 23.85 indicates iron

deficiency anemia, while values = 23.85 indicate thalassemia minor. Sensitivity and specificity for iron deficiency anemia, using this cutoff were 0.94 and 0.87, respectively, while area under ROC curve was 0.895 for the "New Index". **Conclusions:** Considering these preliminary results the "New Index" has shown good performance in discriminating both anemias. Supported by CNPq and FAPEMIG, Brazil.

A1024

TECHNICAL ADVANCES IN RETICULOCYTE COUNTING BY FLOW CYTOMETRY. Viana KA*, Carvalho MG*, Dusse LMS*, Komatsuzaki F*, Avelar RS**, Antonelli L***, Olindo Assis Martins-Filho OA***. *Department Of Clinical and Toxicological Analysis, Faculty of Pharmacy, Federal University of Minas Gerais, Belo Horizonte, Brazil; **Patologia Clínica São Paulo, Belo Horizonte, Minas Gerais, Brazil; ***Centro de Pesquisas René Rachou, FIOCRUZ, Belo Horizonte, Minas Gerais, Brazil.

Introduction: Reticulocyte counts in clinical laboratories are performed by manual or automated method, presenting the first one a greater chance of errors, besides to be a very laborious technique. These limiting factors may compromise the results and contribute to the high coefficient of variation of the manual method. Advances in automated reticulocyte analysis have renewed the clinical interest and the biological relevance of this count. **Objective:** The objective of this study was to evaluate the performance of a new laboratory protocol for counting reticulocytes by flow cytometry (FC) using acridine orange (AO) as a dye. At the present study we compared the performance for three different techniques including the manual technique, the laboratory protocol using AO and a commercial kit, being the latter two techniques conducted by CF. **Material and Methods:** To reduce the error of manual counting, the technique was performed in duplicate. We analyzed 150 blood samples with reticulocyte count ranging from 0.5% to values exceeding 10% by manual counting. The results were analyzed according to recommendations of the National Committees for Clinical Laboratory Standards (NCCLS) to assess interchangeability between techniques using linear regression. **Results:** A similar and good correlation was found between the manual technique and the protocol using AO ($R=0,953$), between manual technique and a commercial kit ($R=0,960$) and between the protocol using AO and a commercial kit technique ($R=0,961$). Coefficients of variation (CV) for both manual technique and protocol using AO were evaluated, and a lower CV was found for the automated technique for low, medium and high reticulocyte counts (16.76 x 26.29; 13,40 x18, 98 and 6.23 x9, 61, respectively). Since the use of commercial kits has a higher cost compared to the protocol using AO and considering that the diluted dye AO can be prepared in clinical labs from a concentrated solution, use of this protocol becomes economically suitable to small and large laboratories working with flow cytometric assays. The protocol using AO is easier to perform and is similar to the commercial kit. **Conclusions:** These results

suggest that the proposed protocol can be a good alternative to the manual technique of reticulocyte count, since it presents the best cost-effectiveness and increases the accuracy of the results. SUPPORT: CNPq / CAPES / FAPEMIG –Brazil

A1031

HEMOLYTIC ANEMIA INDUCED BY CYTOMEGALOVIRUS IN INFANCY. Lee KS*, Shim YJ*, Ahn HS**, Yoon HJ***. *Department of Pediatrics, Kyungpook National University School of Medicine, Daegu, Republic of Korea; **Department of Pediatrics, Seoul National University College of Medicine, Seoul, Republic of Korea; ***Department of Hematology-Oncology, Kyung Hee University School of Medicine, Seoul, Republic of Korea.

Introduction: Cytomegalovirus (CMV) is known to cause several hematologic disorders including thrombocytopenia, atypical lymphocytosis, and infrequently anemia. We experienced seven cases of CMV induced hemolytic anemia (CMV-HA) in infancy. **Objective:** Thus we demonstrated the characteristics of age, laboratory tests, and treatment outcome.

Material and Methods: We reviewed the medical records of CMV-HA infants from January 2003 to December 2011 in Kyungpook National University Hospital. CMV infection was confirmed by polymerase chain reaction, early antigen, virus isolation, and Immunoglobulin M. We investigated their initial blood tests and follow-up outcomes after treatment.

Results: There were seven children (male: female = 2:5) who were diagnosed as CMV-HA. The mean age at diagnosis was 88 days (1 day - 10 months). The mean hemoglobin was 9.3 g/dL (4.7 - 15.6g/dL), reticulocyte was 5.9% (3.1 - 13.4%), corrected reticulocyte was 3.5% (2.5 - 4.7%), and reticulocyte index was 2.3 (1.1 - 4.7). Four of them also showed thrombocytopenia (mean 42,000/mm³, ranging from 6,000 to 98,000/mm³). One infant was recovered of itself without any treatment. Another one was cured after being administered intravenous immunoglobulin because of severe thrombocytopenia before verification of CMV. We used intravenous ganciclovir to treat five children, but one of them demonstrated recurrence of HA despite of additional two doses of ganciclovir and supplementary CMV immunoglobulin. She was completely recovered after splenectomy. The other four infants were cured after being treated with ganciclovir. **Conclusions:** We report seven CMV-HA infants with various clinical manifestations and different treatment responses. It is important to examine CMV for those who had HA with unknown etiology.

A1040

HEMOGLOBIN, HEMATOCRIT, RED BLOOD CELLS, AND WHITE BLOOD CELLS IN MEN AND WOMEN IN THE SIBERIAN POPULATION. Pello E, Maljutina S, Simonova G, Nikitin Y. Institute of Internal Medicine, Siberian Branch of the Russian Academy of Medical Sciences, Novosibirsk, Russia.

No	Age (days)	Sex	Hb (g/dL)	Hot (%)	Reti (%)	C-reti (%)	RI	WBC (μ L)	PIT (μ L)	CMV IgM	CMV early Ag	CMV PCR Blood/Urine	CMV culture	Treatment
1	1	F	15.6	47.2	5.6	4.7	4.7	9,700	98,000	+	-	+/+	-	Ganci
2	41	M	8.9	24.3	6.0	3.4	1.8	9,360	6,000	-	+	+/+	+	Ganci
3	43	F	4.7	14.2	13.4	4.7	3.7	18,690	455,000	+	-	+/+	+	Ganci + CMV IVIG + Splenectomy
4	52	F	6.5	19.1	4.5	2.5	1.1	9,430	387,000	+	-	+/+	-	Ganci
5	61	F	9.4	28.5	4.2	3.4	1.6	11,920	588,000	-	-	+/+	+	ND
6	115	M	8.5	25.6	4.4	3.2	1.6	13,190	55,000	+	+	-/-	-	Ganci
7	300	F	11.6	35.1	3.1	2.7	1.8	10,240	9,000	+	ND	ND	ND	IVIG

Hb, hemoglobin; Hot, hematocrit; Reti, reticulocyte; C-reti, corrected reticulocyte; RI, reticulocyte index; PIT, platelet; CMV, cyto-megalovirus; IgM, immunoglobulin M; Ag, antigen; PCR, polymerase chain reaction; Ganci, ganciclovir; IVIG, immunoglobulin G; ND, not done

Introduction: Currently, on the one hand, much attention is attracted to the prevalence, genetic control, and structure-function relations of anemia and anemia associated diseases. On the other hand, the relatively rare clinical entities of myeloproliferative and myelodysplastic syndromes with high mortality rate are remained as the significant alerts worldwide. Moreover, researchers continue to study the difficulty in diagnosing of anemia of chronic disease and of chronic inflammation. The hematological parameters implement a nontrivial role in pathogenesis of the development and progression of endothelial dysfunction, cardiovascular disease, and heart failure. **Objective:** From pathophysiological point of view, the recently launched necessary health initiatives have underscored the critical importance of renal, thyroid, and androgen functions and erythropoiesis wellbeing in men and women. The estimation of marrow stem cell and erythropoietic reserve changes in the integration with enhancement of age commitments has a tremendous value attuned to disorders. Our investigation was devoted to a pivotal purpose to assess the age- and gender-dependent discrepancies of blood characteristics. **Material and Methods:** We measured (EPOGH study) venous hemoglobin, hematocrit, red (RBCs) and white blood cells (WBCs). **Results:** Conceptually, the ascertainment of the deplorable hematological instabilities demands a major challenge to the appropriate serious risk factors and adjuvant mix of convergent risk factors for creation of the general presumptions about pathways of illnesses formation. Stating alternatively, we revealed that men had higher hemoglobin (16.0 ± 1.5 vs 13.9 ± 1.4 , $P < 0.001$), hematocrit (45.1 ± 3.0 vs 40.3 ± 3.2 , $P < 0.001$), RBCs (4.9 ± 0.6 vs 4.2 ± 0.4 , $P < 0.001$), and WBCs (5.6 ± 1.2 vs 5.0 ± 1.0 , $P < 0.001$) than women. Aspiring to test theory, we sought to identify the differences of hematological parameters in patients below 40 years, at and above 40 years, but vainly. In reality, the additive effects of age and gender combination showed that in men were increased hemoglobin (15.8 ± 1.3 vs 13.7 ± 1.2 , $P < 0.001$), hematocrit (44.8 ± 2.8 vs 39.9 ± 2.6 , $P < 0.001$), RBCs (4.9 ± 0.6 vs 4.2 ± 0.4 , $P < 0.001$), and WBCs (5.5 ± 1.1 vs 5.1 ± 1.1 ,

$P = 0.016$) than in women in group below 40 years as well as in group at and above 40 years concerning hemoglobin (16.1 ± 1.6 vs 14.1 ± 1.6 , $P < 0.001$), hematocrit (45.5 ± 3.1 vs 40.8 ± 3.6 , $P < 0.001$), RBCs (4.9 ± 0.6 vs 4.3 ± 0.5 , $P < 0.001$), and WBCs (5.7 ± 1.4 vs 4.8 ± 1.0 , $P < 0.001$). **Conclusions:** Interestingly, it has been noted that women probably have the more potent risk factors for hematological variations, defining the therapeutic strategies in primary care. In our opinion, such findings further support the intentions imbued with a robust sense to explore the underlying influences of blood profile on the development of cardiovascular disease and prediction of fatal outcomes.

A1050

SICKLE CELL ANEMIA: ADVANCES IN THE PROGNOSIS AND TREATMENT IN ADULT PATIENTS IN CUBA. Espinosa-Estrada EE, Hernández-Padrón C, Machín-García S, Ávila-Cabrera O, Díaz-Barroso LM, Almagro-Vázquez D, Castillo-González D, Macías-Abraham C, Menéndez-Veitía A, Villaescusa-Blanco R, Bencomo-Hernández A, Abreu-Cruz J, Wade-Mateo M, Estrada-del Cueto M, Svarch E, Espinosa-Martínez EE. Instituto de Hematología e Inmunología; Instituto de Cardiología y Cirugía Cardiovascular, La Habana, Cuba.

Introduction: In 1972, the Institute of Hematology and Immunology developed the first study in patients diagnosed with sickle cell anemia in Cuba. **Objective:** Research in morbidity/lethality, biochemical and immunological markers, therapeutic modalities and organ damage. **Material and Methods:** Results obtained between 1997 and 2011. **Results:** 1) Morbidity/lethality: 397 patients evaluated; primary cause of hospitalization: infection, pain vasocclusive crises (VOC), acute chest syndrome (ACS). Causes of death ($n = 38$): hepatic crises (23,5 %), vascular accident (VA) (17,6 %), massive arrest and ACS (9,8 %). Pregnancies: 134 (7 prenatal, 3 newborn, and 4 maternal death). Overall mean survival: 53 years. 2) Anti-band 3 antibodies and pain VOC (25 patients: 8 with VOC; 17 in steady state):

diminished levels of natural antibodies anti-band 3 in VOC (p < 0.0005). 3) Adhesion molecules expression: 20 in steady state; 18 with VOC. Diminished CD11a+/CD18+ and CD49d+/CD29+ (p < 0.05); increased CD 54+ in both groups (p < 0.05). Between groups, significant increase in CD18+ and NK CD57+ and diminished CD 29+ and total CD3+ lymphocytes (p < 0.05) in VOC. 4) Frequency of pulmonary hypertension (PHT) (n = 104): 28,8 %. Significant negative correlation between PHT and hemoglobin (Hb and pulse oximetry (PO) (p = 0,021); significant increase in leukocytes (p = 0,024), lactate dehydrogenase (LDH) (p = 0,003), total and indirect bilirubin (TB p = 0,008; IB p = 0,023), creatin kinase-MB (p = 0,033), creatinine (p = 0,001) and ACS (p = 0,05). 5) Correlation between LDH and biochemical and clinical parameters (n = 130). Biochemical: negative correlation with Hb, fetal hemoglobin (p = 0,000) and PO (p = 0,005); positive correlation with reticulocytes (p = 0,045), aspartate and alanine aminotransferasa, TB, IB, creatin kinase-MB (p = 0,000), alkaline phosphatase (p = 0,017) and creatinine (p = 0,008). Positive correlation with PHT (p = 0,008) and leg ulcers (p = 0,031). 6) Hydroxyurea treatment: patients (n = 29) with VOC, ACS and VA; significant increase of fetal hemoglobin (p = 0,005) and decrease in VOC, VA, ACS; transfusion requirement, infections and hospitalization (p = 0,001). 7) Virological studies (n = 105) (human immunodeficiency virus, hepatitis B serum antigen, antibodies against human C virus; AbHCV): 16,2 % patients with AbHCV. 8) Albuminuria (n = 94): 86 positive cases (90,5 %); 8 with macroalbuminuria (9,3 %). **Conclusions:** At present, the research is focused in the study of hepatic, cardio-pulmonary and renal complications.

A1051

SEVERE COLD IDIOPATHIC AGGLUTININ SYNDROME TREATED WITH RITUXIMAB. Espinosa-Estrada EE, Ramón-Rodríguez LG, Hernández-Padrón C, Ávila-Cabrera O, Sarduy-Sáez S, Bencomo-Hernández A. Instituto de Hematología e Inmunología. La Habana, Cuba.

Introduction: The cold antibody autoimmune hemolytic anemias (AIHAs) are primarily comprised of cold agglutinin syndrome (CAS) and paroxysmal cold hemoglobinuria (PCH) but, in addition, there are unusual instances in which patients satisfy the serologic criteria of both warm antibody AIHA and CAS ("mixed AIHA"). CAS characteristically occurs in middle-aged or elderly persons, often with signs and symptoms exacerbated by cold. The treatment with rituximab is the only well-documented effective therapy. **Objective:** Clinical and hematological response to rituximab treatment in a patient with severe cold idiopathic agglutinin syndrome. **Material and Methods:** The authors present a case of a 21 year-old white female with a history of immunological thrombocytopenic purpura (ITP) in remission for five years. **Results:** In November 2011, she was exposed to low temperatures and presented livedo reticularis, pallor and shortness of breath. The laboratory findings consisted of Hb level 5,6 g/dL, reticulocytosis, hemolytic pattern in the blood smear and

a positive direct Coombs test against C3; autoagglutination 4x corrected only with reductor agents (dithiothreitol; DTT), the red blood cell eluate reaction was negative with a crioagglutinine title of 1:2 000. The thermic range title was of 1.128 at 37oC. There was no anti-I specificity and no other secondary causes of CAS were found. At that time the diagnosis was severe idiopathic CAS. The patient started on rituximab at 375mg/m² on days 1, 8, 15 and 22. **Conclusions:** After treatment the hemoglobin raised up to 10 g/dL and the patient achieved a complete clinical response. There are no previous reports of CAS treated with rituximab in our country.

A1073

PAROXYSMAL NOCTURNAL HEMOGLUBINURIA IS A VERY RARE DISEASE AMONG WEST CENTRAL MEXICO POPULATION. Campos-Cabrera G*, Campos-Cabrera S*, Campos-Cabrera V**, Mora-Torres M***, Gomez-Guijosa MA***, Rivera-Trujillo A****, Hernandez-Rodriguez S****, Pita-Ramirez L⁵, Campos-Villagomez JL*. *Departamento de Hematología de Laboratorios Fatima de Michoacan, Morelia, Mexico; **Departamento de Patología Clínica de Laboratorios Fatima de Michoacan, Morelia, Mexico; ***Servicio de Hematología del H. G. R. No.1 IMSS, Morelia, Mexico; ****Servicio de Hematología del Hospital Civil "Dr. Miguel Silva", Morelia, Mexico; ⁵Servicio de Hematología del Hospital General "Vasco de Quiroga" ISSSTE, Morelia, Mexico.

Introduction: Paroxysmal nocturnal hemoglobinuria (PNH) is a rare hematopoietic stem cell disorder characterized by a somatic mutation in the PIGA gene, leading to a deficiency of proteins linked to the cell membrane via glycosphosphatidylinositol (GPI) anchors. The disease has an estimated incidence of only 1.3 new cases per one million individuals per year in the United States. PNH has three distinctive clinical features that vary greatly from patient to patient and during the course of the disease; first, there is complement mediated and predominantly intravascular hemolysis; second, there is a characteristic thrombotic tendency that can be life threatening and occurs not only in the extremities but also in unusual anatomical locations, such as hepatic portal (Budd-Chiari Syndrome), splenic, or mesenteric veins, third, there is underlying bone marrow failure, which occurs to some degree in all patients, and in its most extreme form, presents as immune mediated severe aplastic anemia. **Objective:** To determinate the prevalence of PNH immunophenotype in our regional population. **Material and Methods:** Blood samples referred to Laboratorios Fatima de Michoacan for flow cytometry in suspected individuals that could have PNH. The protocol is based on the Guidelines for the Diagnosis and Monitoring of Paroxysmal Nocturnal Hemoglobinuria and Related Disorders by Flow Cytometry (Cytometry Part B (Clinical Cytometry) 2010;78B:211-230). **Results:** Fifteen cases were studied; RBCs were tested with antibodies against CD55 or CD59 and FLAER (Aeromonas hydrophila aerolysin). Granulocytes were tested with antibodies against CD16 and CD24. Monocytes tested

with antibody against CD14. With this antibodies panel I can be distinguished between routine analysis (defined as identifying an abnormal population of 1% or more) and high-sensitivity analysis (in which as few as 0.01% PNH cells are detected). Only one patient had abnormal PNH populations in this selected group that had clinical indication for the test: erythrocytes CD59 normal cells 52.27 %, PNH II cells 46.72 %, PNH III cells 0.72 %, FLAER normal cells 52.25 %, FLAER PNH II cells 46.75 %, FLAER PNH III cells 1.0 %; monocytes CD 14 82.5 %; neutrophils CD 16 76.7 % and CD 24 76.1 %. **Conclusions:** PNH is a very rare disease. Its incidence is not well established in our population. The use of the guidelines above mentioned (and cited) should enable laboratories interested in beginning PNH testing to establish a valid procedure and allow experienced laboratories to improve their techniques.

A1089

PARVOVIRUS B19 INFECTION IN PATIENTS WITH HEMATOLOGIC DISEASES IN YUCATAN, MEXICO. González-Martínez P*, Valadez-González N*, Alonso-Salomón G*, Quintal-Ortiz I*, Rosado-Paredes P*, Moguel-Rodríguez W*, Vera-Gamboa L*, Pavía-Ruz N*, García-Escalante G*, Chik-Ku D*, Tzab-Chan A*, Gamboa-Maldonado I*, Góngora-Biachi R**. *Centers Regional Research Dr. Hideyo Noguchi-UADY, Mérida, Yucatán, México; **Medical Center of the Americas, Mérida, Yucatán, México.

Introduction: The clinical spectrum of parvovirus B-19 (PVB19) infection in humans is large and the severity depends on the host immune and hematological status. In Yucatan there are no clinical reports of PVB19 infection, rather than in isolated anecdotal cases without confirmatory evidence. Therefore there is no data on the pathogenic relationship between PVB19 and various blood disorders, which has been successfully documented elsewhere in the world. As a result studies to identify this possible causal connection in our environment are justified, through earlier diagnosis of PVB19 infection. **Objective:** To detect serological and/or molecular evidence of acute or recent infection by Parvovirus B-19 in patients of Yucatan with immune purpura (vascular or thrombocytopenic) and other blood diseases. **Material and Methods:** Prior written consent of the patient or guardian 34 patients with blood disorders were included, mainly vascular and thrombocytopenic purpura and 11 patients with HIV/AIDS. IgG and IgM were determined by ELISA (DRG Diagnostics) and the viral DNA presence by PCR real-time PCR (Q-PCR) was investigated. **Results:** 8 out of 21 immune thrombocytopenic purpura patients and 2 lymphoma patients just had IgG+. 6 out of 8 (75%) patients with vascular purpura had IgG+, and 2 (25%) of them were IgM+. Two paroxysmal nocturnal hemoglobinuria patients and one chronic myeloid leukemia patient were negative. 4 out of 11 HIV/AIDS (36%) patients just had IgG+. In none of the 45 patients was the DNA viral by Q-PCR detected. **Conclusion:** The detection of IgM in two cases of vascular purpura is associated with acute infection with PVB19 and supports the role of this

virus in the pathogenesis of these diseases. As described, the production of antibodies mainly IgM specifically is correlative with the disappearance of the virus in the blood, which explains the inability to detect PVB19 by Q-PCR.

A1194

PREVALENCE OF ANEMIA IN AN ELDERLY POPULATION FROM THE NORTHEAST OF MEXICO. Pequeño-Luévano M*, Salazar-Riojas R*, Valdes-Galván M*, Flores-Vázquez M**, Salinas-Martínez R*, Gómez-Almaguer D*. *Departamento de Hematología del Hospital Universitario "Dr. José Eleuterio González". **Departamento de Geriatria del Hospital Universitario "Dr. José Eleuterio González".

Introduction: Anemia is a common problem in the elderly, with a higher prevalence observed in hospitalized or institutionalized individuals. Using the World Health Organization (WHO) definition of anemia (< 13g Hb/dL for men and < 12 g Hb/dL for women), the Third National Health and Nutrition Examination Study (NHANES III) data, stated that 11.0% of men and 10.2% of women from the people aged 65 and older in the United States were anemic. This counts as many as 3,000,000 persons of this population. This is a relevant problem because anemia has been associated with a negative outcome as it seems to play a role in the excess of morbidity and mortality in the elderly. Because of this, the adequate diagnosis and treatment is important. **Objective:** We aimed to establish the prevalence of anemia in apparently healthy elderly subjects living in nursing homes, since in Mexico there are no reports about these data. **Material and Methods:** We analyzed a group of 265 who lived in several nursing homes of metropolitan area of Monterrey, Nuevo Leon, Mexico and who were willing to participate in this research. Clinical and anthropometric evaluations, as well as dietary survey were performed. Complete blood count was measured. We excluded those with known hematologic diseases. **Results:** Prevalence of anemia was 32% for women and 46% for men. Most of these cases corresponded to normocytic normochromic anemia. From the anemic female population, 5% aged between 60 and 75 years old, and 95% aged 76 and over. On the other hand, from the anemic male population, 19% were between 60 and 75 years old, while 81% aged 76 and over. (Fig). **Conclusions:** Anemia is more prevalent in the 76 year-old and over elderly population. In our analysis, once we compared both genders, we found that male population between 60 and 75 years old is more susceptible to present normocytic normochromic anemia than the female population of the same age. Further research about the possible conditions that lead to these results needs to be made.

A1201

ASSOCIATION BETWEEN IRON OVERLOAD AND PAROXYSMAL NOCTURNAL HEMOGLOBINURIA. Neme-Yunes Y*, Guerrero E**, Pérez-Zepeda MU***, López-Karpovitch J****. *Centro Médico ABC, México D.F., México, ** Hospital Angeles, México D.F., México *** Instituto de Geriatria, México

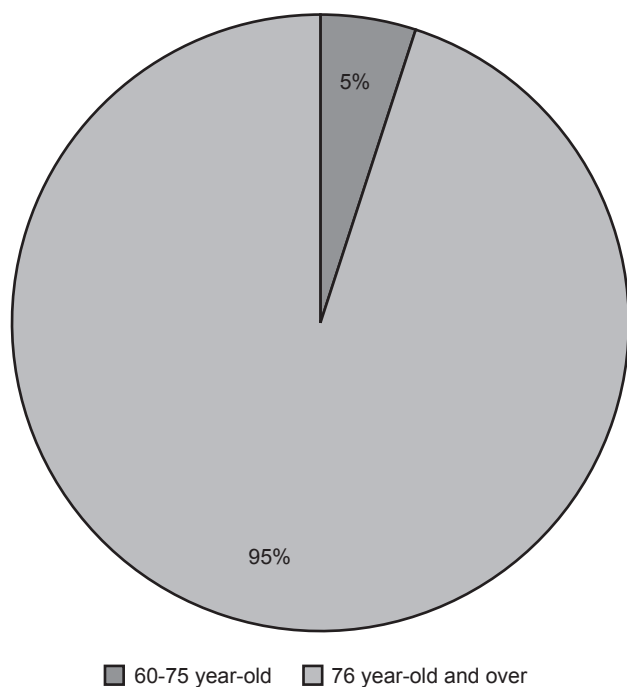


Figure. Anemia prevalence according to age in female population

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Introduction: Paroxysmal Nocturnal Hemoglobinuria (PNH) is an acquired clonal bone marrow disorder produced by a somatic mutation of the PIG-A gene which results in a poor expression of both glucosyl-phosphatidylinositol (GPI) and of the proteins anchored to GPI on the surface of hematopoietic cells. Despite the transfusion requirements associated with PNH, iron overload (IO) appears to be a rare condition in this disease (probably because of hemoglobinuria and hemosiderinuria), its frequency is unknown and there are only a few case reports that describe not systemic IO but renal hemosiderosis in PNH. **Objective:** The aim of this study was to determine the frequency of iron overload in a cohort of patients with PNH; one of the largest in Latin America to our knowledge. **Material and Methods:** All patients at our department are studied thoroughly and followed up. As a way to indirectly determine iron overload three biochemical parameters were used: serum ferritin, serum iron levels and transferrin saturation. The number of red blood cells transfused packages was recorded. Ultrasensitive reactive C protein (usRCP) was also obtained to control for inflammatory response. Descriptive statistics were done, and a multiple linear regression model to test the independent association between ferritin and number of red blood cells transfused packages and usRCP. **Results:** We studied 30 patients with a diagnosis of PNH, 46.7% of which were women (n=14). The median age was

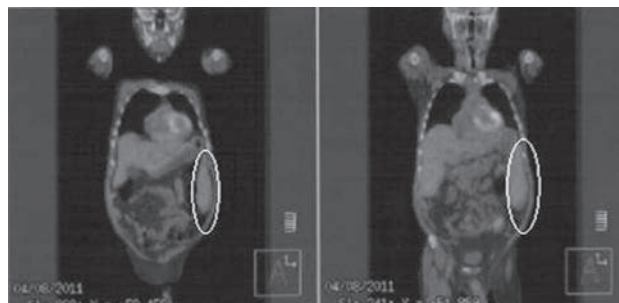
37. A significant correlation was found between ferritin and the number of RBC transfused packages (0.488), independent from the usPCR levels. In the multivariate analysis the number of RBC packages transfused was the main association with ferritin, with a B coefficient of 28.47 ($p < 0.0001$). **Conclusion:** Although the study sample is small, the association between transfused red blood cell packages and ferritin appears to be strong. These findings should alert about the possibility of iron overload and its complications in PNH.

A1203

POLYCYTHEMIA RUBRA VERA (PRV): A CASE REPORT TREATED WITH RUXOLITINIB. Ovilla MR, Guerra UN, Terrazas MR, Herrero MR, Amador AD, Vazquez A, Calles PD, Angeles LA, Buganza TE, Torres FJ, Barrera CC, Madinaveitia TJ, Enríquez OC. Hospital Angeles Lomas

Introduction: The PRV is one of the myeloproliferative disorders which its physiopathological way is the JAK2 gene mutation of the in the erythroid precursors, that determines “gain of function” of the erythropoietin receptor, giving it hyperactivity and function independence, conducting an uncontrolled upregulated eritropoiesis. Ruxolitinib is a selective JAK1/JAK 2 inhibitor mainly developed for mild and moderate risk myelofibrosis, decreasing constitutional syndrome symptoms and splenomegaly.

Objective: Determine treatment outcomes with Ruxolitinib treatment in patient with JACK 2+ mutation PRV, because of its pathway similarity. **Material and Methods:** Case report. A 61yo male, no medical history. He started at January, 2002, with reddishness, widespread itching, migraine, and nocturnal sweats; a complete blood count showed Hb 20.2 g/dL, Hct 60.8 %, WBC $9.8 \times 10^3/uL$, Plt $417 \times 10^3/uL$, plasmatic iron 55. Bone marrow aspiration showed hypercellularity +++, and bone marrow biopsy with erithroid hyperplasia +++. Abdominal US showed splenomegaly (17 x 8 cm). Initial management was erithroferesis in August, 2002, hydroxyurea 500 mg qid, (max dosage 1500 mg qid) until August, 2011. PET SCAN showed splenomegaly (23.7 cm). We documented JAK2 V617F gene mutation, initial treatment with Ruxolitinib 20 mg bid started at September, 2011, with baseline values Hb 12.3, WBC 15.3, Plt 467. **Results:** Laboratories after Ruxolitinib: Hb 12.3/ WBC 9.8/ Plt 376, 11.9/8.8/423, 10.6/6.2/456 and 10.4/7.4/675 in October, November and December, 2011 and January, 2012 respectively.



Abdominal CT showed 17 x 10.2 x 7.8 cm spleen. In January, 2012. He referred itching and nocturnal sweat remission. **Conclusion:** Ruxolitinib has shown remission in constitutional syndrome symptoms, nevertheless splenomegaly was mildly reduced, as it is used as myelofibrosis treatment. Haemoglobin decrease was seen because of Ruxolitinib pharmacokinetics, and was a desired collateral effect to achieve normal hemoglobin level. At this time no thrombocytopenia has been reported but higher platelet count.

A1221

PRECLINICAL EXPRESSION OF IRON OVERLOADS (FE) AND HFE POLYMORPHISMS IN MALE ADULTS. CASE-CONTROL DESIGN. Baptista GH, Rosenfeld MF, GR Trueba, Santamaria HC, Martinez RC, Mendez SN. Instituto Nacional de Perinatología. Fundación clínica Médica Sur Ciudad de Mexico D.F.

Introduction: Iron overload (IO) has drawn attention for its association with various conditions such as diabetes mellitus, liver cirrhosis, cardiomyopathy, cancer, reproductive failure or vascular damage. The international prevalence of IO varies from 9.3-22.5%, while in Mexico, our group has determined a prevalence up to 29% of the healthy population studied. **Objectives:** To establish the association size of HFE mutations (C282Y, H63D), iron overload (IO) and lifestyle in adult males. **Material and Methods:** In a Case-control design, we selected consecutively adult males 18-64 years of age, accepted as blood donors, living in Mexico City. We defined as cases group (IO) subjects with serum ferritin (SF) = 300 ug / L and normal iron store (NIS) with FS 20-299 ug /L and iron deficiency (ID) with FS <30 ug/L. The FS was determined in duplicate by ELFA method (Minividas Ferritin), in genomic DNA sample, we searched for C282Y and H63D mutations, evaluating the interaction with other factors involved in intestinal iron absorption (gastrointestinal bleeding due to illness or heme Fe drug intake in the diet, supplementation with Fe medicinal, alcohol intake abuse). **Results:** We studied a population base of 1849 subjects, 630 adults selected, identifying 254 with IO, 308 adults with NIS and 68 cases ID. The C282Y prevalence (all heterozygotes) was 1.1, 1.9 and 0%, respectively. In bivariate analysis, IO was associated with H63D mutation (OR 1.73, 95% CI 1.14-2.63), as well as alcohol intake abuse (OR 3.7, CI 95 2.2-6.1) and heavy use red meat (OR 4.4, 95% CI 2.2-8.6); but not associated with HFE C282Y mutation, oral Fe supplementation, intestinal bleeding history or drug exposure. In multivariate analysis, age and mutations show statistically significant influence (p 0.0009 and 0.0.11) iron store. **Conclusion:** The IO was associated with HFE H63D mutation, but not with HFE C282Y in healthy adult male Mexican mestizo origin. However, there was more interaction with age, frequent alcohol intake and high consumption of heme Fe in food.

A1222

UGT1A1 POLYMORPHISMS IN FAMILIES AND UNRELATED SUBJECTS UNDER STUDY CONGENITAL HEMOLYTIC DISEASE. Bouchan VP, Coeto BG, Zamorano JA, Baptista

GHA, Rosenfeld MF, GR Trueba. Granados ZM. Hematología Perinatal. Instituto Nacional de Perinatología.

Introduction: UGT1A1 gene variants are associated with partial deficiency of glucuronil transferase, an enzyme responsible for bilirubin conjugating activity in the liver. This condition is known as Gilbert's syndrome (GS). GS prevalence varies according to each population group and variable impact described further in patients with congenital hemolytic defects. **Objectives:** To establish the occurrence of UGT1A1 gene mutations in two groups of subjects: patients and their families and unrelated patients in the study of congenital hemolytic anemia and the presence of HFE mutations, HbS and beta-thalassemia (B-Thal). **Material and Methods:** In a cohort design, we included patients and their families under study congenital hemolytic anemia from abnormal neonatal screening (G6PD deficiency, hemoglobin S and B-Thal) and unrelated subjects studied for the same reason. In genomic DNA samples were evaluated UGT1A1 gene polymorphisms: TA [5'-cctactatataatataatgcaaaaacc] and CAT [5'-ccaagttatgatgtttgactgtg] additionally identified HFE mutations (H63D, S65D, C282Y), B-Thal and HbS (CD6-A, IVS1-110, -28, and Cd-41/42 Cd-39), all using the methodology of real-time PCR hybridization probes format. **Results:** We included 88 subjects, 20 families with 55 subjects and the remaining 33 cases of unrelated patients. The sample prevalence of variant UGT1A1 TA was 24 cases (27.2%), with the proportion of mutated 0309 and 0212, for group I and II, respectively. The occurrence of mutations for G6PD, HFE, B-Thal and HbS was at 0.080, 0.159, 0 and 0.045, respectively. There was no association between UGT1A1 mutation compared with G6PD (OR 1.07, 95% CI 0.19-5.93) or HFE (OR 2.33, 95% CI 0.71-7.62). **Conclusion:** The UGT1A1 gene mutations studied are not associated with the presence of G6PD polymorphisms, HFE, Hb S or B-Thal. These results do not support include the study of these polymorphisms in patients Gilbert neonatal screening for abnormal glucose deficiency.

A1223

CLINICAL COURSE OF NEWBORN INFANT WITH GLUCOSE-6-PHOSPHATE DEHYDROGENASE DEFICIENCY (G6PD). Baptista GHA. Trueba GR, Coeto BG, Rosenfeld MF, Zamorano JCA, Bouchan VP. Granados ZM. Laboratorio de Hematología Perinatal, Instituto Nacional de Perinatología. Estudiante de Posgrado en Ciencias Químico-Biológicas de la Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional. Ciudad de Mexico, Mexico.

Introduction: G6PD deficiency is an X-linked enzymopathy clinically expressed in a male hemizygotas and female homozygotas newborns, and a small proportion of female heterozygotas manifest the disease; the association of this deficiency with neonatal jaundice and hiperbilirrubinemia (HB) depends on factors such as type of mutation and the level of enzyme activity in erythrocytes; exogenous factors such as gestational age, type of feeding (exclusive breastfeeding), infections (ompha-

Group	Gilbert	G6PD		HFE		Hb S	B-Thal	
		WT (81/0.920)	Mutated (7/0.080)	WT (74/0.841)	Mutated (14/0.159)	WT (88/1.0)	WT (84/0.955)	Mutated (4/0.045)
I(55)	WT (38/0.690)	34 (0.850)	4 (0.105)	34 (0.835)	4 (0.105)	35 (1.0)	35 (0.921)	3 (0.079)
	Mutate (17/0.309)	15 (0.882)	2 (0.118)	13 (0.765)	4 (0.235)	17 (1.0)	17 (1.0)	
II(33)	WT (26/0.787)	25 (0.962)	1 (0.038)	22 (0.846)	4 (0.154)	26 (1.0)	25 (0.962)	1 (0.038)
	Mutate (7/0.212)	7 (1.0)		5 (0.714)	2 (0.286)	7 (1.0)	7 (1.0)	

litis, sepsis), and others, increase the severity of jaundice and hiperbilirrubinemia neonatal (HBN). **Objectives:** Describe the clinical course of newborn infant (NB) with positive neonatal screening for G6PD deficiency and theirs molecular variants. **Material and methods:** Using cohort design, we concentrated relevant clinical information of neonates with G6PD deficiency detected by neonatal screening and confirmed by real time-PCR (G202A, A376G, C563T, and T968C). Clinical follow-up were integrated for evaluation in the first year of life. **Results:** We included twenty-six infants with G6PD deficiency, twenty-four male and two female NB. The mean gestational age was 37.5 weeks, Birth Weight in grams was 3305, first minute Apgar was seven and 5 minutes was eight. The average hospitality stays in days was 10. Fourteen NB (54%) remained in nurse ring and others eleven (42.%) NB in the intermediate cares units.

One (4%) NB stays in the intensive care unit for prematurity of 29 SDG. In 16 cases (61.5%) developed jaundice and only 9 cases (56.3%) received phototherapy, with 1.6 days of duration. The serum bilirubin value was 11.3 mg/dL. **Conclusions:** The confirmed cases correspond to functional class III of the WHO, with moderate hemolytic deficiency. More than half-developed jaundice and the third part of them required phototherapy. The association of G6PD deficiency had jaundice and hiperbilirrubinemia with other hematologic morbidity was not common, as Gilbert syndrome, ABO group incompatibility. Is required to expand the universe of study screening, to detect weak cases and establish their clinical course and possible partnerships with other morbidities. In 5 cases without molecular identification of G6PD deficiency, only a case was associated with other risk factor as prematurity, very low birth weight and enteral feeding delayed.

LEUKOCYTE, INFLAMMATION, IMMUNOLOGY

POSTERS

A1064

CONGENITAL NEUTROPENIA ASSOCIATED TO STRABISMUS AND NYSTAGMUS: NEW SYNDROME LINKED TO THE X CHROMOSOME. Ron-Guerrero CS. Centro Estatal de Cancerología de Nayarit de los SSN.

Introduction: Congenital neutropenia (CN) is a primary immunodeficiency whose mode of inheritance is heterogeneous, affected genes can be located in the autosomes or in the gonosomes, therefore its prevalence throughout generations covers the entire spectrum of Mendelian inheritances as well as of chromosome X linked inheritances. CN is characterized by chronic neutropenia due to a congenital constitutional defect. In the past decade the molecular bases of several entities that facilitate a classification have been discovered. **Objective:** To report the case of a woman with CN, with maturation halt in the myelocytes to metamyelocytes phase: partial albinism, nystagmus, convergent strabismus and with a genetic history linked to the X chromosome. **Material and Methods:** A 4 year old girl was identified with CN, convergent strabismus, horizontal nystagmus and partial albinism; she was followed up for 17 years until she died of respiratory failure secondary to pulmonary fibrosis. During this period of time we expected a similar syndrome description to arise, however no other syndrome appeared. Nevertheless, three family members appeared with the same characteristics of the syndrome and we did the investigation through a family tree, we found it was linked to the X chromosome and lethal in males affected. **Results:** The patient had repeated upper and lower respiratory tract infections at a young age as well as otitis, gingivitis and skin infections. It was characterized by severe neutropenia with normalization of neutrophils with the application of G-CSF. The patient died at 21, she developed pulmonary fibrosis one year earlier. In her genetic history three more women were identified with the same syndrome: two first cousins, one of them died at age 10 from an intra-abdominal infection, the other one survives but suffers from repeated infections and moderate to severe neutropenia; the other one is 2 years old, she is first cousin of the patient that still survives, with the same characteristics of the syndrome. Comparisons were carried out with the different syndromes known that could be considered as one more of the

ones already described; however, not all phenotype, genetic or histological characteristics were linked. Therefore, it is important to investigate a genetic molecular defect to include it in the current classification of congenital neutropenias. **Conclusions:** This is a new syndrome not described yet in medical literature; it is characterized by congenital neutropenia with a halting of granulocyte maturity in the myelocyte to metamyelocyte phase, partial albinism, convergent strabismus, nystagmus and linked to the X chromosome.

A1068

DOES IT INCREASE MORTALITY IN PATIENTS WITH FEBRILE NEUTROPENIA NOT RECEIVE ANTIMICROBIAL TIMELY? Alvarado-Ibarra M, Juan Lien-Chang L, López-Hernández M, Alvarez-Vera J. Centro Médico Nacional "20 de Noviembre". ISSSTE

Introduction: Febrile neutropenia is a complication of chemotherapy that have a threat for patient's life, because the rate of bacterial growth is logarithmic. It has been observed that a delay ≈ 24 hours after the first fever spike occur in patients with absolute neutrophil count less 100 cell was associated with a high incidence of mortality. **Objectives:** Determine the impact of delay in the administration of antimicrobials in patients with high risk febrile neutropenia on morbidity and mortality. **Material and Methods:** Patients over 15 years old from January 2010 to August 2011 with diagnosis of acute lymphoblastic leukemia, acute non lymphoblastic leukemia and ambiguous lineage, who received intensive chemotherapy and had neutrophils less than 500 cell. Who courses with temperature up 38°C. Was quantified in minutes elapsed time between the onset of fever or identification of infection, the medical indication and antibiotic administration. Blood cultures were taken at baseline and every 5 days, we assessed the success or failure of the antimicrobial scheme as well the development of septic shock and death. **Results:** We evaluated 166 events of febrile neutropenia, 69.3% with initial absolute number of neutrophils less 100 cell. 76.5% had success of the program presented with fever and neutropenia, 9% had treatment failure and 10.8% died from infectious causes. 22% coursed with septic shock. Those without septic shock, 63.8% and 36.2% had and did not have attachment to the program, respectively ($p=0.007$). Time between the presentation of fever and indication of the antimicrobial in patients with septic shock

and those who had and did not have an average of 63 and 38 min ($p=0.16$), time between indication of antimicrobial and administration was an average of 163 and 76 min ($p=0.003$) and total time from the onset of fever to antibiotic administration was an average of 229 and 115 min ($p=0.0003$). Total time between onset of fever and administration of antibiotic had an average of 120 minutes in patients who had success, 72 minutes for patients who had failure and 291.67 minutes for patients who died ($p=0.0001$). In multivariate analysis, statistical significance was found between time elapsed from time of medical indication to time of antimicrobial administration ($p=0.0001$) and total time from the onset of fever to antibiotic administration ($p=0.0001$). **Conclusion:** Delay in the administration of antimicrobials in patients who present febrile neutropenia after intensive chemotherapy has a direct impact on the presentation of septic shock and death.

A1072

PREVALENCE OF IMMUNOPHENOTYPES IN NEOPLASTIC HEMATOLOGICAL DISEASES IN LABORATORIOS FATIMA DE MICHOACAN. Campos-Cabrera G*, Campos-Cabrera S*, Campos-Cabrera V**, Mora-Torres M***, Gomez-Guijosa MA***, Rivera-Trujillo A****, Hernandez-Rodriguez S****, Pita-Ramirez L⁵, Campos-Villagomez JL*. * Departamento de Hematología de Laboratorios Fatima de Michoacan, Morelia, Mexico; ** Departamento de Patología Clínica de Laboratorios Fatima de Michoacan, Morelia, Mexico; *** Servicio de Hematología del H. G. R. No.1 IMMS, Morelia, Mexico; **** Servicio de Hematología del Hospital Civil "Dr. Miguel Silva", Morelia, Mexico; ⁵Servicio de Hematología del Hospital General "Vasco de Quiroga" ISSSTE, Morelia, Mexico. **Introduction:** Medical indication of flow cytometric immunophenotyping includes diagnosis, classification, prognosis and disease monitoring. This is an important tool that each time is more used in hematology. **Objective:** To determine the prevalence of immunophenotypes in neoplastic hematological diseases in our region. **Material and Methods:** Bone marrow samples referred to Laboratorios Fatima de Michoacan for flow cytometry immunophenotyping for neoplastic hematological disease. The protocol is based on the Report on the Second Latin American Consensus Conference for Flow Cytometric Immunophenotyping of Hematological Malignancies (Cytometry Part B (Clinical Cytometry) 2005;70B:39-44). **Results:** One hundred and thirty three cases were diagnosed. Fourteen myelodysplastic syndromes. Thirty two acute myeloid leukemia; M0-M1 15 cases, 4 with aberrant expression of CD7 and 1 with CD19; M2 2 cases, 1 with aberrant expression of CD 2; M3 3 cases; M4 1 case; M5 9 cases, 2 aberrant expression of CD7; M6 and M7 one case each. Forty nine B cell precursor acute lymphoblastic leukemia; BI 21 cases, 2 with aberrant expression of CD7, and 2 aberrant expression of CD13; BII 16 cases, 1 aberrant expression of CD2, 1 aberrant expression of CD5, and 2 aberrant expression of CD33; BIII 11 cases, 1 aberrant expression of CD3; BIV 1 case. Five T cell precursor acute lymphoblastic leukemia. Twenty six chronic lymphoproliferative disorders; 11 B cell chronic lymphocytic

leukemia, 2 T cell chronic lymphocytic leukemia, 7 follicular lymphoma, 3 mantle cell lymphoma, 3 splenic lymphoma with villous lymphocytes. One adult T cell leukemia/lymphoma. Six monoclonal gammopathies. **Conclusions:** It is important to create the experience with new diagnostic tools based on the regional protocols. Low prevalence of AML M2 presumably because of classic morphologic features. Low prevalence of monoclonal gammopathies because for recent incorporation of diagnosis, treatment and response criteria; it is expected that in future this prevalence will arise.

A1129

AUTOIMMUNE LYMPHOPROLIFERATIVE SYNDROME: DIFFERENTIAL DIAGNOSIS OF CYTOPENIAS. CASE REPORT. Moreno-González AM, Quesada-Chalita CT, Enciso-Peláez S, Maldonado-Bernal MC, Sánchez-Zauco NA, Castillo-Martínez ID. Hospital Infantil de México Federico Gómez, México DF, México.

Introduction: Autoimmune lymphoproliferative syndrome (ALPS) is a disorder of T cell dysregulation caused by defective Fas-mediated apoptosis. Testing for ALPS should be considered in patients with unexplained lymphadenopathy, hepatosplenomegaly and/or autoimmunity. An increased number of a T cell population termed "double negative T cells" (DNTs) are present; cell phenotype CD4-/CD8-, CD3+, TCRαβ+. DNTs typically represent a small subset of T cells (<1%) in peripheral blood in unaffected individuals. Almost all mutations are in genes associated with the Fas apoptotic pathway. **Objectives:** Describe three patients with ALPS. **Material and Methods:** Case report. **Results:** Case 1. A 12-year old male with family history of Wegener's granulomatosis. Epistaxis, easy bruising and gingival bleeding. At admission severe neutropenia and thrombocytopenia were found with multiple lymphadenopathies in neck. Bone marrow aspiration showed left shift in myeloid cells, hyperplastic mature megakaryocytes, unremarkable lymphoid cells. EBV serology reports positive VCAG, EAD and EBNA. ANA, anti-DNA, direct Coombs and lupus anticoagulant were positive, IgG elevated. Thoracic-abdominal CT showed diffuse lymphadenopathy. A cervical lymph node was biopsied and atypical cortical and paracortical hyperplasia with progressive transformation of germinal centers was found. The DNTs cells were elevated. After prednisone treatment cytopenias remitted. Case 2. A 13-year old male with splenomegaly and bicytopenia. Bone marrow aspiration showed normal cellularity, hyperplastic megakaryocytes, and abundant plasma cells. Primary immune thrombocytopenia was diagnosed but patient did not respond to prednisone. ANA with a homogeneous pattern, anti-DNA, c-ANCA and lupus anticoagulant with a direct Coombs were positive. DNTs elevated. Patient 3 years later the present developed sepsis and neutropenic colitis requiring IVIG and wide spectrum antibiotics, at discharge with prednisone. Because of persistent bicytopenia mofetil mycophenolate was started. Case 3. A 11-year old male with epistaxis, gingival bleeding

and bruising. Admitted with fever and otorrhea. Physical exam was positive for splenomegaly. Laboratory work-up showed pancytopenia, positive direct Coombs, polyclonal hypergamma globulinemia. Bone marrow aspiration reported hypercellularity with hyperplastic immature megakaryocytes and maturation arrest in the myeloid serie. ANA with a speckled pattern was positive as well as lupus anticoagulant. IgG serology for CMV and EBV (viral capsid and EBNA) were positive. High levels of vitamin B12. DNTs were also elevated. ALPS was diagnosed 11 months later, starting treatment with prednisone with resolution of patient symptoms and cytopenias. **Conclusions:** ALPS is a rare syndrome caused by defective lymphocyte apoptosis with a wide range of symptoms including cytopenias. This diagnosis should be considered in any child with autoimmune cytopenias.

A1153

HAEMOPAGOCYTIC SYNDROME SECONDARY TO CITOMEGALOVIRUS (CMV) IN IMMUNOCOMPETENT HOST. Gutiérrez-Serdan R, Armenta-San Sebastian JA, Gómez-Plata E. Centro Oncológico Estatal ISSEMyM.

Introduction: Hemophagocytic lymphohistiocytosis (HLH) is a disorder of the mononuclear system that is characterized by nonmalignant generalized histiocytes proliferation with uncontrolled haemophagocytes and cytokine overproduction. The syndrome can be primary or secondary associated with malignancies, autoimmune diseases, variety of infections. The HLH has been associated with a variety of viral infections. **Objectives:** Describe a case of haemophagocytic syndrome associated with mononucleosis secondary to cytomegalovirus. **Material and Methods:** A 24 year-old woman presented to the hospital with a history of fever, fatigue, cough and cervical lymphadenopathy with pain. she had received non steroidal anti-inflammatory drugs and ceftazidime 5 days before her admission. On initial evaluation the patient appeared acutely ill. A skin lesions in her face only. There was cervical and axillary lymphadenopathy, pulmonary and cardiovascular evaluation did not reveal any pathology. She has hepatosplenomegaly, genitourinary and neurological examination was unremarkable. The laboratory revealed a pancytopenia, lactate dehydrogenase 3316 U/L, serum ferritin 800 ug/l, VSDG 38, aspartate amino transferase 313 U/L, alanine aminotransferase 74 U/l, hypertriglyceridaemia 353 mg/dl, fibrinogen 203 mg/Dl. Multiple cultures of blood, urine were negative for bacteria, mycobacteria or fungi, serological study for HIV 1 and 2, hepatitis B and C was negative. For the suspicious of mononucleosis we did serological studies for EBV and CMV, CMV IgM antibody detected by ELISA was positive and the biopsy of lymph node showed lymphohistiocytosis. Bone marrow aspiration showed hypocellularity revealed activated macrophages with engulfed leucocytes, erythrocytes, platelets. **Results:** We did not find any clinical or laboratory features of systemic disease. The therapy was initiated with ganciclovir and symptoms and laboratory abnormalities improved, and the patient was discharge 7 days after admission. **Conclusions:** HLH is an

unusual disorder, according to the Histiocyte Society five of the eight criteria were fulfilled. Primary CMV infection in the immunocompetent host rarely causes serious illness, however CMV infection is the first causes of HLH in immunocompromized hosts, representing 10%. Our case illustrates the possibility of HLH during the course of CMV early infection. Few cases are described in the literature. In our case we use antiviral treatment and the patient improved. Hemophagocytic syndrome should always be taken into account in the differential diagnosis of fever with obscure etiology.

A1200

WAS GENE MUTATION IN A FEMALE PATIENT WITH SEVERE INHERITED THROMBOCYTOPENIA. Boonyawat B*, Dhanraj S*, Abbas FA*, Zlateska B*, Gruenbaum E*, Steele L*, Shago M*, Meyn S*, Blanchette V*, Ochs H**, Dror Y*. *The Hospital for Sick Children, University of Toronto, Toronto, Ontario, Canada; ** Seattle Childrens Research Institute, University of Washington, Seattle, Washington.

Introduction: Wiskott-Aldrich syndrome (WAS) is an immunodeficiency disorder characterized by recurrent infection, eczema, thrombocytopenia with small platelets, and increased risk of autoimmune disorders and cancers. WAS is caused by mutation in the WAS gene located in Xp11.22-p11.23. Mutations in the WAS gene can also cause X-linked thrombocytopenia (XLT), both disorders are typically seen in males. Nevertheless, there have been few reports of female cases of WAS. Herein, we describe a female infant who presented with clinical features resembling WAS phenotype. Molecular analysis revealed a spontaneous heterozygous mutation in exon 4 of WAS on the paternally derived X chromosome and extremely skewed X-chromosome inactivation with preferential selection of the maternally derived wild-type X-chromosome to be inactivated. WASP expression revealed no WASP protein. **Objectives:** To Identify genetic cause in patient with immunodeficiency syndrome. **Material and Methods:** Direct sequence analysis of the WAS gene was performed in patients Genomic DNA and cDNA samples from peripheral blood lymphocytes and expanded T cells. Analysis of X-chromosome inactivation was performed by amplification of androgen receptor (AR) gene and studying methylation status. The X-inactivation ratio was calculated by peak height difference for each parental allele in the daughter. Genome-wide analysis of genetic alterations in the patient was performed using Affymetrix SNP 6.0 Array. Alterations were deemed as true hits only if they were detected by at least 2 of the 3 algorithms (GTC, HMM and Birdsuite). For evaluation of WASP protein expression, nucleated blood cells were ficoll separated, washed and incubated with anti-WASP antibody conjugated to FITC. Cells were washed and analyzed by flow cytometry. **Results:** Analysis of genomic DNA revealed mutation in exon 4 of WAS gene and cDNA sequences suggesting the absence of normal WAS cDNA (Fig. 1). In contrast, sequence analysis showed normal DNA sequence in both parents. Patient's

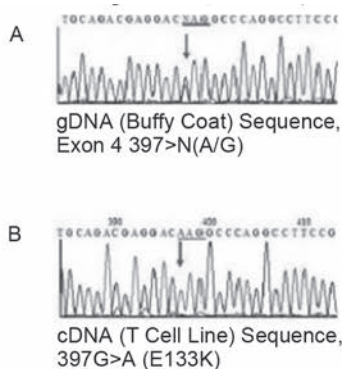
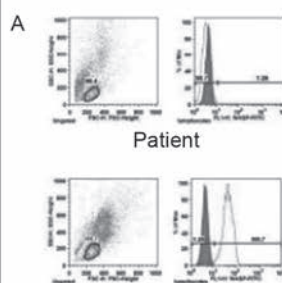


FIG - 1



B

Sample Name	WASP%	WASP MFI
Control R-IgG	0.39	4.63
Control, anti WASP	98.7	51.6
Patient R-IgG	0.2	4.52
Patient, anti-WASP	1.26	4.14

FIG - 2

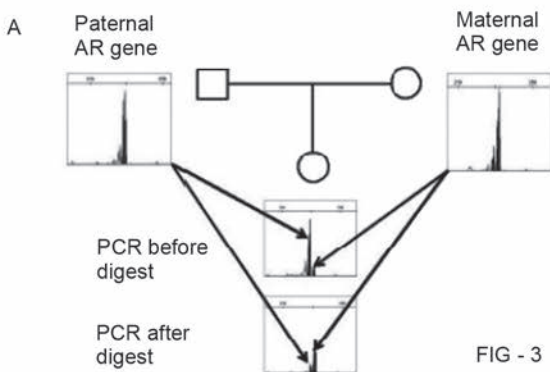


FIG - 3

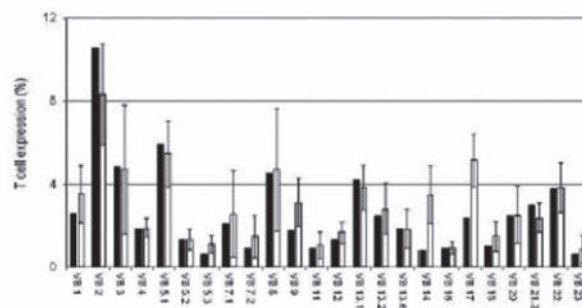


FIG - 4

peripheral blood showed no WASP expression (Fig.2).The XCI analysis in the patients and her parents revealed non-random or skewed pattern of XCI (Fig.3). T cell repertoire demonstrated the presence of all V-beta families without clonal expansion, with reduced percentage of Vb 14 at 0.8 and Vb 17 at 2.4 (Fig.4). **Conclusions:** We describe a female infant with clinical features of WAS with heterozygous mutation in the WAS gene and X-inactivation. The cumulative findings demonstrate how female carrier with heterozygous WAS mutation can manifest classical X-linked WAS phenotypes and the need to test selected female patients with complete or incomplete disease expression for X-linked disorders, particularly when genetic and protein expression assays become widely available.

A1207

PRIMARY AND SECONDARY IMMUNE THROMBOCYTOPENIA (IT). PRIVATE PRACTICE EXPERIENCE. Ovilla-Martínez R, Guerra-Urbe NB, Terrazas-Marín RA, Herrero-Maceda MR, Amador-Acosta D, Vázquez-Gutiérrez JR, Angeles-Labra AR, Buganza-Torio E, Torres-Flores JC, Barrera-Carmona CC, Madinaveitia-Turcott JR, Enríquez Ocaña JC, Calles-Payan DI. Hospital Angeles Lomas, México.

Introduction: IT is a platelet antibody mediated destruction which can be primary (isolated thrombocytopenia <math><100 \times 10^9/L</math>)

or secondary as a consequence of any other primary immune disease, such as infections. Our objective is to assess each treatment based on as primary or secondary disease. **Material and Methods:** We included pediatrics and adults diagnosed with IT between 1998 and 2012, documented on their medical history, clinical presentation and laboratory findings. As the criterion established, the initial treatment was based on steroids and IV immunoglobulin and a subsequent treatment based on the clinical subset, Mycophenolate Mofetil (MMF), Rituximab or splenectomy, all but one patient were monitored until their medical discharge. **Results:** 64 patients were studied. Platelet initial count was $2-54 \times 10^9$, with the following characteristics: 23 patients presented previous infections within 4 weeks before diagnosis. 25 patients had a concomitant disease, however only 12 had a relation with thrombocytopenia. There was no steroid dependent reported. Complete response was reported in all groups; those who didn't reached it with first line treatment, were managed with splenectomy, with (2 cases) or without rituximab following splenectomy. **Conclusion:** In this review, it was identified a greater incidence of primary IT, mostly female, as well as better response at the initial management. There is important to treat the causing disease in patients with secondary IT.

A1217

HEMOPHAGOCYTIC LYMPHOHISTIOCYTOSIS SYNDROME. NEONATAL CASE REPORT. Paredes-Aguilera R,

Lopez-Santiago N, Nuñez-Toscano M. Instituto Nacional de Pediatría. Departamento de Hematología..

Introduction: Hemophagocytic lymphohistiocytosis syndrome (HLH) is rare clinical condition characterized by prolonged fevers in association with hepatosplenomegaly, cytopenias, coagulopathy, and central nervous system (CNS) manifestations. HLH results from a pathological activation of macrophages leading to hyperproduction of cytokines, such as gamma interferon (IFN γ) and tumor necrosis factor alpha (TNF α), that is believed to be the cause of many of the clinical symptoms. HLH is currently classified into a familial form, affecting primarily infants and young children, and a secondary form, which usually occurs in older children. Detection of molecular genetic abnormalities in genes involved in immune response pathways, Clinical diagnosis is largely made according to HLH-2004 criteria. However, a new finding of the Th1/Th2 cytokine pattern is a useful biomarker for the early diagnosis, differential diagnosis, and the monitoring of the disease. Intensive immunosuppressive therapy is generally accepted as treatment for the relief of clinical symptoms/signs, while allogeneic hematopoietic stem cell transplantation is currently the only potentially curative therapy option for severe familial forms of HLH. **Objective:** Describe a case and evolution a neonatal case of HLH. **Material and Methods:** infant of 32 weeks of gestation, mother with Lupus Systemic Erythematosus and history for EBV infection with a history of thrombocytopenia, fever, hepatosplenomegaly, elevation of renal function tests and liver, little response to standard management. It showed the presence of hemophagocytosis in bone marrow to start handling with dexamethasone, etoposide, cyclosporin A. Notice is sent cytokines, TH1, TH2, as well as EBV viral loads for CMV. **Results:** response was measured weekly with biochemical parameters as well as reduction of response clinically Megalies. **Conclusions:** The identification of a variety of genetic abnormalities that lead to the deficiencies of the cytotoxic granules exocytosis pathway has provided a deeper understanding of

these disorders. However, these genetic forms only account for 30–70% of FHL cases. Other inherited HLH conditions remain to be molecularly identified, which is essential for the establishment of accurate diagnosis and efficient therapy. In clinical practice, still often overlooked since the clinical symptoms are also found in immune-competent patients with certain special infections. Survival of HLH patients has improved from close to 0% to over 60% with the use of cytolytic and immunosuppressive drugs combined with stem cell transplantation. Most deaths occur due to the disease itself; therefore, it is important for us to recognize and treat this condition earlier and physicians should learn to move more rapidly to salvage therapy for non-responding patients.

A1220

ANTIBIOTIC RESISTANCE RATES IN BACTERIA ISOLATED FROM BLOOD CULTURES IN FEBRILE PATIENTS OF AN ADULT GENERAL HEMATOLOGY WARD ORGANISMS BLOOD CULTURES. Malaga-Centeno J* San-Martin-Bustinza A**. *Hematology Unit .Carlos A. Seguin Hospital. Arequipa-Peru; **Clinical Pathology Unit. Carlos A. Seguin Hospital. Arequipa-Peru.

Introduction: Antibiotic resistance can be dangerous for immunodepressed patients . Infections due to antibiotic resistant bacteria can be difficult to treat and can mean longer lasting illnesses, extended hospital stays, and the need for more expensive and toxic medications. Some resistant infections can even cause death. It is important to know antibiotic resistance rates among bacteria in hematological patients. **Objectives:** To determine antibiotic resistance rates in bacteria isolated from blood cultures in febrile patients of an adult general hematology ward. **Material and Methods:** We performed a retrospective study of 183 patients with a febrile episode from November 2010 to January 2012 who were hospitalized in an adult general hematology ward of the Carlos Seguin Hospital in Arequipa - Peru. They were diagnosed with Acute leukemias, Lymphoma , Aplastic anemia,

	Pediatrics		Adults	
	Male	Female	Male	Female
Primary IT	16 (25%)	8 (12.5%)	11 (17.18%)	17 (25.56%)
Secondary IT	1 (1.56%)	1 (1.56%)	2 (3.12%)	8 (12.5%)

Treatment	Primary IT			Secondary IT		
	Complete response	No response	Refractory disease	Complete response	No response	Refractory disease
Prednisone	37	7	13	17	5	2
Dexamethasone	1	1	0	0	0	0
IVIG	10	3	4	3	4	2
MMF	2	0	1	1	1	0
No Treatment	2	2	0	0	2	2

Myelodysplastic syndrome, Multiple Myeloma. Nearly 70 % patients were neutropenic. The results of 425 blood cultures were reviewed. **Results:** The rate of blood culture positivity was 2.53%. Of 435 blood cultures results, 11 were positive. Of 183 patients with a febrile episode, 9 (4.92%) had a positive blood culture. Blood samples obtained for culture yielded Gram positive bacteria (63.4%) and Gram negative bacteria (36.6%). They yielded staphylococcus epidermidis (27.27%), E. coli (27.27%), staphylococcus hominis (18.18%), klebsiella pneumoniae (9.09%), coagulase-negative staphylococcus (9.09%), staphylococcus auricularis (9.09%). Around 75% of Gram negative bacteria were found to exhibit multidrug resistance. 100% of Gram negative bacteria were extended spectrum beta-lactamase (ESBL) positive. 50% of Gram negative bacteria were resistant to Ertapenem. Around 85% of Gram positive bacteria were beta-lactamase positive. None of the gram positive bacteria were resistant to Vancomycin. **Conclusions:** The antibiotic resistance rates in bacteria isolated from blood cultures in febrile patients of an adult general hematology ward are high. The rate of blood culture positivity was very low.

A1237

WEEKLY CULTURES INCREASES THE FREQUENCY OF ISOLATION OF BACTERIA IN PATIENTS WITH NEUTROPENIA ASSOCIATED WITH CHEMOTHERAPY. Ramos Peñafiel CO*, Cabrera García A*, Balderas Delgado C*, Olarte Carrillo I**, Martínez Tovar A**, León González G*, Hernández

Sánchez M***, Rozen Fuller E*, Collazo Jaloma J*, Martínez Murillo C*. *Department of Hematology, Hospital General de México O.D; **Department of Molecular Biology, Hospital General de México O.D; ***Department of Infectology, Hospital General de México O.D.

Introduction: Sepsis is one of the mayor cause of death in cancer patients. The frequency of bacteria isolates is low. Gram-negative bacteria like Escherichia coli are the most frequent followed by Staphylococcus and Pseudomonas aeruginosa. **Material and Methods.** We performed a retrospective study in patients with acute leukemia who received intensive chemotherapy regimens. **Results.** 70 patients were studied, 80% correspond to an acute lymphoblastic leukemia. The most frequent bacteria were Gram-positive (56%) (Staphylococcus epidermidis, Staphylococcus aureus) followed by Gram-negative (Escherichia coli, Klebsiella pneumoniae). The most frequent bacteria was S epidermidis in a 32% followed by Escherichia coli in a 14.7%. The frequency of Pseudomonas aeruginosa was 4.3%. The major resistance was observed with ciprofloxacin especially in Pseudomonas (67%), in this the greater sensibility were for piperacilin and cefepime (85%). For Escherichia coli the main sensitivity was for imipenem, meropenem and amikacin (95%, 50 y 85% respectively). All Staphylococcus were sensitive to vancomycin (>90%). **Conclusion.** This strategy increased the frequency of bacterial isolates to 91%, the nasal cavity being the main site. Mortality was not impacted by the type of isolated germ.

ACUTE LEUKEMIA

ORAL PRESENTATION

A1043

BIOLOGICAL DOSIMETRY TO ASSESS THE EXPOSURE OF HANDLING ANTINEOPLASTIC DRUGS BY MICRONUCLEUS COUNT IN ORAL MUCOSA CELLS. Rodríguez L*, Rodríguez E**, Zúñiga R***, Retamales E****. *Subdpto. de Asuntos Científicos (ISP); **Dpto. Salud Ocupacional (ISP); ***Hospital Carlos van Buren de Valparaíso; ****Hematología y Banco de Sangre; Instituto de Salud Pública de Chile (ISP).

Introduction. The antineoplastic drugs are widely used in therapy against the cancer and others pathologies. The action mechanisms above mentioned have been useful to stop the advance of malignancies in spite of the adverse reactions that they could present. The health workers that manipulates oncologic treatments in hospitals or chemotherapy centers, they would be potentially exposed to the genotoxic effect but they adopt safety strategies for prevention. This effect is described in the literature and will be measured using cytogenetic methods such as study of chromosomal aberrations, sister chromatid exchange, and Micronuclei (MN) analysis. Likewise, citostatic metabolite chemical analyses in urine has shown the presence of this substances. The Micronuclei analysis is a simple, low complexity and rapid laboratory analysis. It is one of the tests used in search and quantification of this nuclear alteration in cells. **Objective.** The aim of this study it's determine the level of association between the number of micronuclei(MN) counted in exfoliated cells from oral mucosa in health care staff of 14 Chilean chemotherapy centers and the degree of exposure to antineoplastic drugs. **Material and Methods.** The participating subjects of the study were 140 volunteers, 97 exposed and 33 controls. The samples were stained with acridine orange and MN were counted in 1200 cells, using the likelihood ratio method and statistical analyses were performed using STATA Version 9.1. **Results.** The likelihood ratio method, obtained a cutoff of 16 MN confidence interval: 13 to 18. It is suggested that could be used as possible indicators of high levels of exposure. The 140 officers who participated in this study, the most exposed are: Nurses (EU) in 27/29 cases, Pharmacists (QF) in 17/24 cases and paramedical (TPM) in 31/39 cases. Administration and Preparation of cytostatics, while for Pharmaceutical Chemists, Nurses

and Paramedical Technicians presented the highest frequently of counts above 16 MN. **Conclusions.** Can be concluded that exposed people labor to antineoplastic drugs (manipulation and administration) have an increased number of MN in their oral mucosa compared with individuals other people's to that activity, would demonstrate a risky expose damage to the DNA of the epithelial cells. The suitable nonuse of the PPE for the manipulation of these drugs, could favor the MN increase, which would indicate the importance of the use of these elements for the exposed health workers.

A1083

MINIMAL RESIDUAL DISEASE AT DAY 14 AND 28 OF REMISSION INDUCTION AS A PROGNOSTIC FACTOR OF SURVIVAL IN CHILDREN WITH ACUTE LYMPHOBLASTIC LEUKEMIA TREATED AT HOSPITAL CIVIL DE GUADALAJARA DR. JUAN I. MENCHACA. Gallegos-Castorena S, Chavez-Panduro P, Cruz-Ororio RM, Sanchez-Zubieta F. Servicio de Hemato-Oncología Pediátrica, División de Pediatría, Hospital Civil de Guadalajara Dr. Juan I. Menchaca, Guadalajara, Jalisco; Instituto de Investigación en Cáncer en la Infancia y Adolescencia; Universidad de Guadalajara; Centro Universitario de Ciencias de la Salud Guadalajara, Jalisco.

Introduction: Response to treatment of Acute Lymphoblastic Leukemia is the most important prognostic factor for survival. Detection of minimal residual disease (MRD) is the most important accurate measurement to define this response during induction remission treatment in order to allow early modifications of treatment to improve survival. **Objective:** To define the prognostic value of minimal residual disease at day 14 and 28 of induction remission treatment on survival of patients with Acute Lymphoblastic Leukemia (ALL) treated at Hospital Civil de Guadalajara Dr. Juan I. Menchaca from August 2007 to December 2009. **Material and Methods:** We performed a descriptive clinical prospective study in children with ALL treated at our institution from August 2007 to December 2009. MRD was measured by immunophenotype by flow cytometry at day 14 and 28 of induction remission. It was considered negative with 0.00% of leukemic cells and positive divided in three groups: A > 0.00% to < 1%; B > 1% to 5% and, C > 5%. Statistical analysis was performed with SPSS 17. Survival was analyzed with Kaplan Meier test and compared with log rank

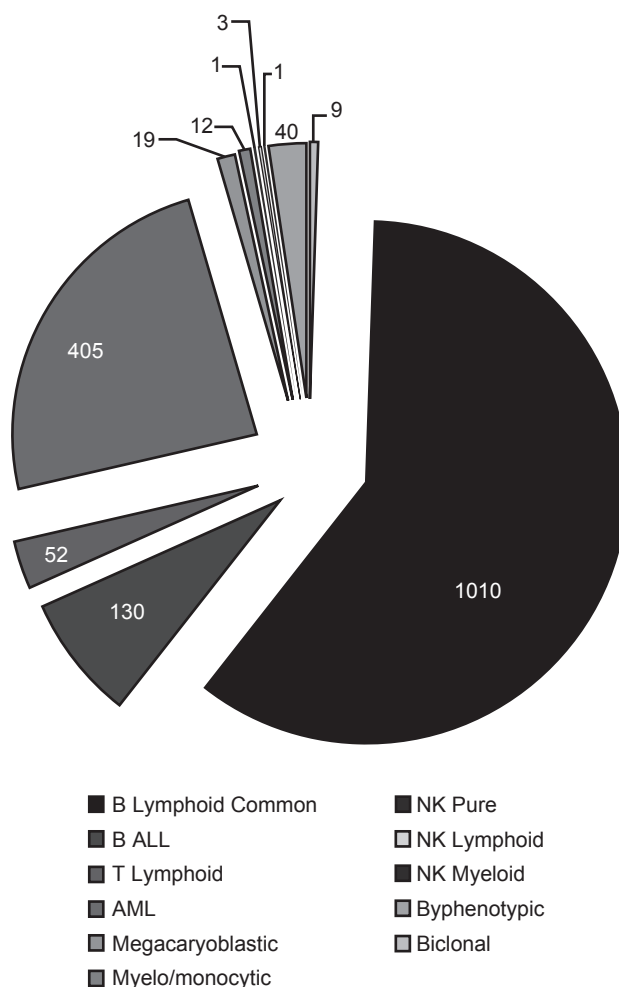
test. Frequencies were used for descriptive variables. **Results:** There were 113 patients with ALL during study period, 102 were evaluable. Patients had mean age of 6.2 years (1-15); 57 male, 44 female; 99 had PreB, 1 T-cell and 1 B mature cell immunophenotype. Fifteen were classified as standard risk and 86 as high-risk at diagnosis. Minimal residual disease at day 14 was negative in 73.5%; > 0-1% in 9.8%; > 1-5% in 9.8% and > 5% in 6.9%. Event-free survival according to day 14 MRD was 76.4%; 30%, 50% and 14.3% respectively, with a median follow up of 37 months. Log rank test was 0.001. MRD at day 28 was negative in 87%; > 0-1% in 6.9%; 1-5% in 2.9% and > 5% in 2.9%. Event-free survival was 70% for patients with less than 1% at day 28 and 0% for 6 patients > 1% at day 28 of induction. Standard-risk cases had 80% event-free survival and high-risk had 67.6% when MRD was negative at day 28. **Conclusions:** Minimal residual disease status at day 14 and 28 induction was a prognostic factor for event-free survival in children with ALL treated at Hospital Civil de Guadalajara Dr. Juan I. Menchaca.

A1087

PREVALENCE OF ACUTE LEUKEMIA IMMUNOPHENOTYPES IN CLINICA RUIZ-LABORATORIOS. Perez-Romano B, Fragoso J, Ruiz-Arguelles GJ, Ruiz-Arguelles A. Laboratorios Clinicos de Puebla and Centro de Hematología y Medicina Interna, Puebla, Puebla, Mexico.

Introduction: The results of the immunophenotypic analysis of newly diagnosed cases of acute leukemia, performed at Laboratorios Clinicos de Puebla, from January 1998 to December 2011, were reviewed and classified according to the Latin American Consensus Conferences for Immunophenotyping of Hematological Malignancies. Inasmuch as classification criteria were different at each Conference, cases were divided into those studied from January 1998 to April 2005, and those studied hereto from. **Objectives:** To determine the prevalence of acute leukemia phenotypes in cases studied at Laboratorios Clinicos de Puebla, and to know the prevalence of aberrant antigen expression in such cases. **Material and Methods:** Flow cytometric immunophenotyping was performed in either blood or bone marrow samples following the recommendations derived at the Latin American Consensus Conferences. **Results:** During the first period, a total of 1682 cases were classified. The vast majority of them corresponded to B cell precursor acute leukemia (1140 cases, 88%), followed by myeloid leukemia (436), T cell acute leukemia (52 cases) and NK acute leukemia (5 cases). Biphenotypic and bclonal cases were 40 and 9 respectively. During the second period, a total of 4115 cases were studied, and the proportions of B cell, T cell and myeloid precursor cases were almost identical. Of the 2487 B cell precursor acute leukemia cases, 90 were classified as B-I, 1934 as B-II, 102 as B-III and 66 as B-IV. In 295 cases, subtyping was not possible. Aberrant antigen expression is a common finding. In our cases, only 8% of B cell precursor acute leukemia cases expressed CD13 and/or

CD33; expression of CD10 was seen in 28% of T cell cases while expression of lymphoid cell antigens in acute myeloid leukemia cases was demonstrated in 23% of the cases. **Conclusions:** The prevalence of immunophenotypes and subtypes of acute leukemia was established at Laboratorios Clinicos de Puebla, and it is clear that the relative proportions of types has not changed over time. The aberrant expression of certain antigens in acute myeloid leukemia is similar to that reported in caucasian populations; while that in both B and T cell acute leukemia is different. These differences might be explained on ethnic rather than on environmental factors.



A1109

COMPARISON OF TWO INDICES OF SERUM GALACTOMANNAN ASSAY FOR DIAGNOSING INVASIVE PULMONARY ASPERGILLOSIS IN HEMATOLOGY PATIENTS. Prem S*, Singh R**, Xess I**, Kumar R***, Mahapatra M**, Saxena R**, Sharma S**. *Regional Cancer Centre, Trivandrum, India; **All India Institute of Medical

Sciences, New Delhi, India; ***Cancer Care, Manitoba, Winnipeg, Canada

Introduction: In this prospective blinded study, we included patients of acute leukemia and febrile neutropenia unresponsive to broad spectrum antibiotics. **Objective:** We compared two different positive cut-offs of galactomannan assay for diagnosing Invasive Pulmonary Aspergillosis (IPA). **Material and Methods:** Blood samples for galactomannan were taken from leukemia patients with clinical or radiological signs of pulmonary infection & tested for galactomannan by Platelia Aspergillus test (Bio-rad) by an investigator blinded to the sample source and clinical data. Optical densities were read at 450 and 620 nm. To get the galactomannan index the O.D. of the test serum was divided by the mean O.D. of the calibrator serum. All samples were tested in duplicate. The ELISA results were analyzed at two different positive cut-offs- >1.5 (manufacturer's recommended positive cut off) and >1.0. After all samples were analyzed, data were combined with the clinical data which was collected independently. IPA was classified according to EORTC-MSG criteria. Results of galactomannan detection were excluded from the criteria. Statistical analysis. Total group of patients with IPA was defined as sum of patients with proven, probable & suspected IPA. Patients without IPA were defined as patients not classified in any IPA category and who did not receive empirical antifungal treatment. Calculations of sensitivity, specificity, positive predictive value & negative predictive value were based on these two groups. **Results:** A total of 80 patients with pulmonary infection were studied. Based on microbiological and radiological criteria and histopathology the final diagnoses were: a) definite / proven IPA-none, b) probable IPA -1 c) suspected IPA-32. The mean number of samples per patient was 2. Using a positive cut-off >1.5, sequential (>2 consecutive) positive results were recorded in 14 febrile episodes. On reducing the positive cut-off to >1.0, there was 28 sequential positive results. The sensitivity, specificity, positive predictive value and negative predictive value of the test were 72.7%, 94.4%, 96% and 65.4% respectively using an index of 1.0. With an index of 1.5 the sensitivity dropped to 42.4%. We did not find any change in specificity when the cut-off was reduced from 1.5 to 1.0 and this remained at 94.4%. **Conclusions:** We observed that the sensitivity of the Platelia galactomannan assay dropped sharply from 72.7% to 42.4% without any increase in specificity when the cut-off was increased from 1.0 to 1.5. Therefore the lower cut-off may be more appropriate for the early diagnosis of IPA in patients of acute leukemia with pulmonary infection.

A1118

INTERACTION BETWEEN THE SUSCEPTIBILITY TO DEVELOP ACUTE LEUKEMIA IN CHILDREN AND PATERNAL SMOKING. Mejia-Arangure JM, Perez-Saldivar ML, Flores J, Bernaldez R, del Campo MA, Medina A, Palomo MA, Paredes R, Martínez A, Rodríguez MC, Zarco J, Rivera R, Flores H, Goro-

dezky C, Salamanca F, Fajardo-Gutiérrez A. Instituto Mexicano del Seguro Social; Hospital Infantil de México Federico Gómez; Instituto Nacional de Pediatría, Secretaría de Salud.

Introduction: It has been suggested that acute leukemia (AL) can be the result of the interaction between inherited susceptibility to the disease and exposure to different carcinogenic factors. **Objective:** The present study evaluates the interaction between the susceptibility to develop acute leukemia and paternal smoking as a risk factor. **Material and Methods:** A Case-control study with selection by the susceptibility was realized; we had two types of cases and controls: 47 children with Down syndrome (DS) and AL who are known to be highly susceptible to develop AL, 423 children with AL without DS from the same population-base, a group of 207 children with DS without AL, and another group of children neither AL nor DS. The interaction was assessed with logistic regression. The results were adjusted by different variables. In our data there was not interaction between susceptibility for developing AL and paternal smoking. **Results:** The paternal smoking prior to the conception had an odds ratio (OR) of 5.9 (95% confidence interval, CI, 2.3, 15.8) when entire population was analyzed. In the population with DS the OR was 0.8 (0.4, 1.7) and in the group without DS was 5.4 (1.9, 15.1). With the postnatal paternal smoking the ORs were 2.6 (1.1, 6.5), 0.4 (0.2, 0.8) and 2.6 (1.1, 6.43), for entire population, children with DS and children without DS, respectively. **Conclusions:** The results did not show an interaction between the susceptibility for developing AL and paternal smoking. However the risks for paternal smoking before and after the conception were very high in the population without DS. The project was supported by the Fondo Sectorial de Investigación en Salud CONACYT.

A1228

SNP'S IN REDUCED FOLATE CARRIER (RFC) GENE, AS PREDICTOR OF SURVIVAL IN PATIENTS WITH ACUTE LYMPHOBLASTIC LEUKEMIA. Candelaria M***, Ojeda J*, Labardini J*, Gutiérrez-Hernández O**, Vidal-Millán S**, Corrales C**, Espinoza R*, Galvan-Salazar G*. *Hematology Department. **Pharmacogenetics Laboratory. Instituto Nacional de Cancerología (INCan). Mexico City, Mexico.

Introduction: Treatment for adult ALL (acute lymphoblastic leukemia) offers a long-term disease-free survival ranging from 20 to 35%. The prognosis is associated with host and malignancy characteristics, including leukemia cell phenotype, cytogenetic abnormalities and sensitivity to the treatment given. Hyper-CVAD scheme is used as first-line treatment in patients with ALL at the INCan. Reduced folate carrier (RFC) is involved in methotrexate inflow to the cell. Therefore SNP's associated with a lower activity may influence the sensitivity to this drug. **Objectives:** Assess by DHPLC the frequency of polymorphisms of genes involved in reduced folate carrier, as well as its influence on response and survival of patients with

ALL. Material and methods: A pilot prospective, open, exploratory, trial. Inclusion criteria: Age > 15 years, with normal renal and hepatic function, diagnosed with ALL, treated with Hyper-CVAD. Exclusion criteria: hypersensitivity to methotrexate or other drugs included in the Hyper-CVAD scheme. Sample Process: DNA was obtained by standard methods from peripheral blood, RFC gene was amplified by PCR and thereafter analyzed by DHPLC. The final results are reported as wild-type (wt) or polymorphism detected. Clinical characteristics were registered. All patients were evaluable for response (complete, partial and disease progression), by International Working Group for acute leukemia. Demographic statistical analysis was done for clinical variables & toxicity was tabulated. Cox regression analysis was done to evaluate factors influencing on response & overall survival. **Results:** From January 2010 to

June 2011, 31 (14 male/16 female) patients with ALL treated with Hyper-CVAD were included. 68 % had common B-ALL, median age was 26 years. RFC gene analysis was determined in 27 patients. Complete remission was achieved in 80,6% with the first phase of Hyper-CVAD. At 18 months of follow up the RFS was 83.9% (95% CI, 380-494) and OS of 51.6% [95% confidence interval (CI) 252-367 days. Of all clinical factors analyzed, only the immunophenotype and the allelic variant wild-type (wt) of the RFC, had a statistically significant influence on OS and RFS. **Conclusions:** These preliminary results demonstrated an influence of RFC polymorphism on RFS & OS. A larger sample is needed to prove these results. Pharmacogenetics analysis of genes influencing on transport & drug metabolism may be required for ALL patients before receiving chemotherapy to improve long-term OS.

POSTERS

A1009

HLA-A, -B, -Cw, -DRB1 AND -DQB1 ALLELES IN PATIENTS WITH B-CELL ACUTE LYMPHOBLASTIC LEUKEMIA (ALL). Solís-Martínez RA, Vázquez-Castillo ., Santos-de la Cruz ME, Pintado-González M, Gómez-Mar SG, Bustos-Colorado A. Department of Molecular Biology, Laboratorios Diagnóstica.

Introduction: The frequencies of HLA class I (A, B, Cw) and class II (DRB1 and DQB1) alleles has not been determined in Mexican mestizo patients with B-cell acute lymphoblastic leukemia (B-cell ALL), although several studies have demonstrated relationship between acute lymphoblastic leukemia and HLA allele's frequencies in others countries. **Objective:** The aimed of this study was determine the frequencies of HLA -A, -B, -Cw, -DRB1, -DQB1 alleles in Mexican mestizo patients with B-cells acute leukemia. **Material and Methods:** 19 patients with B-cell ALL were studied and compared with a healthy volunteers control group (n=33). The loci A, B and Cw of the class I HLA and the DBQ1 and DRB1 loci of the class II HLA were analyzed using Morgan® HLA SSP ABCDRDQ trays typing kit 1 test/tray with the SSP (Sequence Specific Primers) technique, according to the manufacturer Texas BioGeneInc®. Statistical analyses were carried out using the SPSS® statistical package, version 9.0 (SPSS Inc., Chicago, IL, USA) for Windows. **Results:** The frequency of HLA-A*02 were significantly higher in patients compared with the controls (74% vs 33%, $p = 0.005$). Not in the same way, but yet revealing, was the frequency difference of B*35 (63% vs 30%, $p = 0.021$). In the Class II HLA the ones that stand out are DQB1*04 and DRB1*08 which rises a 40% in patients compared with the controls ($p = 0.005$, and $p = 0.014$ respectively). **Conclusions:** Have been reported a wide variety of alleles of HLA which are related to the development of acute lymphoblastic leukemia, among them the HLA-A*02, supporting the results of this paper. The risk factor of the dominant alleles

were analyzed and highlighted A*02 with OR=5.6 ($p = 0.005$), B*35 with OR=3.943 ($p = 0.021$), DQ*04 with OR= 9.042 ($p = 0.005$) and DRB1*08 with OR=7.154 ($p = 0.014$). We conclude that HLA-A*02, B*35, Cw*04, DQB1*04 and DRB1*08 alleles are associated with the development of B-cell acute lymphoblastic leukemia in Mexican mestizo population.

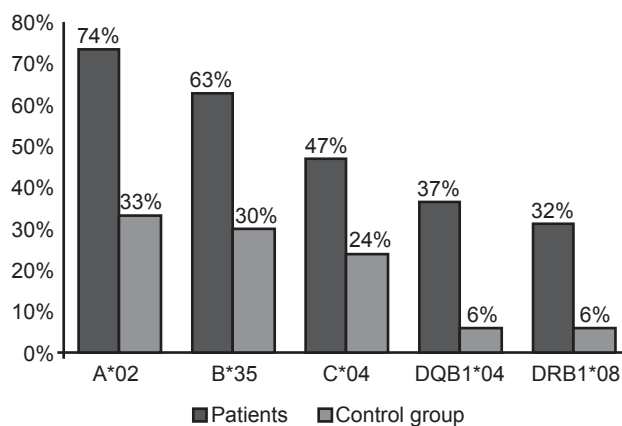


Figure 3. Alleles with significantly differences in frequency between B-cell ALL patients and healthy controls

A1029

IMMUNOPHENOTYPING ANALYSIS IN CEREBROSPINAL FLUID OF PEDIATRIC PATIENTS WITH ACUTE LYMPHOBLASTIC LEUKEMIA. Palomo M*, Zapata M*, Juárez L*, Riosvelasco F**, López B*, Mendoza E*, Huerta Y*, Maldonado C*. *Hospital Infantil de México Federico Gómez, México City, Distrito Federal, México. **Universidad Autónoma de Ciudad Juárez, Chihuahua, México.

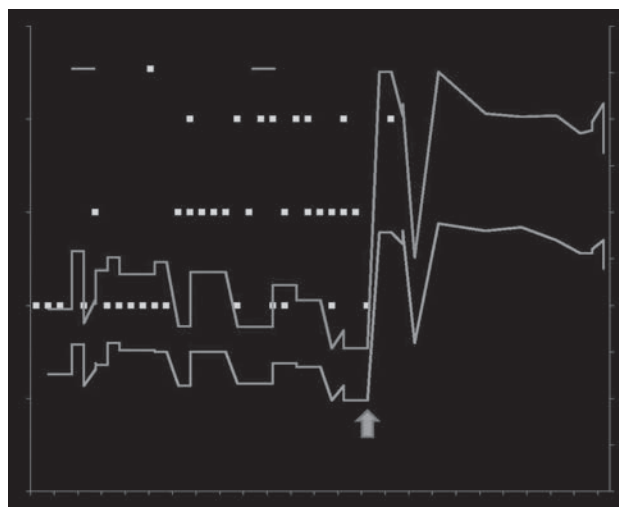
Introduction: Cerebrospinal involvement is a frequent complication of haematological malignancies, with an incidence of up to 25% in leukemias and lymphomas. The diagnostic gold standard to detect cerebrospinal fluid (CSF) involvement is cytologic examination by light microscopy; unfortunately, this technique is characterized by low sensitivity and low specificity. **Objective:** Asses the diagnostic accuracy of flow cytometric (FCM) immunophenotyping in comparison with classic cytology for diagnosing central nervous system (CNS) infiltration in ALL. **Material and Methods:** One hundred four CSF specimens from pediatric patients with ALL were examined by FCM for immunophenotyping. CSF fluid analysis was performed as part of their routine work up. The results were compared to classic cytology routinely done for all samples. Medical ethical committee of HIMFG approved this study. Children with ALL were eligible for inclusion in this study if they met the following criteria 1) they were newly diagnosed with ALL; 2) no prior chemotherapy and radiotherapy had been administered. Cells for immunophenotyping were obtained from 1 to 2 ml aliquot of CSF collected during the initial diagnostic lumbar puncture. All samples were studied within 6 hours of collection. Monoclonal antibodies against cell surface antigens included CD10 and TdT. **Results:** In this work 104 CSF were examined. Nineteen were positive [19/104 (18.2%)] and 85 negative for light microscopy. Twenty-five samples were positive by FCM [25/104(24%)]. A total of 25/104 positive samples were detected ; 14 samples were positive for both FCM and cytology [14/30 (46.6%)]. Eleven samples were positive by FCM and negative by cytology [11/30(36.65)]. Five samples were positive by cytology and negative by FCM [5/30(16.6%)] . Intraobserver agreement for light microscopy was in our study, with a k index of 0.53. Sensitivity was 0.73, Specificity was 0.87 , Positive predictive value 0.56 and negative predictive value 0.93. **Conclusions:** The diagnosis values of FCM are two-three times more than that of cytology. Immunophenotyping by FCM is recommended for routine diagnosis of CSF infiltration combined with cytology to increase the diagnosis yield.

A1030

BETA-THALASSEMIA AND ACUTE LYMPHOBLASTIC LEUKEMIA IN A MEXICAN CHILD. Palomo M, Zapata M, Juárez L, Córdova L. Hospital Infantil de México Federico Gómez, México City, Distrito Federal, México.

Introduction: Beta thalassemia is a hemoglobinopathy caused by decreased synthesis of globin chains. The severity of the deficit depends on the levels of gene expression. BT is associated with alterations in the immune system that predispose to frequent infections and hematologic malignancies. **Objective:** Case Report. **Material and Methods:** Descriptive. **Results:** The patient was an 8-month-old male. He had no known history of European ancestry. At 9 months of age he developed anemia and jaundice. He received iron and folic acid, without

improvement in his symptoms. When he was one year old, he received a transfusion of packed red blood cells. He was referred to due to increased abdominal girth. Upon physical examination, he was noted to have cervical lymphadenopathy, splenomegaly extending to the lower left quadrant and hepatomegaly crossing the umbilicus. The patient's initial complete blood count (CBC) showed the following values: Hb 7.9 g/dL, hematocrit 25.8%, MCV 88.6 fl, MHC 27.1, MCHC 30.6 and platelet count of 91.000/mm³. The patient's blood type was O positive. His reticulocyte count was 23%, HbF was 2.58% and HbA₂ was 8.87%. Treatment was started with folic acid, ascorbic acid and transfusion therapy. Red blood cell (RBC) transfusions (10 – 15 mL/kg/week) were required for 2 years. Results of the iron profile was: iron, 70 ?g/dL; iron binding capacity, 192 ?g/dL; free iron, 122 ?g/dL; saturation, 36.4% and ferritin, 1900 mg/mL. Six months later, chelation with deferoxamine was started at 40 mg/kg/dose. When the patient was 4 years old, a splenectomy was performed for massive splenomegaly and frequent transfusion requirements. A month later, the patient presented with diarrhea and meningitis. He was treated with antibiotics, and phenytoin therapy was prescribed (6.2mg/kg/day) for 2 years. At the age of 5 years and 8 months, the patient presented with an infiltrative syndrome and hepatomegaly. Blood count performed at that time showed : Hb, 12.8g/dL; leukocytes, 90,500/mm³ and platelets, 263,000/mm³. ALL-L1 with a T immunophenotype and without central nervous system (CNS) involvement was diagnosed. The patient then developed tumor lysis syndrome, which was treated. He was treated according to our institutional protocol for high-risk ALL, including CNS radiotherapy (18 Gy). He completed 36 months of treatment. He has been in remission for 58 months. **Conclusions:** Immunodeficiency in patients with BT is due to functional and quantitative defects in T- and B-lymphocytes, immunoglobulin levels and chemotaxis. Iron overload reduces phagocytosis and regulates the expression of surface markers on T cells, thus affecting cellular immune function.



A1037

INCIDENCE OF PROGNOSTIC FACTORS IN MEXICAN PEDIATRIC PATIENTS WITH ACUTE LYMPHOBLASTIC LEUKEMIA. A SINGLE INSTITUTION EXPERIENCE. Perez-Rivera E*, Gutierrez-Castellanos S**, Cruz-Borja* P, Chavez-Gallegos S*. *Department of Hematology-Oncology, Hospital Infantil de Morelia Eva Samano de Lopez Mateos, Morelia, Michoacan, Mexico. **Facultad de Ciencias Medicas y Biologicas Ignacio Chavez, Universidad Michoacana de San Nicolas de Hidalgo, Morelia, Michoacan, Mexico.

Introduction: Continuing research on the clinical and biological aspects of Acute Lymphoblastic Leukemia (ALL) has identified numerous features with prognostic potential. The identification of such factors has become essential in the design and analysis of modern therapeutic trials. In underdeveloped countries such as Mexico, there is little information about these factors, for this reason this research was conducted. **Objectives:** Identify the incidence of prognostic factors in Mexican pediatric patients with Acute Lymphoblastic Leukemia. **Material and Methods:** A retrospective analysis was conducted in 91 patients with newly diagnosed ALL from January 1, 2009, through December 20, 2011. Marrow samples were collected before treatment. Diagnosis was performed using cytomorphology and cytochemistry when 25% or more blast cells were present in the bone marrow. Flow cytometric immunophenotyping and DNA content were performed. The initial leukocyte count, characteristics of the cerebrospinal fluid and determination of minimal residual disease were performed. Final analysis was performed only in patients who had at least six of the variables considered. **Results:** 79 eligible ALL patients with a media age of 7 years (range 9 months to 15 years) were analyzed. Gender: 44 (55.7%) males, 35 (44.3%) females. Age at diagnosis: 1 year to <10 years, 55 (69.2%), <1 year 1 (1.2%), 10 or more years 23 (13.8%). White blood cell count at diagnosis: <50,000/ul 62 (86.1%), >50,000/ul 10 (13.8%), unknown 7. Central Nervous System disease: positive 4 (5.7%), negative 66 (94.3%), unknown 9. Prednisone response: good response 50 (74.6%), bad response 17 (25.3%), unknown 12. DNA Index: <1, 1 (1.3%), 1, 51 (64.5%), 1.01-1.15, 7 (8.8%) and >1.16, 20 (25.3%). Immunophenotype: precursor B-cell ALL 73 (97.3%), T-cell ALL 2 (2.6%), unknown 4. CD10-positive precursor B-cell ALL 66 (88%), CD10-negative precursor B-cell ALL 9 (12%), unknown 4. Minimal residual disease: <0.01% 57 (86.3%), >0.01- <1 6 (8.9%), >1 4 (5.9%). See table 1. **Conclusions:** Our group concluded that our population presents adverse prognostic factors more frequently than reported internationally. Age equal to or greater than 10 years and the poor response to steroid are more frequent. Minimal residual disease is the most important prognostic factor and the response of our patients was similar to that reported by international groups.

A1045

HIGHLY SENSITIVE MONITORING OF MINIMAL RESIDUAL DISEASE IN FLT3-ITD-POSITIVE ACUTE MYELOID

LEUKEMIA PATIENTS WITH A MUTATION SPECIFIC QUANTITATIVE-PCR METHOD. Schiller J, Praulich I, Hallek M, Kreuzer KA. Department I of Internal Medicine, University at Cologne, Cologne, Germany.

Introduction: Minimal residual disease (MRD) monitoring in patients with acute myeloid leukemia (AML) can predict relapse clearly in advance and therefore allows early therapeutic intervention. Moreover, recent studies have highlighted the significance of personalized treatment on the basis of MRD status for improving outcome in AML. The FLT3 internal tandem duplication (FLT3-ITD) occurs in 15-35% of all AML. Clinically, FLT3-ITDs have been strongly associated with poor outcome. However, due to the high sequence variability of individual FLT3-ITD commonly a universal PCR approach is applied which has a low sensitivity (approx. 1: 5x10²). **Objective:** To develop a novel cDNA-based, highly sensitive quantitative real-time reverse transcription polymerase chain reaction (qRT-PCR) assay for the detection of the FLT3-ITD mutation level. **Material and Methods:** On the basis of individual FLT3-ITD, mutation-specific primers were designed. The expression of FLT3-ITD was determined using complementary DNA samples at different points in time diagnosis and subsequent treatment. **Results:** From a total of 394 newly diagnosed AML patients 55 (14%) were FLT3/ITD positive. Retrospectively we analyzed ITD mutation of FLT3 in 39 available cases. The length of ITD ranged from 3 to 144 base pairs (median 46). Nine patients had extra insertions of 2-38 base pairs between two repeats. For the FLT3-ITD quantification we developed patient-specific qRT-PCR for 29 individuals with mutation-specific forward primers and studied 83 peripheral blood and 61 bone marrow samples. Three cases with WT1, fifteen with NPM1, three with MLL-PTD and five with PML/RARa fusion genes expression levels were compared with FLT3-ITD expression levels. 26 of 29 assays (90%) were highly specific (1:104 - 1:105) and yielded similar results when compared to other high sensitive assays for molecular markers like NPM1 or PML/RARa. In three cases (10%) a co-amplification of the wild-type could not be avoided resulting in lower sensitivity (1:103). We could show that FLT3-ITD positivity reliably predicted relapse up to 10 months in advance. 92% patients, who achieved FLT3-ITD negativity with our assay, did not relapse. Furthermore we compared paired PB and BM samples at diagnosis and after induction therapy in 5 cases. The difference in FLT3-ITD expression were not statistically significant (p=0.8) which is in line with recent studies. **Conclusions:** We conclude that highly sensitive detection of individual FLT3-ITD poses equal prognostic power in AML like established molecular MRD markers. Using this approach MRD guided treatment decisions appear to be justified and should be incorporated in future studies.

A1066

PULMONARY INFILTRATES IN NEUTROPENIC PATIENTS- OUTCOME AND PROGNOSTIC FACTORS. Prem S*, Kumar R**, Mahapatra M***, Saxena R***. *Regional Cancer Centre,

Trivandrum, Kerala, India; **Cancer Care Manitoba, Winnipeg, Canada; ***All India Institute of Medical Sciences, New Delhi, India.

Introduction: Pneumonias represent the most unfavorable infectious complication occurring during the course of neutropenia in patients with acute leukemia on chemotherapy and are associated with high mortality rates. **Objective:** The aim of this study was to determine the predictors of mortality in neutropenic patients with fever and pulmonary infiltrates/pneumonia. **Material and Methods:** Patients of AML or ALL on intensive chemotherapy protocols were included in this study. 25 variables were studied in prognostic analysis. For purpose of analysis, the following 4 groups of prognostic variables were assessed. 1. Parameters associated with the prognosis of the underlying malignancy and clinical performance of the patient at the time of hospital admission, eg. age, primary or relapsed disease; co-morbidity; ECOG score. 2. Parameters qualifying the circumstances prior to the development of pulmonary infiltrates, eg., type of chemotherapeutic regimen, number of days until development of neutropenia, fever and pulmonary infiltrates, pretreatment with antibiotics or antimycotics, prior steroid medication. 3. Parameters reflecting the severity of illness—vital signs, laboratory parameters; radiographic parameters (type of infiltrate, lung areas involved); microbiological positivity. 4. Evolutionary parameters during antimicrobial treatment—eg., neutrophil count at treatment end point and outcome of treatment (complete remission/failed complete remission). Invasive Pulmonary Aspergillosis (IPA) was classified according to EORTC case definitions. Statistical analysis. Differences between survivors and non-survivors were assessed using the chi-square test for categorical variables and Fisher's exact test in case of small expected frequencies. A multivariate logistic regression analysis was done on a model incorporating variables with $p < 0.05$ on univariate analysis. **Results:** 80 febrile neutropenic episodes with pneumonia were studied. The overall mortality in our patients was 43.75%. Mortality in the IPA group was 43.8% and that in control group was 33.3% ($p=0.04$). Response to antibiotic therapy was seen in 12.5% of febrile patients, response to anti-fungal therapy in 40% and to anti-tubercular therapy in 2.5%. In our study survivors and non-survivors were not significantly different with regards to steroid or ATG pretreatment, performance status, prophylactic medications or co-morbid conditions. Evolutionary parameters like presence of organ dysfunction (hypotension, jaundice or renal failure), failure to attain complete remission with chemotherapy and $ANC < 1000/mm^3$ at treatment end-point correlated significantly with outcome on univariate analysis. In a multivariate model these 3 factors were independently associated with poor outcome. **Conclusions:** We found that independent predictors of mortality in patients of febrile neutropenia with pneumonia were failure to attain complete remission at treatment end point, organ dysfunction and the presence of invasive pulmonary aspergillosis.

A1074

EFFECT OF THE PREDNISONE WINDOW RESPONSE ON THE EVENT-FREE SURVIVAL IN CHILDREN WITH LYMPHOBLASTIC ACUTE LEUKAEMIA. Jiménez-Hernández E, Jaimes-Reyes EZ, Arellano-Galindo J, Dueñas-González MT, Berges-García A. Bekker-Méndez C, Espinoza-Hernández L, Franco-Ornelas S, Pérez-Casillas R, Sánchez-Jara B, Tiznado-García HM, Balam-Canún AM, Núñez-Villegas NN, Mejía Aranguré JM. Hematología Pediátrica UMAE, Centro Médico Nacional "La Raza" IMSS; Laboratorio De Investigación Hospital Infantil de México "Federico Gómez" SS; División de Laboratorios de Vigilancia E Investigación Epidemiológica UMAE, Centro Médico Nacional "La Raza" IMSS; Unidad de Investigación en Epidemiología Clínica, UMAE Hospital de Pediatría Centro Médico Nacional "Siglo XXI", IMSS. Ciudad de México.

Introduction: Prednisone Response Is the Strongest Predictor of Treatment Outcome in Infant Acute Lymphoblastic Leukemia.

Objective: To know the effect of prednisone window response on the event-free survival (EFS) in children with acute lymphoblastic leukaemia (ALL). **Material and Methods:** A retrospective and analytical cohort study from August 2006 to December 2010. Patients less than 15 years old, with ALL, who had received prednisone window, with an intrathecal chemotherapy of metotrexate followed of prednisone 60 mg/m²/day for 7 days as prophase of treatment to the remission induction (protocol of chemotherapy CMR 2002) developed in the pediatric haematology department at the UMAE Centro Médico Nacional la Raza IMSS. The response was considered as follows: good when less than 1×10^3 blasts/L and poor when more than 1000 blast were found on 7 days in peripheral blood. Statistical analysis. All cases were taken consecutively. A value of $P .05$ was considered significative. The effect of RWP was evaluated by the Kaplan-Meier method and the groups were compared with a log-rank test using SPSS 16 software. **Results:** A total of 300 patients that met inclusion criteria, median of 7 years old, most of the from the masculine sex (55 percentage) and a median of leukocytes to the diagnosis $10,7 \times 10^9/L$ (min 720 and max $939 \times 10^9/L$), hyperleukocytosis 15 percentage, standard risk (SR) 48 percentage, high risk (HR) 52 percentage. On the total of population, the EFS was of 74.7 percentage. The EFS for the group of good responder of SR was of 65 percentage, and high risk 45 percentage ($P.043$) paradoxical results. According to the Cox regression. Be of HR with more than 1000 blasts to the diagnosis the risk of died of was of 1.7 times compared with the responders on 7 day, the risk of died was of 1.8 times, be of HR with more than 1000 blasts on 7 day the risk of died was protector (0.793). The group with more risk of relapse adjusted with the count of leukocytes was found between $20-50 \times 10^9/L$ classified as SR in our population. **Conclusions:** The EFS of our population was of 74.7 percentage lower than the reported in developed countries and higher than reported in undeveloped countries, The leukocytes count at the diagnosis was a main confuser variable. The prednisone window response

have influence on the EFS but is depending on the risk assigned to the diagnose, which suggest that is necessary the classification of three group of risk.

A1077

IMPACT OF DURATION OF BREAST-FEEDING AND THE RISK OF ACUTE LYMPHOBLASTIC LEUKEMIA IN MEXICAN CHILDREN. Nuño-Vázquez LM, Ramírez-Cázares C, Villarreal-Martínez L, Hernández-Juarez I, Ríos-Solis J, Mancías-Guerra MC, Gómez-Almaguer D, González-Llano O. Servicio de Hematología, Hospital Universitario "Dr. José Eleuterio González", Universidad Autónoma de Nuevo León, Monterrey, Nuevo León, México.

Introduction: Breast-feeding has been studied for a possible protective rol in the risk of childhood leukemia, the proposed hypothesis arise from the one that suggests that breast-feeding protects from infections. This association has mixed reports. There are no reports of this relationship in our population.

Objectives: Evaluate the rol of breast-feeding and its duration in children with acute lymphoblastic leukemia in a Mexican population. **Material and Methods:** This was a retrospective case-control study comprising 50 patients with acute lymphoblastic leukemia (ALL), aged 1-16, as well of 55 controls matched for age and sex. The information was collected by interviewing the children's parent, during the appointment with pediatric hematologist and/or pediatrician. **Results:** Patients with ALL had less breast-feeding versus controls (P=0.013). Breast-feeding for more than 6 months of duration was higher among controls compared with leukemia patients (P=0.001). **Conclusions:** Our findings suggest that breast-feeding lasting longer than 6 months may have a protective effect against childhood acute lymphoblastic leukemia.

A1078

WILMS' TUMOR 1 PROTEIN EXPRESSION STATUS IN AML CELLS AND POSSIBILITIES FOR PHARMACOLOGICAL MODULATION. Zaharieva M*, Dobrev N**, Spassov B**, Balatzenko G**, Guenova M**, Berger MR***, Konstantinov S*. *Lab for Experimental Chemotherapy, Dept. of Pharmacology, Pharmacotherapy and Toxicology, Faculty of Pharmacy, Medical University of Sofia, Bulgaria **National Specialized Hospital for Active Therapy of Hematological Diseases, Center of Excellence Translational Research in Hematology, Sofia, Bulgaria ***German Cancer Research Center, Unit of Toxicology and Chemotherapy, Heidelberg, Germany.

Introduction: Erufosine is an ether lipid analogue with strong antineoplastic activity and lack of bone marrow cytotoxicity. It interferes with signal transduction and induces apoptosis in leukemic cells but its exact mode of action remains poorly understood. Wilms' tumor 1 (Wt1) protein is expressed in 50-60% of AML patients and high expression of Wt1 gene confers adverse prognosis in these patients. **Objective:** The estimation

of Wt1 protein expression status in AML cells as well as its modulation by erufosine were the focus of the study. **Material and Methods:** In vitro experiments were performed on 8 AML-cell lines (Eol-1, Nomo-1, MV-4-11, NB-4, U-937, KG-1, HL-60 and HL-60/Dox). Cytotoxicity of erufosine was measured by the colorimetric MTT-assay and IC50 values were calculated. Changes in the MiRNAs expression were investigated using a multiplex miRNA-assay (Luminex). Protein expression levels of Wt1, Bcl-2 and Lamin B were determined by Western blotting. Additionally, bone marrow samples of 30 AML patients were analyzed for Wt1 protein expression. **Results:** Erufosine was found to be cytotoxic in all cell lines except HL-60/Dox and the most sensitive cell line was found to be NB-4. The chemical substance altered the levels of tumor suppressor and oncogene miRNAs in NOMO-1 and U-937 cells, inhibited WT1 expression in U-937 cells and induced apoptosis in both cell lines as evidenced by miRNA and signaling protein changes. Wt1 protein expression was detected in 53.3% (16/30) and high levels of the marker were observed in 30% (9/30) of AML patients. High Wt1 protein expression correlated with significantly lower rate of complete remission (P=0.023). **Conclusions:** AML patients with high Wt1 expression have poor prognostic and therapeutic outcomes. Dynamic changes in WT1 expression during therapy occur and can influence the remission induction. Erufosine is an effective antileukemic agent in vitro that may have special therapeutic relevance since it leads to Wt1 protein inhibition. **Acknowledgements:** This work was supported by the National Science Found grant No. CVP/01 0119-DO-02-35/09 and the Alexander von Humboldt Foundation.

A1079

POLYMORPHISMS C677T AND A1298C OF METHYLENE-TETRAHYDROFOLATE REDUCTASE (MTHFR) AND RISK OF ACUTE LYMPHOBLASTIC LEUKEMIA IN MEXICAN CHILDREN. Villarreal-Martínez L*, Martínez-Garza LE**, Mancías-Guerra MC*, Hernández-Juárez I*, Nuño-Vázquez LM*, Ríos-Solis JE*, Zamudio-Osuna MJ**, Aguirre-Rodríguez AA**, Gomez-Almaguer D*, González-Llano O*. *Servicio de Hematología Hospital Universitario "Dr. José Eleuterio González" Universidad Autónoma de Nuevo León; **Departamento de Genética, Hospital Universitario "Dr. José Eleuterio González" Universidad Autónoma de Nuevo León.

Introduction: Leukemia is the most common cancer in pediatric age. Its etiology has a combination of genetic susceptibility and adverse environmental exposure during early development, fetal life and infancy. Folic acid deficiency affects nucleic acid synthesis and DNA repair resulting in chromosomal damage which is related to tumorigenesis process. Polymorphisms of the methylenetetrahydrofolate reductase (MTHFR) gene C677T and A1298C have been associated with modification in cancer risk including leukemia. We conducted a study to determine the frequencies of these polymorphisms in our population. **Objective:** Establish allele frequencies of the C677T and A1298C poly-

morphisms of the MTHFR gene in a pediatric ALL population and compare them with healthy controls to determine if there is an association with leukemia risk. **Material and Methods:** We performed a case-control study that involved 50 patients aged 1-16 with ALL in a Mexican population and compare them with historical controls. The polymorphisms genotyping was assessed using real time polymerase-chain reaction. **Results:** The frequencies of MTHFR polymorphic variants in patients versus controls were: 46% vs 53.7% for CT (OR 0.82), 24% vs 18 % (OR 1.25) for TT. For the A1298: 20% vs 28% for AC (OR 0.63) and 2% vs 4% (OR 0.44) for CC. No significant difference was found in allele frequencies between ALL patients and healthy controls. **Conclusions:** This report provides information about the allelic polymorphisms C677T and A1298C in pediatric ALL patients of a population of northern Mexico. We didn't find any association between MTHFR allelic frequencies and leukemia risk.

A1084

FREQUENCY OF THE MAIN FUSION TRANSCRIPTS IN ACUTE LYMPHOBLASTIC LEUKEMIA: A STUDY OF 148 ADULT BULGARIAN PATIENTS. Balatzenko G***, Guenova M***, Ganeva P***, Jankova T*, Varbanova V*, Spassov B***, Mihailov G*, Konstantinov S****, Stoimenov A*, Toshkov S*. *National Specialized Hospital for Active Treatment of Hematological Diseases – Sofia, Bulgaria; **Center of Excellence for Translational Research in Hematology – Sofia, Bulgaria; ***Lab for Experimental Chemotherapy, Dept. of Pharmacology, Pharmacotherapy and Toxicology, Faculty of Pharmacy, Medical University of Sofia, Bulgaria.

Introduction: Genetic abnormalities are one of the most important prognostic factors in acute lymphoblastic leukemia (ALL) patients. Several studies support the idea that the frequency of some chromosome translocations and the respective fusion transcripts may vary among the different countries. To better understand these “geographic-associated” differences it is important to compare the frequency of genetic abnormalities in individual countries. **Objective:** To establish the prevalence of the main fusion transcripts in adult Bulgarian ALL patients. **Material and Methods:** A total 132 (58 females; 74 males) newly diagnosed ALL Caucasian patients with a mean age of 40.0 ± 16.8 years were studied. The group comprised 101 patients with B-cell ALL (B-ALL) and 31 cases of T-cell ALL (T-ALL). All patients were screened by reverse transcription polymerase chain reaction (RT-PCR) (BIOMED-1 Concerted Action) for the recurrent fusion transcripts: BCR-ABL [both Major (M) and minor (m) forms]; ETV6-RUNX1 (TEL-AML1); TCF3-PBX1 (E2A-PBX1); MLL-AF4; and SIL-TAL1 rearrangements. **Results:** Overall, a positive result for the presence of fusion transcripts was detected in 43 patients, including 40 positive cases of patients with B-ALL, and 3 SIL-TAL1-positive cases out of 31 (9.7%) patients with T-ALL (2.3% of all cases). The most frequent molecular abnormality in B-ALL patients was the BCR-ABL rearrangement, found in

27 (26.7%) patients (b2a2 n=4; b3a2 n=6; e1a2 n=17). The frequency of the remaining markers was as follows: 7/101 (5.9%) MLL-AF4(+) (4.5% of all cases); 4/101 (4.0%) TCF3-PBX1(+) (3.0% of all cases) and 1/101 (1%) ETV6-RUNX1(+) (0.8% of all cases). BCR-ABL-positive patients (20.4% of all cases) were clearly older compared to the negative group (51.6 ± 17.2 vs. 34.4 ± 14.9 years; $p=0.00$). No similar correlation was found in regard to the remaining abnormalities. **Conclusions:** The estimated prevalence of the main fusion transcripts in our study does not differ significantly from the generally reported values. Therefore it is unlikely that the local genetic background is associated with a specific molecular mechanism of leukemogenesis in adult ALL. **Acknowledgements:** Studies were supported by the National Science Fund, Ministry of Education, Science and Youth.

A1086

FLOW CYTOMETRIC DETECTION OF RESIDUAL ACUTE LEUKEMIA. NOSOGRAPHIC PERFORMANCE IN A SINGLE INSTITUTION. Perez-Romano B, Coutiño R, Ruiz-Argüelles GJ, Ruiz-Argüelles A. Laboratorios Clínicos de Puebla and Centro de Hematología y Medicina Interna, Puebla, Mexico.

Introduction: Several technical approaches have been proposed and used to detect residual disease after completion of treatment in patients with acute leukemia. Inasmuch as crucial decisions are taken upon the results of such studies, the nosographic performance of each test or combination of tests must be known. Flow cytometry is one of the most popular tools to detect residual disease in hematological malignancies, however, its nosographic performance may be different in different samples, different types of leukemia and different laboratories. **Objective:** To determine the nosographic performance of flow cytometry for the detection of residual disease. **Material and Methods:** A total of 905 flow cytometric studies to detect residual acute leukemia were performed at Laboratorios Clínicos de Puebla from 2002 to 2011. The usual study comprised 7 antibodies in multiparameter flow-cytometry, that were selected according to the diagnostic immunophenotype of each case. For this particular study, a positive or a negative result was considered as true positive or true negative, if the result was consistent in at least one consecutive study. Inconsistent results in consecutive studies were tagged as false positive or false negative. A result switching from positive to negative is not necessarily an equivocal one, since it may reflect medical intervention. **Results:** The vast majority of the studies were performed in peripheral blood (744), followed by bone marrow (137) and cerebrospinal fluid (24). The proportions of positive and negative results were similar in all these samples. The type of leukemia was B cell precursors in 443 cases, T cell leukemia in 137 and myeloid in the remaining 325 cases. Overall sensitivity and specificity of the method were 74.34 % and 78.40 % respectively. The predictive value of a positive result was 79.5%, while that of a negative result

was 73.0%. The statistical significance of these results is solid (Chi-square 197, $p < 0.0001$). **Conclusions:** In our experience, a positive result of the flow cytometric detection of residual acute leukemia is more reliable than a negative result, mainly if the decision to cease treatment is to be taken. The most reasonable explanation is that flow cytometric immunophenotype is incapable to detect small numbers of malignant cells that suffice for the disease to relapse. For practical purposes, the recommendation is to further treat patients with a positive flow cytometric results, but not to withdraw treatment in a patient with a negative flow cytometric result until another analytical approach proves the absence of disease.

A1095

FMS-LIKE TYROSINE KINASE 3 (FLT3) MUTATIONS IN MEXICAN PATIENTS WITH DE NOVO ACUTE MYELOID LEUKEMIA. Ruiz-Argüelles GJ*.,**** Garces-Eisele*.,**** J, Alarcón-Urdaneta C*.,****, Julia Lutz-Presno J*.,**, Ruiz-Delgado GJ*.,****. *Centro de Hematología y Medicina Interna de Puebla; **Laboratorios Clínicos de Puebla. ***Universidad Popular Autónoma del Estado de Puebla; ****Benemérita Universidad Autónoma de Puebla.

Introduction: Constitutive activation of the FMS-like tyrosine kinase 3 (FLT3) receptor tyrosine kinase by internal tandem duplication (ITD) as well as activation loop mutation D835X has been researched in patients with de novo acute myeloid leukemia (AML). **Objective:** To assess the prevalence of FLT3 mutations in Mexican patients with AML and analyze its prognostic significance. **Material and Methods:** A total of 31 consecutive patients with AML in a single institution were prospectively studied. Genomic DNA was extracted from peripheral blood and polymerase chain reaction was performed. GeneScan was used to analyze the mutant to wild-type ratio. The sequencing of mutated genes was performed to confirm the mutation types and exclude false positives. **Results:** A total of 4 / 31 cases (13%) were associated with mutations, ITD in 3 cases and activation loop mutation D835X in one. The overall survival (OS) of AML patients without FLT3 mutations was 67%, whereas the OS of patients with FLT3 mutations was 25%, the difference being statistically significant. All patients (3/3) with FLT3 ITD have died, whereas one with the D835X is alive. **Conclusions:** In this small series of Mexican patients with AML we have found a lower prevalence of FLT3 mutations when compared to Caucasian populations, where it has been described in around 20%. Moreover, the adverse prognostic value of the mutation has been confirmed.

A1096

BLAST CELL DNA CONTENT IN CHILDHOOD ACUTE LYMPHOBLASTIC LEUKEMIA. DETECTION OF CHILDREN WITH LOW PROBABILITY OF RELAPSE. Gutiérrez-Castellanos S*, Pérez-Rivera E**, Cruz-Borja P**, Chávez-Gallegos

S**, Nava-Miranda J**. *Laboratorio de Citopatología Molecular, División de Estudios de Posgrado, Facultad de Ciencias Médicas y Biológicas Ignacio Chávez, Universidad Michoacana de San Nicolás de Hidalgo; Laboratorio Clínico, Unidad de Medicina Familiar con UMAA No. 75, Instituto Mexicano del Seguro Social, Morelia, Michoacán, Mexico. **Department of Hematology-Oncology, Hospital Infantil de Morelia Eva Sámano de López Mateos, Morelia, Michoacán, Mexico.

Introduction: Acute lymphoblastic leukemia (ALL) is the most common malignancy of childhood and has served as a model system for clinical and basic studies. Certain features of leukemic cells, such as the immunophenotype, central nervous system disease (CNS), age, leukocyte count (WBC), cytogenetic abnormalities and DNA index (DI), are considered important prognostic factors. DNA index greater than 1.16, occurs in 20% to 25% of cases of precursor B-cell ALL and has been associated with a better prognosis. Flow cytometric analysis of cellular DNA content is a powerful tool for assessing prognostic variables as the DNA ploidy of malignant G0/G1 cells and the percentage of S-phase cells. **Objective:** The aim of this work was to determine the percentage of children with high DNA index, and its relation with prognostic factors. **Material and Methods:** Analysis was conducted in 79 eligible patients treated at our institution with newly diagnosed ALL from March 1, 2009, through December 20, 2011. Marrow samples were collected before treatment from each child. DNA content and the percentage of S-phase cells were analyzed by flow cytometry. We performed univariate analysis of sex, immunophenotype, NCI risk criteria, and age, WBC and CNS status at diagnosis. **Results:** 79 ALL patients with a median age of 7 years (range 9 months to 15 years) including 44 (55.7%) males, and 35 (44.3%) females were analyzed. DNA index = <1, one case (1.3%); DI=1, 51 (64.6%), DI=1.01-1.15, 7 (8.9%) and DI≥1.16, 20 (25.3%). Interesting, all patients with DI > 1.16 had WBC < 50 x 10⁹/L, as well as CNS status, patients with DI > 1.16 expressed level 1, while DI < 1.16 expressed level 2 or 3. (Figure 1). **Conclusions:** We identify a group of 20 patients with DI > 1.16 who have features as WBC and CNS status with a low probability of relapse. If they remain disease-free after longer follow-up, it may be advisable to treat with less intensive, hence less toxic, chemotherapy. To our knowledge there are no studies of this type in Mexico and it is necessary to continue researching

Table 1. DNA index and Percentage of S-Phase Cells

DNA index	n(%)
<1.0	1 (12%)
1.0	51 (64.5%)
1.01-1.15	7 (8.8%)
≥1.16	20 (25.3%)
Percentage of S-Phase Cells	
≤6.8	39 (49.3%)
>6.8	40 (50.6%)

in our population because of results of treatment is different from the international ones.

A1098

MIXED PHENOTYPE ACUTE LEUKEMIA. Campos-Cabrera G*, Campos-Cabrera S*, Campos-Cabrera V**, Mora-Torres M***, Campos-Villagomez JL*. * Departamento de Hematología de Laboratorios Fatima de Michoacan, Morelia, Mexico; ** Departamento de Patología Clínica de Laboratorios Fatima de Michoacan, Morelia, Mexico; *** Servicio de Hematología del Hospital Infantil de Morelia “Eva Sámano de López Mateos”, Morelia, Mexico.

Introduction: Acute leukemia with a mixed phenotype is a rare disease and comprises 2–5% of all acute leukemias. These disorders have been known historically by a variety of names, such as mixed lineage leukemia, bilineal leukemia and biphenotypic leukemia, and the criteria for diagnosis have often been arbitrary. The scoring criteria proposed by the European Group for the Immunological Characterization of Leukemias (EGIL) represented a major attempt to define this disorder. **Objective:** We present a case of mixed phenotype acute leukemia. **Material and Methods:** The diagnosis and classification of acute leukemia relies on a multidisciplinary approach including morphology, immunophenotyping, karyotype analysis and more specific molecular genetic analysis when available. **Results:** Female of 10 years old with a history of lower back pain, low intensity fever at night, and anemia was sent for evaluation. Among other lab tests; bone marrow aspiration with flow cytometry and karyotype samples was performed. The morphology showed acute lymphoblastic leukemia with L1 morphology (Figure 1). Flow cytometry report mixed acute leukemia (Figure 2): No lineage associated markers: CD45+, CD34+, CD38+, HLA-DR+; B cell markers: CD10+, CD19+, CD20+, CD22+, CD24+; Myeloid markers: CD13+, CD33+, CD117+; Other markers were negative Karyotype (Figure 3): 46,XX. **Conclusions:** Catovsky’s scoring criteria were based on the number and different weights given to diagnostic markers according to their generally accepted specificity at that time (Table 1 - Ann Hematol 1991; 62: 16–21). The EGIL later proposed an immunological classification and characterization of acute leukemias, which also included a definition for MPAL (Table 2 - Leukemia 1995; 9: 1783–1786). There is no single chromosomal aberrancy that is uniquely associated with MPAL, 68% had a clonal abnormality whereas 32% had a normal karyotype, the most common include rearrangement of 11q23, the site of MLL, followed by the Philadelphia chromosome; additional abnormalities included deletion of 6q, 5q and 12p (Leukemia 2010; 24: 1844–1851). The prevalence and clinical significance of MPAL using the 2008 WHO definition remains to be determined. Treatment for MPAL is not well established and the cytogenetic and molecular genetic changes have emerged to be of greatest biological importance. If that is true, the controversy on how to diagnose and treat these disorders will shift away from the immunophenotype and towards more

Table 1. Catovsky et al scoring system for biphenotypic acute leukemia

Points	B lineage	T lineage	Myeloid lineage
2	cCD22 cμ chain	cCD3	MPO
1	CD10 CD19 CD24	CD2 CD5 TCR rearrangement (β or δ chain)	CD33 CD12 CD14 AML morphology or cytochemistry (other than MPO)
0.5	TdT IgH rearrangement		CD11b CD11c CD15

Abbreviations: AML, acute myeloid leukemia; MPO, myeloperoxidase; TCR, T-cell receptor; TdT, terminal deoxynucleotidyl transferase. ≥2 points are required to assign a lineage.

specific molecular genetic markers. The program used to upload images do not support JPG and related files.

A1099

ACUTE MYELOID LEUKEMIA IN ADULTS OVER 60 YEARS: INTENSIVE CHEMOTHERAPY INCREASE OVERALL SURVIVAL? Lopez-Hernandez MA, Mena-Zepeda V, Alvarez-Vera JL, Alvarado-Ibarra M. Hematology Department of the National Medical Center “20 Noviembre”.

Introduction: The acute myeloid leukemia (AML) in elderly patients of >60 years old, the expected frequency of remission and overall survival is lower than in patients under. Increased morbidity and mortality is related to age, leukocytosis, initial infection and comorbidity. It is generally accepted that the initial remission and overall survival are better with use of intensive chemotherapy. **Objective:** Compare the overall survival (OS), in patients over 60 years with diagnosis of AML, treated with intensive chemotherapy (IC) or non intensive chemotherapy (NIC), attended during the past 11 years, in our Department. **Material and Methods:** Longitudinal study, non-randomized, comparative and retrospective. We included patients over 60 years with AML which received IC: induction with cytarabine/daunorubicina (7/3), and two courses of postremissions with cytarabine high doses/etoposide and cytarabine high doses/daunorubicine. Another group was treated with NIC: cytarabine low dose (30 mg/m² sc, ten days of each month), alternating with busulfán (15 mg/day, three days of each month), and mercaptopurine (60 mg/day for a month). Were analyzed: sex, age, co-morbidity, index of Charlson (IC), leukocytes, platelets initial and number of blasts in the blood or marrow; diagnosis was specified with immunophenotype and investigated whether they were second-

ary or associated with myelodysplastic syndrome. We quantified the frequency of complete remissions. **Results:** Sixty and six patients were included. Nine were excluded since they did not receive treatment. Twenty-three were given IC and 34 NIC. The median age was 66 and 77 years ($p = 0.001$). No difference was found ($p > 0.19$) in sex, co-morbidity, IC, account of white blood cells, platelets, blasts, secondary leukemias, associated with myelodysplastic syndrome and with a history of chemotherapy. Twenty achieved remission in the IC and none in NIC. The OS was 28 months for IC and 26 for NIC, with median of 4 months for both ($p = 0.87$). The hospitalization was 46 and 22 days ($p = 0.03$). None of the variables showed influence at the destination ($p > 0.08$). The OS was equal in those who attained remission or not ($p = 0.518$). **Conclusions:** Overall survival is the same using QTI or QTNI with less need for hospitalization in the second.

A1106

ANALYSIS OF THE BIOLOGICAL AND CLINICAL CHARACTERISTIC OF ACUTE LYMPHOBLASTIC LEUKEMIA IN ADULTS. Díaz-Ruiz R*, Garcés-Ruiz OM*, Aguilar-López L*, Iris F García-Rincón IF*, Vega Ruiz A*, Rubio-Jurado B*.*. *Hematology Service, **Clinical Epidemiology Research Unit, UMAE, CMNO, Instituto Mexicano del Seguro Social. Guadalajara, Jalisco. México.

Introduction: The acute lymphoblastic leukemia (ALL) corresponds to 15% of acute leukemia of the adult. The rate of remission is variable of 60-75% and in Mexico a median of global survival of 15 months is considered, with a mortality of 26% in induction. The poor results are related to biological characteristics of high risk, resistance to drugs, low tolerance and complications of the treatment. It is important to identify the related factors to remission and mortality in our population.

Objective: Describe the frequency of clinical features and biochemical markers in patients with ALL. **Material and Methods:** A descriptive study. We included patients who were consecutively included in the period April 2010 to December 2011 at the Hematology Service, Specialties. IMSS CMNO, who identified ALL. **Results:** We included 20 patients, 13 women and 7 men. Age (median, minimum to maximum) 36, 17, 84 yrs. Frequencies of clinical and biological variables: bleeding 43%, fever 20%, lymphadenopathy 52%, organomegaly 19%. Leukocytosis $> 30\,000$ cells / μ L in 11 cases (55%). Lactic deshidrogenasa in 11 cases high (55%). Classification of the FAB: 17 cases with L2. Positive immunophenotype for markers lineage B 22/22 cases. Karyotypic in 2 / 21 cases, one normal case and another with t (4; 11). Treatment of Induction, 14 BMF cases (70%), Hyper-CVAD 3 cases (15%), other schemes are: CFA+VCR; R+VCR; DNR+araC+Dexa. Treatment of Intensification: Hyper-CVAD 10 cases (50%), 2 LARSON, cases. Complete remission in 12 pts. (60%). 3 pts. relapse after 8 months of treatment. 8 pac died., Sequimiento median 4.7 m (1-to 11 months), 4 in remission induction (20%), 2 in relapse and 2 from sepsis in RC. They are

found living in treatment 8 pts; 2 pts. with CNS relapse. Disease free survival is 7 months and overall survival of 9 months.

Conclusions: The ALL is a disease with poor prognosis. In a short follow-up to 40% of these cases remains in remission. Mortality is related to what was reported. Not routinely available cytogenetic study. Is required to generate a risk-adapted treatment according to their initial characteristics.

A1107

HIGH DOSE METHOTREXATE IN PATIENTS WITH ISOLATED CNS RELAPSES OF ALL: A PROTOCOL MODIFICATION. Rios-Solis JE, Nuño-Vazquez LM, Villarreal-Martinez L, Hernandez-Juarez I, Mancias-Guerra MC, Gomez-Almaguer D, Gonzalez-Llano O. Servicio de Hematología, Hospital Universitario "Dr Jose Eleuterio Gonzalez", Universidad Autónoma de Nuevo León, Monterrey, Nuevo León, México.

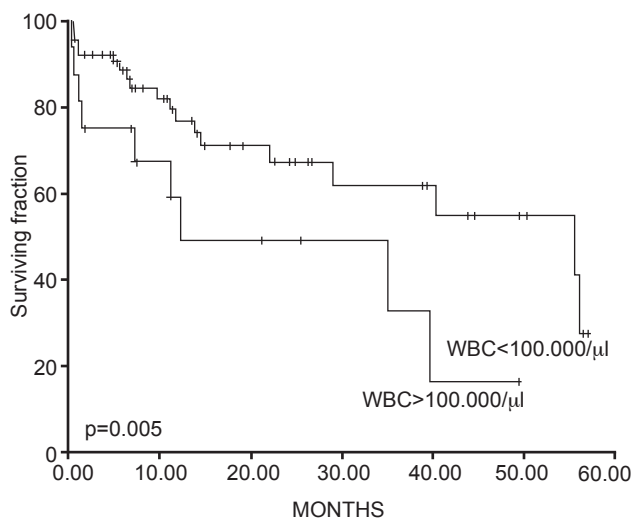
Introduction: Patients that present a late isolated CNS relapse are enrolled into an intensified systemic therapy, including high dose methotrexate, plus craniospinal radiation. Because space limitations in our institution, we performed a modification in dose and time for methotrexate infusion, using a 2 gr/m²/day in 6-hour infusion. **Objective:** Demonstrate that high dose methotrexate in 6-hour infusion, isn't related to an increased risk for acute toxicity. Corroborate serum levels of methotrexate and its relationship with toxicity symptoms. **Material and Method:** ALL patients with isolated late CNS relapse treated in our service, without previous radiation, and a remission duration of =18 months. They were enrolled in COG 94 protocol, and received the 6-hour infusion of 2 gr/m²/day methotrexate. We measured methotrexate serum levels 24 hours from onset of the 6-hour methotrexate infusion, followed by folinic acid rescue at up 24 hours, grading the mucositis through OMS classification, and search for other acute toxicity data. **Results:** Five patients underwent 4 cycles of this modification. Four of them presented mucositis 0-2 (OMS). Only one patient had elevated liver enzymes, and graded 4 mucositis. All methotrexate levels where in therapeutic range. **Conclusions:** We need more patients to make final conclusions, but right now it seems to be a safe way to administrate high dose methotrexate in these patients.

A1114

CHARACTERISTICS AND OUTCOME OF ADULT PATIENTS WITH ACUTE LYMPHOBLASTIC LEUKEMIA TREATED AT A SINGLE INSTITUTION IN MEXICO. Jaime-Pérez JC*, Pinzón-Uresti MA*, Gutiérrez-Aguirre C.H.*, Cantú-Rodríguez O*, Tarín-Arzaga L*, Gómez-Almaguer D*. *Hospital Universitario " Dr. Jose Eleuterio González", Universidad Autónoma de Nuevo León, Monterrey, Nuevo León, México.

Introduction: Acute lymphoblastic leukemia (ALL) in adults is a biologically heterogeneous disease, characterized by lymphoid malignant cell proliferation; differences in clinical behavior,

response to therapy relapse and overall and event-free survival rates exist in different populations. **Objective:** We documented the characteristics in adults with ALL receiving therapy at a single institution in Mexico. **Material and Methods:** Adults with ALL attending the Hematology clinic at a public University Hospital in Northeast Mexico from January 2006 to April 2011 were studied. Clinical files and electronic records were scrutinized. Descriptive analysis was performed, obtaining means and standard deviation, medians and ranges. Overall survival (OS) and event-free survival (EFS) were determined using the Kaplan-Meier method, calculating time, survival, and standard errors with 95% confidence interval (CI); hazard ratios (HR) for death and relapse were estimated by uni- and multivariate Cox regression analysis. **Results:** 86 adults were included, median age was 27 years, 61 (70.9%) were ≤ 40 years, 17 (19.8%) had a WBC = $100 \times 10^9/L$, 52 (60.5%) had high risk and 34 (39.5%) standard risk ALL, OS was 26% and 59%, respectively, whereas EFS was 25% and 60%; 62 (72.1%) achieved complete remission (CR), 20 (23.2%) relapsed at a median of 18 months, 28 (32.6%) died at a median of 8.6 months, 58 (67.4%) are alive at a median of 13.9 months. OS and EFS were 42% and 40% at five years; a leukocyte count $\geq 100 \times 10^9/L$ at diagnosis was identified in uni and multivariate analysis as the only significant factor influencing in these study, $p < 0.05$. **Conclusions:** Is important to report data on the clinical picture, evolution and prognosis. Adults with ALL from northeast Mexico were younger and had an elevated proportion of high-risk characteristics than in other populations. A WBC count $> 100 \times 10^9/L$ was found to be the only significant adverse prognostic factor in B cell cases.



A1120

EVALUATION OF PROPHYLACTIC AND THERAPEUTIC INTRATHECAL CHEMOTHERAPY FOR ADULTS WITH ACUTE LYMPHOBLASTIC LEUKEMIA. Flores-Jiménez JA, Sánchez-Cárdenas M, Saldaña-Vázquez R, Ceballos-López

AA, Martínez-Hernández RA, Cuervo-Sierra J, Pequeño-Luévano MP, Tarín-Arzaga L, Jaime-Pérez JC, Cantú-Rodríguez O, Gutiérrez Aguirre CH, Gómez-Almaguer D. Universidad Autónoma de Nuevo León, Hospital Universitario, Monterrey, N.L., México

Introduction: Central nervous system (CNS) involvement in acute lymphoblastic leukemia (ALL) patients remains a challenge affecting 5-10% at diagnosis. Before the use of CNS prophylaxis the relapse rate was 75% in children and 30% in adults. Besides the use of intrathecal (IT) therapy CNS relapses rates may range from 1-15%. Several strategies have been implemented for the prophylaxis and treatment of CNS disease which include: IT chemotherapy with single drug or multidrug agents, craniospinal radiation therapy and high-dose systemic chemotherapy. **Objective:** The aim of the study was to evaluate the effectiveness of IT chemotherapy in the prevention and treatment of CNS relapse.

Material and Methods: We analyzed medical records ALL adult patients from January 2003 to December 2011. We included patients ≥ 15 years diagnosed with ALL morphologically or by flow cytometry at the University Hospital "Dr. Jose Eleuterio Gonzalez", who had received triple intrathecal chemotherapy during its treatment approximately 15 during the first year. In patients with CNS leukemic infiltration at diagnosis intrathecal chemotherapy was up to 2 times per week. CNS involvement was defined as morphologic evidence of lymphoblasts in CSF. Overall survival was estimated by the Kaplan-Meier method. **Results:** Fifty-five patients were included, the ALL immunophenotype was B-cell in 50 (91%), T-cell 3 (5%) for and B-mature 2 (4%). With regard to gender, 67% were males and 33% females. The median age was 31 years (range 15-78). CNS-involvement at diagnosis was present in 5 (9%) patients; with a median age of 20 years (range 15-76). The median number of IT chemotherapy applications to become CSF-negative was 6 (range 2-7). Seven (13%) out of 55 patients had CNS relapse with a median age of 23 years (range 15-45), 89% showed B-phenotype and 11% T-phenotype. The median number of IT chemotherapy applications to become CSF-negative in relapsing patients was 3 (range 1-7). The median overall survival at 2 years for the 55 patients was 68%. We found no difference in survival between patients with and without CNS-involvement (Figure-1). **Conclusion:** Comparing both groups, those with and without CNS-involvement at diagnosis, we found no statistically significant differences in overall survival and clinical and laboratory characteristics, which may be due to the sample size. We had 13% CNS relapse rate, higher than in other reports; this could be explained by a delay between beginning of symptoms and diagnosis and treatment as well as to lack of adherence to treatment. Prospective larger studies are needed to confirm our findings.

A1125

THE NKG2D RECEPTOR EXPRESSION IN NK CELLS AND MIC-A AND MIC-B LIGANDS EXPRESSION IN LEUKEMIC

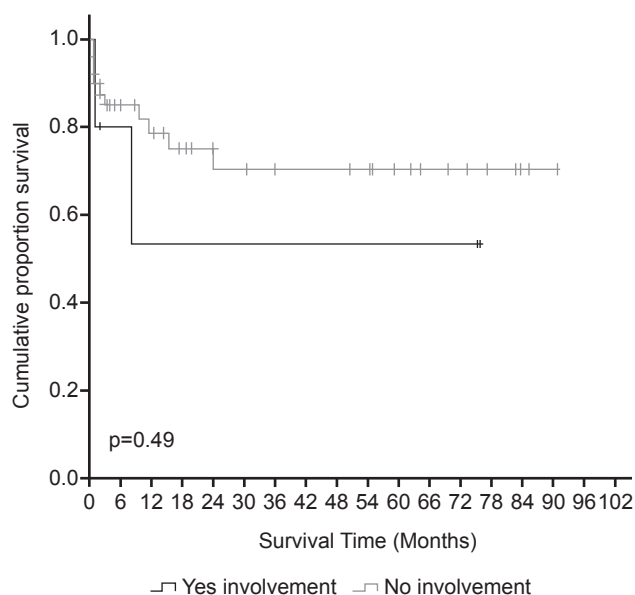


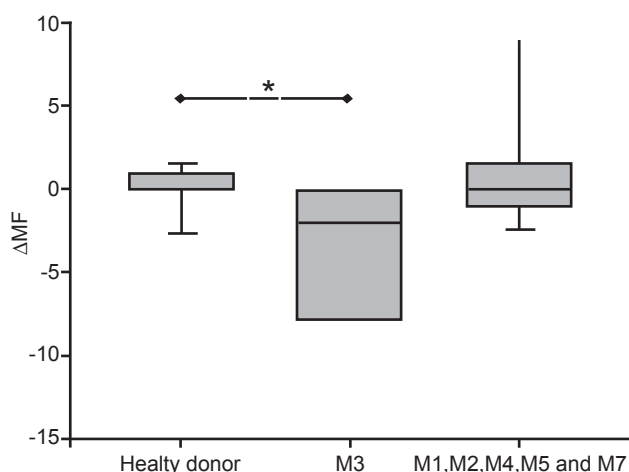
Figure 2. Survival in patients with CNS involvement at diagnosis

BLAST FROM MEXICAN PATIENTS WITH ACUTE MYELOID LEUKEMIA. Sandoval-Borrego D*, Gutiérrez-Hoya A*, López-Santiago R*, Vela-Ojeda J**, Montiel-Cervantes**, Moreno-Lafont M*. *Departamento de Inmunología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, México DF, México; ** Centro Médico Nacional “La Raza”, Instituto Mexicano del Seguro Social, México DF, México.

Introduction: Natural killer (NK) cells are an important part of the organism’s innate immune system. They can destroy virally infected and malignant cells without prior sensitization. These cells recognize through their activation receptors, such as NKG2D, abnormal or damaged cells expressing the ligands MICA and MICB. **Objective:** In this project we searched for the expression of the NKG2D receptor and its ligands MICA and MICB, on leukemic blast from acute myeloid leukemia patients.

Material and Methods: We analyzed heparinized peripheral blood samples from 14 newly diagnosed AML patients. NKG2D was searched in purified NK cells from 5 patients, and in peripheral blood mononuclear cells from the other 9 patients. All the samples were activated with IL-2 (10 ng/uL) and IL-12 (5 ng/uL); after two days of culture, the cells were stained with mouse mAb anti-human CD3-FITC/CD16+CD56-PE and anti-NKG2D-PerCP/Cy5.5. To determine MICA and MICB, peripheral blood mononuclear cells were cultured for two days in the presence of magnesium valproate. The cells were stained with mouse mAb anti-human CD34-APC, anti-CD45-PerCP; MICA/B were stained with primary mouse IgG followed by anti mouse IgG-FITC. All of the cells were analyzed by flow cytometry. **Results:** It was observed that cells from patients with AML M1, M2, M4, M5 and M7 expressed NKG2D on its surface; it was also showed

a slight tendency to increase the expression of MICA and MICB on leukemic blast in response to stimulus with valproate. On the other hand, cells from patients with AML LMA M3, showed a tendency to decrease the expression of NKG2D, even when they were stimulated with cytokines. It was not observed expression of MICA and MICB on the blast cells of these patients. **Conclusions:** NK cells of patients with AML M3 have lower expression of NKG2D receptor on the surface of the membrane. NK cells of patients with AML M1, M2, M4, M5 and M7, no change in the expression of NKG2D on its membrane, even when stimulated with interleukins. Blast cells from patients with M1, M2, AML M3, M4, M7 5 and slightly expressed on the surface ligands MICA and MICB.



A1131

NEUROPSYCHOLOGICAL ABNORMALITIES SECONDARY TO CHEMOTHERAPY AND RADIATION THERAPY FOR CHILDHOOD WITH ACUTE LYMPHOBLASTIC LEUKEMIA IN REMISSION AT MEXICAN POPULATION. de Diego Flores Chapa JE, Altawel-Pérez GS, Tlatempa-López E. Universidad Nacional Autónoma de México Centro Médico Nacional 20 de Noviembre del ISSSTE.

Introduction: Acute lymphoblastic leukemia (ALL) is a neoplasm that usually occurs in childhood, comprises 80% of this kind of illness affecting a large number of children under 18 years in the world. Treatments based on chemotherapy and radiotherapy have increased life expectancy up to 75%. The treatment involves impairment of healthy cells, particularly from central nervous system because these cells and the white matter are very sensitive to minimum biochemical changes observed in different cognitive functions. There have been several studies to determine neuropsychological impairments of ALL and its treatment. **Objective:** The aim of this study was to prove the existence of neuropsychological impairment associated with the treatment of ALL in mexican children. **Material and Methods:** The children selection was made according the diagnostic criteria

established by the Hematology service of CMN 20 de Noviembre del ISSSTE. Children selected had to be school-age children diagnosed with acute lymphoblastic leukemia, as inclusion criteria they should know to read and write, do not have infiltration to CNS, and do not present any psychiatric disorders. Parents signed an agreement form which authorized the implementation of the protocol and the filming of the executions of the child as well children sign an approbation letter. The application of the neuropsychological battery was developed in 4 sessions and held individually, with a duration of 1 ½ hours each. It should be mentioned that during the application of these tests children were provided with neuropsychological support to help us determine the level of deficit of the process in assessment. We evaluated 12 children with ALL whose treatment ended at least 6 months before the study. The neuropsychological test included WISC IV, TAVECI, Oral Trail, Rey's Figure, the ENI memory text, a Child Depression Inventory, Corsi Cubes, Tower of London (children's version), Test Neuropsicológico de Memoria Visual y Aprendizaje and Wisconsin Card Sorting Test, while the parents go through an interview filling a questionnaire about the development of their child. **Results:** The evaluation results show alterations in cognitive functioning mainly in processing speed, verbal memory and working memory. **Conclusion:** Neuropsychological assessment must be based on a comprehensive model, embracing each element: individual, family, and sociocultural. As we know these deficits affect social, emotional and school performance necessary to have a healthy biopsychosocial development. It is important to do an accurate diagnosis, to look forward into an intervention aimed not only to help them with cognitive impairments but also with the everyday life.

A1132

ACUTE MYELOID LEUKEMIA IS A DISEASE OF YOUNG ADULTS IN MEXICO. Ceballos-López AA, Sánchez-Cárdenas M, Flores-Jiménez JA, Martínez-Hernández RA, Cuervo-Sierra J, Saldaña-Vázquez R, Pequeño-Luevano MP, Tarín-Arzaga LC, Gómez-Almaguer D. Hospital Universitario de la Universidad Autónoma de Nuevo León.

Introduction: Acute myeloid leukemia (AML) is the second most common type of leukemia in developed countries and the most deadly, with a median age at diagnosis of 65 years and rarely diagnosed before the age of 40 years. The overall U.S. survival rate associated with AML from 1996 to 2002 was 21.7%. In Mexico there are few epidemiological data about it, with a report of a median age of 45 years in a hospital in the center of the country. Nowadays, despite various available therapeutic approaches improvements in survival and cure are not remarkable. **Objective:** Describe the demographic characteristics of AML patients and their survival in a northeast institution of Mexico. **Material and Methods:** We analyzed medical files of patients with newly diagnosed AML between January 2003 and December 2011, in the hematology service of the Hospital Universitario de

la Universidad Autónoma de Nuevo León. **Results:** We included 157 patients for this analysis. In regard to gender 73 (46.5%) were females and 84 (53.5%) males. The median age at diagnosis was 35 years (range 1-85). In the M3 group, with 24 patients (15.3%), median age was 27 years (6-59) while the No M3 group (84.7%) had a median age of 33 years (1-85). The most common type was M2 with 38.2% followed by M4, with 19.7% and M3 15.3%. Of the 157 patients enrolled, 49 were alive (31.2%), 83 patients died (52.9%) and 25 patients had an unknown current status (15.9%). Survival at 6 months was 64.5%. The median overall survival was 12.9 months (CI 95% 4.4-21.3), with 39.6% at 24 months. **Conclusions:** In our patient population, the median age (35 years) is lower in comparison with data from the western countries including the United States; M2 is the most frequent subtype and overall survival is very similar to international data and the previous information from central Mexico. A leukemia national survey is needed in order to confirm these findings.

A1142

ASSESSMENT OF NUTRITIONAL STATUS OF CHILDREN WITH ACUTE LYMPHOBLASTIC LEUKEMIA BY BIOCHEMICAL PARAMETERS AND BODY COMPOSITION. Leal-Solis BL*, De La Torre-Salinas A*, Lopez-Herrera M*, González-García B*, Ancer-Rodríguez P*, Jaime-Pérez JC*, Gómez-Almaguer D*. *Hospital Universitario "Dr. José E. González", Universidad Autónoma de Nuevo León, Monterrey, Nuevo León, México

Introduction: Acute lymphoblastic leukemia (ALL) is characterized by the uncontrolled proliferation of lymphoid cells from the blood and explains 75% of all cases during childhood, with the peak of incidence at four years of age. Nutritional assessment is a key component of the clinical history of a child diagnosed with ALL; it allows an early intervention in the treatment and support during the illness and helps in preventing the effects of malnutrition on the course of leukemia. **Objective:** To assess the nutritional status and body mass index (BMI) in children newly diagnosed with ALL attending the "Dr. Jose Eleuterio Gonzalez" Nutrition Service, Department of Hematology, "Dr. José E. González" University Hospital and School of Medicine of the Universidad Autónoma de Nuevo León in the period from January 1st to December 31st 2010. **Material and Methods:** Prospective analysis of 55 children younger than 16 years of age diagnosed with ALL. The information was gathered in two stages: the first included anthropometric assessment of body composition which was determined employing the following instruments: electronic scale, body fat analyzer and the tables of children of the National Center for Health Statistics (NCHS, USA). The second stage included sequential clinical review of the children and measurement of biochemical parameters including serum albumin concentration (< or >3.5 g / dl) to assess the nutritional risk index. **Results:** Of the 55 children evaluated, 27 were females and 28 males. With respect to age, 22% had between

1 and 5 years, 49% between 6 to 10, and 29% between 11 to 16 years of age. At diagnosis 55% of the girls had a normal BMI, as did 42.8% of the boys. Obesity was present in 11% of girls compared to 37% of boys. 66% of the girls had albumin levels below the parameter used as an indicator of nutritional risk, compared with 35% of the boys. 33% of the girls and 64% of the boys had normal levels of albumin. **Conclusion:** The BMI determination and biochemical assessment of key nutritional markers is of capital importance at the initial evaluation of children with ALL, who otherwise can start treatment with a compromised nutritional status, which largely determines the response to chemotherapy and the development of complications during treatment. Thorough nutritional evaluation at ALL diagnosis is essential for a better clinical follow-up and to optimize clinical outcomes during treatment of ALL of childhood.

A1151

CONGENITAL LEUKEMIA. Miranda-Madrado MR, Ortiz-Torres G, de Diego-Flores-Chapa J. ISSSTE, Hospital 20 de Noviembre UNAM.

Introduction: Describe a case report of a patient with congenital leukemia. **Objective:** Evaluate the approach, value the possible anomalies and response to therapy in patients with congenital leukemia. **Material and Methods:** Starts during the first 24 hrs of life with a hard nodule, not painful, of 1.5 cm on the chest. By age 2 weeks small nodules and papules, hiperchromic start appearing in all his body. At 3 weeks of age he looks pale, irritable and has tachypnea, he is taken to the pediatrician who diagnosis the flu. He is taken to the hospital where they perform a CBC, with anemia, so they transfuse him 2 BP and refer him to our center. At his arrival he has disseminated hard hiperchromic nodules, 0.5-1 cm in diameter, not painful; lesions obstructing air passage through the nose. He had hepatosplenomegaly, no enlarged lymph nodes. **Results:** CBC: L 18,370, B 43%, Hb 8.2, Hct 24.8, Pq 26,000. PBS: blasts 85%. BMA: blasts 90%, basophilic cytoplasm, with azurophilic granules, some with cytoplasmic vacuoles, nucleus folded or convoluted, fine chromatin pattern, 2-4 nucleoli per cell. Ophthalmology: without infiltrate. Ecocardiogram: FEVI 76%, FA 42%. Cytochemical stain: 86% non specific esterase, 83% peroxidase. FISH: positive MLL rearrangement. Immunophenotype: positive CD 33, CD 15, HLA-DR, CD 71a. Day -3: cytoreductive therapy with cytarabine and 6-mercaptopurine. Day 0: induction therapy with cytarabine and idarubicin. Day +6: he had neutropenic colitis and difficulty breathing. CBC with anemia and thrombocytopenia. He started with respiratory pauses which lead to cardiopulmonary arrest. Advanced life support is started; he is intubated and requires application of epinephrine. He has hiperkalemia, acidosis, hemodynamic instability, IDC and starts bleeding through the endotracheal tube. He again falls into cardiopulmonary arrest, advanced life support is started but after 30 minutes time of death is determined. **Conclusion:** Congenital leukemia is defined as a

neoplasm that is diagnosed from birth to the first 6 weeks of life. It is a rare disease, representing less than 1% of childhood leukemias, and is characterized by non-specific symptomatology. It has been associated with trisomy 21, turner syndrome, mosaic trisomy 9 and mosaic monosomy 7 as well as mutations on chromosome 11 and other neonatal malformations. A nodular cutaneous infiltrate, "blue berry muffin" baby, and hepatosplenomegaly are characteristic features. On the CBC we usually find high white blood count. Most have acute non-lymphocytic leukemia, with M4 and M5 accounting for over half the cases. Progression is usually fatal.

A1161

VORINOSTAT, CYTARABINE, MITOXANTRONE AND ETOPOSIDE AS FIRST LINE TREATMENT IN ACUTE MYELOID LEUKEMIA: PHASE II STUDY. PRELIMINARY REPORT. Martínez-Hernández RA, Ceballos-López AA, Cuervo-Sierra J, Flores-Jiménez JA, Saldaña-Vazquez R, Pequeño-Luevano MP, Tarín-Arzaga LC, Gómez-Almaguer D. Hospital Universitario "Dr. Jose Eleuterio Gonzalez". Universidad Autónoma de Nuevo León.

Introduction: Because of poor results in the treatment of AML alternatives have been investigated such as epigenetic therapy. Vorinostat has demonstrated activity against AML in patients with refractory disease; therefore it was added to conventional first line. **Objective:** Describe the response and safety of treatment with vorinostat scheme. **Material and Methods:** Prospective study of patients with AML treated at the Hospital Universitario de Monterrey since September 2011. Vorinostat was administered 300 mg orally every 8 hours in the first 3 days of induction chemotherapy and the first two consolidations. Induction was performed with cytarabine 100mg/m² daily for 7 days and mitoxantrone 12 mg/m² daily for 3 days. The consolidation was performed by 3 monthly cycles of cytarabine 3gr/m² daily for 4 days and etoposide 150mg/m² daily for 3 days. Prophylaxis was administered with ciprofloxacin, itraconazole and acyclovir, and transfusion support as required. Infectious processes were studied and treated conventionally. Response assessment was performed after 25 days of induction, and after the third consolidation. **Results:** We have included 6 patients, 4 female, median age 23 years. Good gastrointestinal tolerance was observed to vorinostat. At the end of induction therapy 5 patients were evaluable, with CR 4 (80%) and 1 refractory 1 patient was not evaluable because he died on day 6 post induction (sepsis). The median recovery was 23.5 days for myeloid cells and 16.5 days for platelets. All patients developed at least 1 episode of fever and received intravenous antibiotics. First consolidation was applied to 3 patients (2 patients continued treatment). After consolidation the median myeloid recovery was 20 days and platelet 20 days. Second consolidation was applied to 2 patients. The median myeloid recovery was 16 days and 13.5 days platelet. 3 patients currently alive, 2 CR and 1 dead. **Conclusion:** With conventional therapy we had 60% CR. GR was observed in 66% and CR in

80% of evaluable patients. Vorinostat dose administered was well tolerated, and in combination with conventional chemotherapy was safe. Other studies with vorinostat frontline GR have shown 85%, 75% CR (Garcia-Manero et al. 2011:118:763 ASH Annual Meeting Abstracts). It requires a larger number of patients to establish safety response and treatment schedule.

A1167

CYTOGENETIC RESULTS AND ITS IMPACT IN 24 ADULT ACUTE LYMPHOBLASTIC LEUKEMIA IN A HOSPITAL OF NORTHEAST OF MEXICO. A PRELIMINARY REPORT. Saldaña-Vázquez R, Flores-Jiménez JA, Sánchez-Cárdenas M, Cantú-Rodríguez O, Tarín-Arzaga LC, Gutiérrez Aguirre CH, Gómez-Almaguer. Hospital Universitario "José Eleuterio González", UANL, Monterrey, Nuevo León, México

Introduction: Cytogenetic evaluation on childhood acute lymphoblastic leukemia (ALL), has been identified as an important prognostic factor, and has been used for the stratification in different risk groups. Nowadays there is scarce information regarding the prognostic significance of cytogenetics in adult ALL. **Objective:** Describe the characteristics and prognostic significance of cytogenetics in 24 patients with ALL, according to three different risk groups defined by the Medical Research Council and Eastern Cooperative Oncology Group (MRC/ECOG). **Material and Methods:** Adult patients (older than 15 years) diagnosed with ALL in Hospital Universitario "Dr. José Eleuterio González" UANL were analyzed. Characteristics at diagnosis were collected prospectively. Only patients with cytogenetic analysis at diagnosis were included and stratified according to risk groups proposed by the MRC/ECOG. **Results:** Twenty four patients with cytogenetic analysis were included and classified as standard risk (12.5%), intermediate risk (50%), and high risk (37.5%). At end of induction 58.3% were in complete remission, 3 (100%) of standard risk, 7 (70%) of intermediate risk and 4 (57%) of high risk patients. Relapse was documented in 7 (29.2%) of patients; 14.2% of standard risk, 42.8% for both intermediate and high risk patients. The overall survival was 4.78 months (0.43-90.77), event free survival was 11.57 months (0.43-54.37). **Conclusion:** In the present study we observed that both the overall survival and the event free survival are reduce in the high risk cytogenetics group of patients, but there is no statistical difference, maybe because the small group of patients included. We are prospectively increasing the number of patients studied.

A1176

ACUTE LYMPHOBLASTIC LEUKEMIA. TEN YEARS OF CYTOGENETIC ANALYSIS AT THE NATIONAL CANCER INSTITUTE IN MEXICO. Arcos-Fonseca DM, Cruz-Velazquez J, Chavez-Jacal MS, Diaz-Vargas G, Espinoza-Zamora JR, Cervera-Ceballos EE. Instituto Nacional de Cancerología (INCAN).

Introduction: Acute lymphoblastic leukemia (ALL) is a malignant disorder of stem cells committed to B cell lineage, involving the bone marrow and peripheral blood. The ALL is primarily a pediatric disease, followed by a low incidence in childhood, adolescence and youth, turning them again in the sixth decade in adults accounts for only 15-20%, most frequently the subtype FAB-L2. Historically, the diagnosis of patients based on morphological criteria by bone marrow aspirate, later supported by studies of immunophenotype and cytogenetic analysis of both conventional and molecular. The difference in treatment outcome between children and adults is probably not due to the presence of a different disease, but the high frequency of cytogenetic abnormalities present in adults associated with a poor prognosis such as the presence of the Philadelphia chromosome, on the other hand, a different response of cytotoxic drugs and high rates of hematological toxicity contribute to a worse outcome, particularly in patients over 50 years. **Objective:** Identify ALL cytogenetic abnormalities diagnosed from 2000 to 2010 at the National Cancer Institute of Mexico. **Material and Methods:** Analysis of karyotype reports and epidemiological studies of cases with ALL issued from 2000 to 2010, and the frequency of cytogenetic abnormalities reported in the group. **Results:** 582 samples were identified, of whom 49% are male, and this group had a mean age of 48 years with a range that moves from 16 to 78 years of age. The variety L2 represents the highest percentage of diagnoses in this group (90%), while L1 and L3 varieties represent only the remaining 10% (5% and 5% respectively). Among the chromosomal abnormalities identified in this group, normal karyotype with BCR/ABL positive (13%) by FISH is the most significant event since the two techniques are mutually exclusive. **Conclusions:** While conventional cytogenetic analysis is a basic pillar, the implementation of the analysis of fluorescence in situ hybridization (FISH) using probes specific sequence and analysis of reaction in the polymerase chain are of great importance in the detection of rearrangements gene that can be skipped by chromosome analysis.

A1177

RETROSPECTIVE ANALYSIS OF CYTOGENETIC FINDINGS OF ACUTE MYELOID LEUKEMIA DIAGNOSED AT THE NATIONAL CANCER INSTITUTE. Arcos-Fonseca D, Cruz-Velazquez J, Chavez-Jacal MS, Espinoza-Zamora JR, Diaz-Vargas G, Cervera-Ceballos EE. National Cancer Institute (Mexico).

Introduction: Acute myeloid leukemia (AML) is a clonal expansion of hematopoietic progenitors either nonlymphoid maintaining their capacity for self-renewal, but are limited in their ability to differentiate into functional mature cells, it is also distinguished by recurrent chromosomal alterations present and genetic alterations. The LAM may develop as a result of the clonal progression of other disorders of pluripotent hematopoietic cell, including chronic myeloid leukemia, polycythemia vera, primary myelofibrosis,

essential thrombocythemia and myelodysplastic syndrome. It is estimated that approximately two thirds of patients with detectable cytogenetic abnormalities LAM. Of these approximately 60% show consistent specific aberrations. Approximately 10% of patients with LAM have a translocation involved, although the percentage varies depending on age being approximately 20% in patients younger than 50 years and 6% in patients over this age. **Objective:** Identify findings conventional cytogenetic and molecular cytogenetic (FISH) in cases with a diagnosis of LAM from 2000 to 2010 in the cytogenetics laboratory of the National Cancer Institute. **Material and Methods:** Analysis of reports of cytogenetic (karyotyping, FISH) and epidemiological analysis of cases with LAM issued from 2000 to 2010. **Results:** In the LAM group analyzed 52% (480) of the reports are male, the average age is 44 years with a range of 16 to 90 years of age. The group shows that the promyelocytic leukemia (LP) is the variety most commonly observed (43%). 24% (109 reports) are in the range M4, which was observed in the inversion of chromosome 16 [inv (16)] in 17% by FISH. Only 17% of the group corresponds to the range M2, which is observed in the t(8; 21) in 40%. The remaining subgroups M0, M1, M5, M6 and M7 are below 10%. Cytogenetic markers characteristic of these strains were determined by FISH. **Conclusions:** The molecular cytogenetic technique is a faithful tool in the identification of key markers in LAM. We anticipate an improvement in cytogenetic diagnosis of LAM with the speed of FISH, not to mention the screen by karyotype. The findings of cytogenetic aberrations in AML enrich the diagnosis or monitoring when sample conditions permit.

A1185

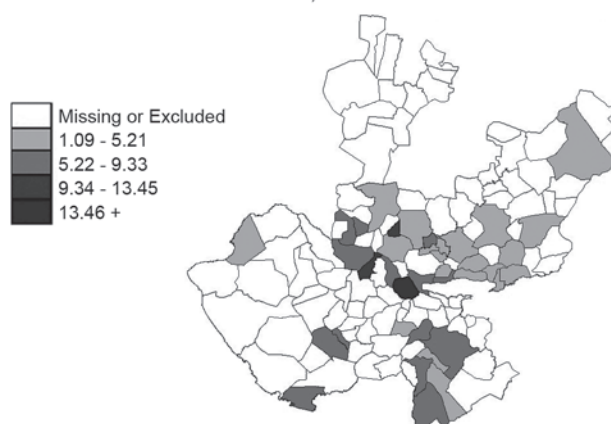
EPIDEMIOLOGICAL PROFILE OF DEATHS FROM LEUKEMIA AND LYMPHOMAS IN JALISCO'S DELEGATION OF MEXICAN SOCIAL SECURITY INSTITUTE (IMSS) FROM 2001 TO 2010. Gutiérrez-Aguirre O*, Castañeda-Lopez G**, Gutiérrez-Guerrero MG***. *UMAE Hospital de Pediatría, Centro Medico Nacional de Occidente, Instituto Mexicano del Seguro Social. **Coordinación de Información y Análisis Estratégico, Delegación Jalisco, Instituto Mexicano del Seguro Social. ***Coordinación de Información y Análisis Estratégico, Delegación Jalisco, Instituto Mexicano del Seguro Social.

Introduction: According to demographic statistics manual version in 2008, there were 67.048 deaths nationally due to malignant tumors, although it's not specified how many are due to leukemia and lymphoma, when searching by age group, we found that the age groups most affected are: 1 to 4 years with 190 deaths, 5 to 9 years with 574 cases, 15 to 24 with 535 cases, 25 to 34 years with 377 cases. In our delegation we don't have studies of epidemiological profile of patients who die of leukemia or lymphoma. **Objective:** To describe the epidemiological profile of deaths from leukemia and lymphomas in Jalisco's delegation of Mexican Social Security Institute (IMSS) from 2001 to 2010. **Material and Methods:** We analyzed the database of institutional mortality (SISMOR) records for the years 2001 to 2010,

the results are presented in frequencies and percentages and we obtained the municipal rates per 10,000 users of IMSS in Jalisco.

Results: In the study period, 1385 deaths were recorded in total, 733 (52.9%) were men, the year that registered more deaths was 2001 with 193 cases (13.9%), the mean age of patients was 47.23 years. The disease most common was non-Hodgkin lymphoma with 435 cases (31.4%), followed by acute lymphoblastic leukemia with 362 cases (26.1%) and acute myeloid leukemia with 246 cases (17.8%). The age group most affected was over 60 years with 533 cases (38.5%). The region of the state with the highest rate of cases per 10,000 users of IMSS Jalisco was Zacoalco de Torres with a rate of 17.5. **Conclusions:** We found that contrary to what is reported in world literature, the age group with higher mortality due to leukemia and lymphomas are patients over 60 years, the study's limitations do not allow us to perform a deeper analysis to measure other specific risk factors in our population. In addition, it's necessary further research to clarify the high rate of cases in the municipality of Zacoalco de Torres.

Municipal rate of deaths due to leukemia and lymphomas per 10,000 users of Mexican Social Security Institute in Jalisco, Mexico.



A1191

ATRA SYNDROME, A CASE REPORT. Galván-López I, Couary-Aguilera P, Tuna-Aguilar E, Guadarrama-Beltrán E, Rosas-López A, Crespo-Solis E, González-Rodríguez K, Espinoza-Macias G. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, México DF, México.

Introduction: The introduction of trans retinoic acid (ATRA), changing the treatment of APL. However, ATRA therapy has potential adverse effects. The ATRA syndrome (differentiation), consists of elevated leukocyte count, fever, respiratory distress, interstitial pulmonary infiltrates, hypoxemia, pleural or pericardial effusion, renal failure or weight gain. In patients treated with ATRA alone, the incidence is approximately 25%, while those treated with ATRA-chemotherapy is re-

duced to 5%. We present a case of AS in a patient treated with chemotherapy-ATRA, leucopenia and infection, resulting in an initial misdiagnosis. **Objective:** 1. Displaying a case of ATRA syndrome and combination therapy. 2. Infer the importance of high clinical suspicion even without meet all criteria. 3. Highlight dexamethasone treatment as valuable tool to control AS. **Material and Methods:** Prospective follow-up and retrospective review of medical history and laboratory studies. **Results:** A woman 44 years, diagnosed in October 2 of an Acute Promyelocytic Leukemia a intermediate risk, made by Bone Marrow Aspirate(BMA) hematic cytology(HC), FISH t(15:17) positive; Fibrinogen 233 and D-dimer, 3225. Treatment with Protocol IC APL 06(ATRA+daunorubicin) started in October 3, HC:leucocytesxmm3, 3.400 and total neutrophils(NT) 1938xmm3. At 48 hours after ATRA start she presented dry cough, normal chest x-ray. On October 7 fever peak associated with a persistent cough, chest CT scan normal. Hospital-acquired pneumonia was suspected and antibiotic treatment started. October 8: Hematologic deterioration: 500xmm3 leucocyte and 305xmm3 NT. A zone of cellulite was detected in a right thigh at the puncture site for BMA, Clindamicyn was added. The CT showed no abscess. Ventilatory impairment, desaturation(80%) in ambient air, then Puritan was placed at 100%. four days after: Ventilatory impairment progressed and required endotracheal intubation and was transferred to the Intensive Care Unit (ICU). Evolution: Dexamethasone 10 mg every 12 hours was started for high suspicion of AS (CT findings in pleural effusion, pericardial effusion, multiple unpolished glass opacities and consolidation areas for bilateral multi-focal pneumonia). Oliguria and creatinine elevation. But, positive cumulative balance decreased progressively without dialysis requirements only diuretics. Antibiotics remained until neutropenia and fever resolution. Dexamethasone was permanent until withdrawal of antibiotics and was gradually decreasing until its suspension. The patient was successfully restart ATRA without pulmonary or hematologic complications. Currently in remission of APL and no recurrence of ATRA syndrome. **Conclusions:** 1. ATRA syndrome requires high clinical suspicion. 2. Infectious processes are the main causes of pulmonary infiltrates. 3 When AS is suspected the immediate use of dexamethasone and stop ATRA (even debatable if leucopenia) until exclude the diagnosis.

A1198

ARSENIC TRIOXIDE (ATO) PLUS ALL-TRANS-RETINOIC ACID (ATRA) IN THE TREATMENT OF THE NOVO ACUTE PROMYELOCYTIC LEUKEMIA (APL). Ovilla-Martínez R, Angeles-Labra AR, Blancas-Ojeda MH, Jiménez-Alvarado RM, Monreal-Alcántara C, Amador Acosta D. Hematology Department, Hospital Ángeles Lomas. Huixquilucan, México.

Introduction The APL is one of the most aggressive leukemias characterized by severe coagulopathy and average life expectancy of less than a month if treatment is not implemented. The

combination of ATRA and anthracycline based chemotherapy is currently the standard approach to treat newly diagnosed APL. The arsenic trioxide (ATO) by itself or combined with ATRA has proven to be effective in the production of complete molecular remission in treatment-naïve patients. **Objective** Our objective was to prove the effectiveness and safety of ATO combined with ATRA in the treatment of newly diagnosed APL. **Material and Methods:** We included 3 patients with newly diagnosed APL 39, 40 and 35 years old, 2 females and 1 male. WBC at the beginning was 0.7, 0.9, and 72.0 x 10⁹ /L. All 3 patients shown t(15;17) and PML-RAR alpha. All of them were treated with ATRA 45 mg/m² per day orally, as a single agent in the first 10 days, then ATO 0.15mg/kg per day intravenously until complete remission (CR) was achieved. Afterwards, a consolidation course was administered consisting of ATO plus ATRA. ATO in cycles at the same dose for 14 days during 7 months and ATRA at the same dose daily for 7 months. After the end of treatment all patients were followed until the present report. We monitored bone marrow, cytogenetic and molecular rearrangement every 3 months in the first year and then every six months until completing 5 years. After a short period of hospitalization all patients were treated in ambulatory care. **Results:** All 3 patients obtained complete hematologic, cytogenetic and molecular remission in 1, 2 and 3 months respectively; all completed the treatment schedule, and persisted without any evidence of APL, during 34, 15 and 12 months respectively. Nobody developed DIC during the induction of remission and they required minimal transfusion support. **Conclusions:** Although this is a small number of patients, we reproduced the results previously reported by others and proved that ATO plus ATRA may serve as an alternative to chemotherapy in untreated patients with APL.

A1202

PANCYTOPENIA IN ADULT AML PATIENT CAUSED BY PARVOVIRUS. González-Zenteno S*, Crespo-Solis E*, Llamas-Gutiérrez F**, Lome-Maldonado C**. *Departamento de Hematología, **Departamento de Patología. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Subiran, México D.F.

Introduction: A 40-year old man with AML and hepatic myeloid sarcoma, in first remission, received chemotherapy with high-dose cytarabine (HDC) as second consolidation. Three weeks later the patient recovered from the aplasia post-chemotherapy. Then, suddenly the patient developed fever associated with severe anemia, neutropenia and thrombocytopenia. A bone marrow aspirate and biopsy was done to rule out relapse of the disease. The biopsy showed giant erythroblasts and cytopathic changes suggestive of parvovirus B-19 infection. Immunohisto-chemistry corroborated the viral infection. No blasts were seen. Intravenous immunoglobulin was administered and chemotherapy was deferred. A complete recovery was obtained 11 weeks later. The patient completed 3 consolidations with HDC. At the end of therapy PCR for parvovirus B-19 was still positive, but cytopenias were

not present. Patient continues in complete remission. **Objective:** Review a case of a patient with pancytopenia associated with parvovirus B19 infection. **Material and Methods:** Case review. **Results:** B19 parvovirus infection in immunocompromised patients, such as a patient with AML chemotherapy may be the cause of pancytopenia. **Conclusions:** Erythroblastopenia due to parvovirus infection has been reported mainly in children with ALL patients. Parvovirus B19 infection should be suspected in leukemic patients with unexplained cytopenias after an acute febrile illness. Very sensitive methods are often needed to confirm the diagnosis, for example immunohisto-chemistry or PCR for parvovirus, since routine serological tests may be unreliable in immune-compromised patients.

A1213

SIMULTANEOUS PRESENTATION OF ACUTE MYELOID LEUKEMIA AND BURKITT LYMPHOMA: CASE REPORT AND REVIEW OF THE LITERATURE, Flores-Villegas LV, Merino.Pasaye LE, Trejo-Gomora J, de-Diego-Flores-Chapa JE, Reyes-Zepeda NC, Ortiz-Torres G. Pediatric Hematology Service; CMN "20 de Noviembre" ISSSTE

Introduction: The incidence in the pediatric population with acute myeloid leukemia is 1 case per 100 000 / year and non-Hodgkin lymphoma is 6 cases per 100 000 / year. In the review of the literature, we don't found reports of two simultaneous hematologic malignancies in children. In studies reviewed found that the onset of acute myeloid leukemia may present over a range of 5 to 15 years after being diagnosed and treated non-Hodgkin lymphoma, this is secondary to the use of chemotherapeutic agents, radiation as well as the dysregulation of c-Myc and BCL-2. **Objective:** Perform a case report and review of the literature of a simultaneous presentation of two hematologic malignancies: Burkitt lymphoma and acute myeloid leukemia in a child in the Pediatric Hematology Service CMN "20 de Noviembre" ISSSTE. **Material and Methods:** All the pathological, flow cytometric, cytogenetic and assays were performed in certified clinical laboratories using standard techniques. Immunohistochemistry was performed with antibodies to CD18, CD54, LFA-1, Ki-67, CD45, BCL-6, CD10, CD20, CD22, CD79a, sIgM, TdT. **Results:** This paper report a male patient, 6 years old with a presentation of 4 months of evolution and history of abdominal pain, weight loss, anorexia and fever. He's never received chemotherapy treatment. He presented with generalized lymphadenopathy, pleural effusion and retroperitoneal mass. Laparoscopy was performed to obtain a biopsy. The histopathologic diagnosis was Burkitt's Lymphoma. Stage III. No cytogenetic abnormalities are found, discard infection by Epstein Barr Virus. Treatment was initiated with the scheme of COPADM. After four months of treatment the patient presented leukocytosis, cephalgia and bilateral proptosis. The bone marrow aspiration showed us 86% of myeloblasts. The second diagnosis was acute myeloid leukemia M2. Was determined CNS infiltration with involvement the optic nerves

and meninges. The kid received systemic chemotherapy and intrathecal according to the protocol of acute myeloid leukemia of High Risk, High-doses of methotrexate and Radiotherapy. He achieved complete remission (CR) after the chemotherapy. We don't found compatible donors for bone marrow transplantation. At three months of treatment he relapsed of Burkitt's lymphoma with central nervous system activity and retroperitoneum. Finally the patient died. **Conclusion:** This is the first report in the review of the literature of simultaneous presentation of acute myeloid leukemia and Burkitt lymphoma in children. This kind of patients had a short response to treatment with early relapse and poor prognosis

A1219

EFFECTIVITY OF LAL-T 98 TREATMENT PROTOCOL IN CHILDREN WITH LEUKEMIA IN INSTITUTO NACIONAL DE PEDIATRIA, MEXICO CITY. Paredes R, Lopez N, Taboada C, Monsivais A, Gonzalez L, Romero L, Tavera G. Instituto Nacional de Pediatría.

Introduction: In the first decade of 1970, the evident that acute lymphoblastic leukemia (ALL) of T cell progenitors has a worse prognosis than acute lymphoblastic leukemia of B progenitor cells was made. However, in the end of the decade of 90", a greater improvement in the prognosis of these patients with rotatory schemes comprising of high dosis of methotrexate (MTX), cyclophosphamide (CFA), cytarabine (Ara-C), and L asparaginase has been observed. **Objective:** The objective of this work was to know the event-free survival (EFS) of pediatric patients with ALL -T treated with "ALL T 98" protocol at Instituto Nacional de Pediatría (INP), Mexico city from 1998 to 2010. **Material and Methods:** From 1998 until 2010, 40 patients less than 18 years old diagnosed with ALL-T in INP were enrolled in trial "LAL-T 98". The patients were classified in accordance with the criteria of Nacional Cancer Institute (NCI) as standard-risk group (SRG) and high- risk group (HRG). Treatment protocol principally consisted of the administration of high dosis of MTX, CFA, Ara-C during consolidation and reconsolidation, and an upkeep treatment with multiple qtxx combined with 4 agents plus 20 to 30 dosis of L-asparaginase at high dosis until the stage of elective cessation of chemotherapy is reached. **Results:** The median age was 8 years (interval 1 – 16 years); 9/40 (22%) of the patients were more than 10 years old; 72% were male. The median WBC count at presentation was 20,000 mm³ (interval 1,100 – 880,000 mm³). The major extramedullary disease manifestations were a mediastinal mass in 15% of patients. The average time of follow-up was 5 years (interval 0.2 – 12 years). EFS was 64% at 5 years. Only a patient presented therapeutic failure. Reoccurrence of the sickness was seen 12 (30%) of the patients and 8 of them recovered with treatment while the remaining 4 patients were subjected to elective cessation of chemotherapy. Only one of the patients with reoccurrence is still alive bone marrow allogenic transplant with a follow-up of 11 months. There were three death

during remission due to treatment complications. We did not find any difference in EFS between the two risk groups. **Conclusion:** Treatment with high dose rotator scheme with MTX, CFA, Ara-C, and a high upkeep dose of L-asparaginase was useful for children with diagnosis of T ALL.

A1229

TREATMENT OF ACUTE LYMPHOBLASTIC LEUKEMIA PHILADELPHIA CHROMOSOME POSITIVE WITH TYROSINE KINASE INHIBITORS IN PEDIATRIC PATIENTS. Amador R, Ledesma C, Lugo Y, Castellanos J, Hernandez P, Guzman L, Garcia M, Solis L. Regional General Hospital No. 1 "Dr. Carlos Sanchez Navarro Mac Gregor" IMSS

Introduction: Chromosome Acute Lymphoblastic Leukemia F + (LLACrPh +) characterized by a reciprocal translocation between the chromosome 9 and 22, determining gene fusion protein Bcr / Abl. Occurs in 3-5% in children with LLACrPh +, is associated with poor prognosis. The development of tyrosine kinase inhibitors has been a therapeutic advance in combination with chemotherapy regimens, as treatment may include transplantation. **Objective:** know the response to treatment in children with tyrosine kinase inhibitors. **Material and Methods:** We reviewed medical records of pediatric patients diagnosed with ALL CrPh + over a period of 10 years (2000-2010), a retrospective, descriptive. **Results:** Were 4 patients diagnosed with ALL CrPh +, three men and a woman in a period of 5 years range from 5 to 10 years (mean 7.7 years). Morphology (FAB) 3 patients L1, and 1 L2. 3 patients B-cell and 1 lineage T. All received chemotherapy with remission induction, 2 with high-risk conventional scheme Total XIII, St Jude, and 2 hiperCVAD, all achieved complete remission (CR), at day 28, continued chemotherapy, a patient could not be performed karyotype at diagnosis and relapse two months shows bone marrow, this time a sample was taken for karyotype and the report begins with imatinib mesylate, although in poor condition general rescue myelosuppression by chemotherapy and died of septic shock. In the remaining three cases was initiated tyrosine kinase inhibitor to remission, one with imatinib mesylate, presenting nausea and vomiting as side reactions but those with symptomatic control, remained in CR for 21 months, with the sixth study negative molecular months of treatment, but died of tuberculosis meningitis. In the other 2 patients administered dasatinib, currently an RC (13 months) with proper medication tolerance and molecular remission from the third month after start of medication has not been transplanted as donor and has continued with chemotherapy maintenance. The fourth patient in CR 16 months, with molecular remission at 3 months of initiation of appropriate medication and tolerance, at 11 months of diagnosis was unrelated allogeneic transplant (umbilical cord) with complete graft and negative molecular study that dasatinib continued. **Conclusions:** CrPh + ALL is a very poor prognosis in children with chemotherapy alone has seen a range of disease-free survival of 25- 30% increased 60 to 75% with transplantation. With TKI inhibited the growth of posi-

tive cells Bcr / Abl, induces apoptosis and there is suppression of cell proliferation, thus was able to increase the response, which correlate with our cases.

A1234

HEMATOLOGY IN LATIN AMERICAN. ¿WHERE WE ARE? del Campo-Martínez MA, Gabús R, Galeano S, de Souza C, Howard SC, Milone JH, Tejerina-del-Valle ML, Alfaro-Lucero J, Rosales C, Zarza J, Cauvi F, Borelli G, Prieto M. Sociedades de Hematología Latinaamericanas.

Introduction. In recent years there has been an increase in ties between the societies of Latin American Hematology and the American Society of Hematology. **Objectives:** Provide knowledge about reality of hematology in our continent, through which strengthen the role of Latin American societies of hematology each other and with the American Society of Hematology, and encourage coordination of cooperative projects, to improve diagnosis and treatment of patients suffering from blood disorders in the Latin community. **Material and Methods:** We asked hematology societies the distribution of "evaluation forms" and general data summary and status of the hematology of the country, reflecting the opinion of the society they represented. The information was collected and analyzed by the coordinators of HOA-LA 2011 and presented the results in the "Breakfast Session from HOA-LA 2011". **Results:** For the territory and population of the countries studied, there are 4.306 hematologists, rate of 0.9 per 100,000 inhabitants, with heterogeneity by country, region and between them, the 76.7% are members of their society, in 7 countries have formal programs residence. The hematologist in public sector is between 25 and 73% and private assistance between 10 and 20%. There are few national records and limited to certain diseases. Most people can easily access reference centers; there are geographical areas with difficulties such as Brazil and Mexico. The availability of flow cytometry, cytogenetics, molecular biology, immunohistochemistry, CT, MRI and PET, is heterogeneous across countries and within between. The availability of irradiated blood products is limited. Most countries have access to basic treatment hemato-oncology, but only partly to the high cost. There are countries that do not perform HSCT. Funding is guaranteed by the state or medical organization to which it is affiliated. There are almost no national protocols. The number of hematologists and training is "insufficient" and "mislocated". **Conclusions:** Encourage development of records, national guidelines and protocols. Deficiencies were detected in cytogenetic and molecular biology, and the need for better IHC techniques, and exchange of knowledge in these areas. Also needs of imaging centers in some countries due to the limited availability of MRI and PET in particular. The need of completely access to basic hematology-oncology treatments for all people and gradually evaluate various models for access to expensive treatments and cooperation between Latin America

to encourage the development of programs of TPH. In the area of training was assessed need for programs to stimulate the formation of hematology and related areas.

A1235

IMMEDIATE AND LONG-TERM MEDICAL EFFECTS OF NUCLEAR AND RADIATION ACCIDENTS: CHERNOBYL, GOIANIA, BELARUS, TOKAIMURA AND FUKUSHIMA. Gale RP*, Baranov A*, Guskova A**. *Department of Experimental Medicine, Division of Haematology, Imperial College, London, UK; **Burnasyan Federal Medical Biophysical Center (FMBC, Moscow, Russia)

Exposure to ionizing radiations results in immediate and long-term effects. These effects can also be classified as deterministic and stochastic. Immediate effects typically occur within the 1st year but can occur over the 1st 10 years after exposure. In contrast, long-term effects typically begin after 5-10 years following exposure and may extend over 50 or more years. Here, we review these effects in the context of recent nuclear and radiation accidents. Acute exposure to high-doses of ionizing radiations results in acute radiation syndrome features which are variable and dose-dependent. Predominate targets include the gastrointestinal system, skin, bone marrow and central nervous and cardiovascular systems. There are often concomitant and confounding thermal and concussive injuries in the setting of nuclear and radiation accidents. Strategies to treat acute radiation syndrome require a reasonably accurate dose-estimate based on physical, biological and computational analyses. Estimates of dose-uniformity are also important. Identification of appropriate interventions depends on these estimates and include supportive care, antibiotics and anti-virus drugs and transfusions of RBC and platelets. Molecularly-cloned hematopoietic growth factors and hematopoietic cells transplants are considered at higher doses. Drugs directed to prevent/repair gastrointestinal and/or skin damage including radio-protectors and molecularly-cloned hormones are effective in animals but untested in humans with acute radiation syndrome. We discuss our experience using these modalities in ARS after several recent nuclear and radia-

tion accidents. We show several important points: (1) accurate dose-estimation and uniformity is complex and difficult under real-time accident conditions in contrast to theoretical models; (2) calculation of risk:benefit ratio of diverse interventions is also complex and difficult when there is concomitant and confounding injuries; (3) bone marrow recovery is possible at doses much greater than the estimated LD₅₀ in humans; (4) molecularly-cloned hematopoietic growth factors accelerate bone marrow recovery; and (5) there is a small but definite dose-window where HCTs are reasonably-considered. Stochastic effects of exposure to ionizing radiations include cancer, birth defects and genetic abnormalities. Radiation-induced leukemia and thyroid cancers occurs more quickly (within 2-10 years) whereas other cancers have a latency of 10-60 years. Cancer and birth defects are of greatest concern as genetic effects are unproved in humans. Risk of radiation-induced cancer is greatest in young persons and decreases dramatically with increased age at time of exposure. A linear relationship between radiation-dose and increased cancer-risk is proved for doses >200 mSv but unproved (and controversial) at lower doses. Although some data support a linear, no-threshold relationship, other data are inconsistent with this notion. It is now 25 years since the Chernobyl NPS accident. By considering estimates of radionuclide releases, atmospheric and terrestrial dispersion, population exposure and results of epidemiological studies it is possible to determine cancer consequences of Chernobyl to-date. The most striking finding is a substantial increase in thyroid cancers in persons <16 years old at exposure from ¹³¹I and possibly ¹³⁷Cs. This probably resulted from exposure to radionuclide-contaminated foodstuffs. Convincing evidence of a substantial increase in leukemia or other cancers is lacking; any increase appears to be of a small magnitude, if at all. There are no convincing data of an increase in birth defects or genetic abnormalities. Radionuclide releases from the Fukushima NPS to-date are about 10-fold less than from the Chernobyl NPS (5.2 versus 0.5 TBq). Furthermore, there was more effective control of foodstuff contaminated with ¹³¹I and distribution of non-radioactive iodine. Based on these considerations there will be few, if any, radiation-induced cancers from the Fukushima NPS accident. Birth defects and genetic abnormalities are rather unlikely.

CHRONIC LEUKEMIA

ORAL PRESENTATION

A1047

QUINONE METHIDE RELEASING NITRIC OXIDE ACETYL SALICYLIC ACID (NO-ASA) DERIVATIVES: A POTENTIAL NEW SUBSTANCE CLASS FOR THE TREATMENT OF CHRONIC LYMPHOCYTIC LEUKEMIA (CLL). Poll-Wolbeck SJ, Gehrke I, Liakos A, Krallmann S, Krüger M, Berkessel A, Hallek M, Kreuzer KA. Department I of Internal Medicine, University at Cologne, Cologne, Germany; Institute for Organic Chemistry, University at Cologne, Cologne, Germany.

Introduction: In chronic lymphocytic leukemia (CLL) new therapeutic options are desired, especially for treatment of resistant patients with adverse prognosis. Amongst nitric oxide acetyl salicylic acid (NO-ASA) derivatives quinone methide forming NO-ASA derivatives show superior apoptosis inducing potency. Thus, we could recently show potent apoptosis induction in primary CLL cells in vitro and reduced tumor growth in a xenograft CLL-like mouse model upon para-NO-ASA treatment.

Objective: To further improve the efficacy and specificity of NO-ASA derived compounds, we generated six different NO-ASA derivatives. **Material and Methods:** Primary CLL cells as well as peripheral blood mononuclear cells (PBMCs) of healthy volunteers were incubated with these substances and their parent compound ASA for 24 hours. Cell viability was assessed by determination of the intracellular ATP content in a luminometric assay and flow cytometrically by annexin-V-binding and propidium iodide exclusion. The substances were also tested for their impact on CLL cells derived from bad prognosis patients showing TP53 mutations. **Results:** All synthesized NO-ASA derivatives proved to be far more potent towards primary CLL cells in a viability test than their chemical origin ASA (ASA effective dosage 50% (ED50) = 7851 uM, NO-ASA derivatives ED50 ranging from 0.29 uM to 69 uM). Changes of the ASA part of para-NO-ASA had only little impact on the ED50 as assessed flow cytometrically (para-NO-ASA = 6.4 uM vs. 7.4 uM and 3.6 uM, respectively). In contrast, if the linker was bound to a protective group or the leaving group more firmly attached to the linker, which impedes quinone formation, the ED50 was significantly increased (para-NO-ASA = 4.7 uM vs. 69.0 uM and 53.0 uM, respectively). Compound VI, which was especially

designed to release quinone methides upon metabolism, resulted in a decrease of the ED50 to 0.3 uM in the cell viability assay and 1.0 uM in the flow cytometry based assay. Further, II, III and VI showed selectivity towards CLL cells as PBMCs from healthy donors were less sensitive as assessed by flow cytometry (ED50: para-NO-ASA = 64.1 uM vs. 37.5 uM, 50.0 and 74.3 uM, respectively). II and III also showed high potency towards TP53 mutated CLL cells. **Conclusions:** It can be concluded, that the release of the quinone methide appears to be critical for NO-ASA efficacy on primary CLL cells. Further, the NO-ASAs might be a therapeutic option for patients with treatment resistant forms of CLL.

A1049

LYMPHOID ENHANCER-BINDING FACTOR-1 EXPRESSION IS ASSOCIATED WITH REQUIREMENT OF TREATMENT, ZAP70 POSITIVITY, AND FIBROMODULIN EXPRESSION IN CHRONIC LYMPHOCYTIC LEUKEMIA. Erdfelder F, Praulich I, Rocha CK, Gehrke I, Poll-Wolbeck SJ, Hallek M, Kreuzer KA. Department I of Internal Medicine, University at Cologne, Cologne, Germany.

Introduction: Lymphoid enhancer-binding factor-1 (LEF1) is a Wnt-pathway transcription factor which is specifically expressed at early stages of B-cell differentiation and exerts oncogenic functions. The extracellular matrix protein fibromodulin (FMOD) is a tumor-associated antigen in chronic lymphocytic leukemia (CLL). It was also shown to be associated with p53-mutation and resistance to DNA damage in CLL cells. We and others could previously show that LEF1 and FMOD are highly overexpressed in CLL. **Objective:** Based on the above-mentioned findings, we speculated that LEF1 and FMOD may be associated with poor prognosis and advanced disease stage in CLL. **Material and Methods:** Quantitative real-time PCR was used to determine LEF1 and FMOD mRNA expression in 120 and 100 primary CLL samples, respectively. The ZAP70 assay was performed according to the method of Rassenti et al. using clone 1E7.2 conjugated with Alexa Flour 488. We also compared LEF1 and FMOD expression of patients requiring treatment with the expression of patients in recently diagnosed Binet stage A. Moreover, we investigated correlations of LEF1 and FMOD with each other. **Results:** Patients requiring treatment showed a nearly four fold

higher mean LEF1 mRNA relative expression ratio (RER) compared to patients in recently diagnosed Binet stage A ($p < 0.001$). Moreover, mean LEF1 RERs were 54 and 37 in ZAP70-positive and ZAP70-negative patients, respectively ($p = 0.004$). In addition 90% of the patients with a LEF1 RER over 70 required treatment and had more than 80% of lymphocytes in the peripheral blood. Furthermore, we found a highly significant positive correlation of LEF1 with the percentage of lymphocytes in the peripheral blood (Spearman's $\rho = 0.440$, $p < 0.001$). However, we did not observe a significant difference in LEF1 expression between CD38-positive and CD38-negative patients. We also found LEF1 and FMOD expression to be highly correlated with each other (Spearman's $\rho = 0.405$, $p < 0.001$). Moreover, we performed a LEF1 median split dividing the patients in high and low LEF1 subgroups and found an over 8 fold higher mean FMOD RER in the high LEF1 group ($p < 0.001$). **Conclusions:** Our results suggest that LEF1 expression is associated with poor prognosis, and the requirement of treatment in CLL. Thus, LEF1 might be involved in the process of disease progression and possibly can serve as a molecular parameter for risk assessment and/or monitoring of CLL.

A1097

DEEP SEQUENCING OF ANTIIONCOGENE TP53 IDENTIFIES THE CLONAL EVOLUTION AND SUBCLONAL HETEROGENEITY IN CHRONIC LYMPHOCYTIC LEUKEMIA PATIENTS. Tichy B*, Stano-Kozubik K*, Malcikova J*, Trbusek M*, Tom N*, Lochmanová, Kantorová, Smardova J**, Doubek M**, Mayer J**, Pospisilova S*. *Masaryk University, Central European Institute of Technology, Brno, Czech Republic; **University Hospital Brno, Brno, Czech Republic.

Introduction: Mutations in the TP53 gene are associated with poor prognosis in chronic lymphocytic leukemia (CLL) patients and functional status of p53 became one of the most important prognostic factors in CLL. The frequency of p53 inactivation is substantially higher in pretreated patient cohorts compared to those investigated before first-line therapy. **Objective:** Recent studies showed the selection of TP53 mutated clones by therapy (clonal evolution) as well as rare occurrence of cases with leukemic subclones harboring different mutations in the TP53 gene (subclonal heterogeneity). **Material and Methods:** We performed deep sequencing of TP53 exons 4–9 in selected CLL samples using amplicon sequencing approach (Roche GS Junior system). We sequenced five PCR products representing 6 exons to a depth at least 2500 in each sample. All samples were previously analyzed by at least one of these methods detecting the TP53 mutations – FASAY (Functional Analysis of Separated Alleles in Yeasts), direct sequencing, resequencing microarray and DHPLC. **Results:** Several consecutive samples from each patient were analyzed for TP53 mutations using deep sequencing approach. For example, in three consecutive samples from one CLL patient we observed clear evidence of clonal evolution – in

the first sample (year 2005) only I195T mutation was detected (2.6 % of reads; not detected other methods), in the second sample (year 2007) another two mutations appeared (R248W, 3.6 % and R273C, 3.4 %) in addition to I195T (28.1 %). In this sample we were able to detect only the I195T mutation using other techniques (FASAY, DHPLC, resequencing microarray). The third sample was from relapse after allogeneic transplantation of hematopoietic stem cells and here we detected only R248W mutation (97.8 %) in conclusion with other methods. We have analyzed three other cases suspected of subclonal heterogeneity. We confirmed the presence of more than one mutation (missense, nonsense or splicing site) with frequency > 1 % in each of them. **Conclusions:** Here we show that deep sequencing of TP53 gene is highly sensitive method, which enables an early detection of unfavorable prognostic factors in CLL. All detected TP53 mutations were present as minor clones undetectable by standard techniques. The detection of minor clones of CLL cells with p53 mutation prior to therapy could be of potential relevance for therapy selection or subsequent management of the disease relapse. The work was supported by grant projects of Ministry of Education, Youth and Sports No. MSM0021622430, CZ.1.05/1.1.00/02.0068 a CZ.1.07/2.3.00/20.0045.

A1122

CHRONIC MYELOID LEUKEMIA, CHOOSING THE THERAPEUTIC OPTION ACCORDING TO AGE AT DIAGNOSIS AND ECONOMICAL ASPECTS. Flores-Jiménez JA, Cantú-Rodríguez O, Jaime-Pérez JC, Ceballos-López AA, Martínez-Hernández RA, Cuervo-Sierra J, Gutiérrez Aguirre CH, Gómez-Almaguer D. Universidad Autónoma de Nuevo León, Hospital Universitario, Monterrey, N.L., México.

Introduction: Chronic myeloid leukemia (CML) represents about 15% of leukemias in adults. The incidence is estimated at 1-2 cases per 1 000, 000 population per year. CML is a myeloproliferative disorder characterized by a reciprocal translocation $t(9; 22)(q34, q11.)$, that usually appears with a median age of 60 and 67 years in Europe and United States of America respectively, however, all age groups can be affected. With the high efficacy of tyrosine kinase inhibitors, stem cell transplant has been considerate as second line treatment option in CML patients. **Objective:** In this study we analyzed the demographic characteristics of the study population and compared the cost associated to the therapeutic option used. **Material and Methods:** We retrospectively studied the clinical records of all CML patients of the Hematology Service of the University Hospital "Dr. José E. González", who received treatment with imatinib or hematopoietic stem cell transplant from January 2001 to September 2011. We found 127 patients evaluable for this analysis. **Results:** The median age was 39 years (range: 4 to 80), 41 years in patients who received only imatinib and 35 years for those who were treated with imatinib and allogeneic HSCT. Patients treated with imatinib were older than those

who received imatinib and allogeneic HSCT ($p < 0.004$). In regard to gender 52% were males and 48% females. No statistically significant difference in sex distribution existed. The 3 years median overall survival (OR) was 85% for patients with imatinib and 72% for patients receiving HSCT. Cytogenetic remission was 26% for patients with imatinib and 100% for allogeneic HSCT patients. The cost of one year of imatinib in our institution is approximately 26,000 USD and regarding the

allogeneic HSCT the cost is 30,000 USD. In our institution the cost of the first year of treatment with imatinib covers 86% of the cost of an allogeneic HSCT. **Conclusions:** The median age at diagnosis of patients in this group was 39 years, lower than the found in Europe and the USA. This finding should be investigated in more detail for their potential impact on the costs of treating the disease. In our hands long term treatment with allogeneic HSCT seems to be less expensive than imatinib.

POSTERS

A1034

APOPTOTIC EFFECTS OF NEW IRON(III) CHELATES OF S-METHYL-THIOSEMICARBAZONES ON K562 CELLS. Kuruca SE*, Ülküseven B**, Bal-Demirci T**, Akgün-Dar K***, Arslan M*, Gürel E***. *Department of Physiology, Istanbul Faculty of Medicine, Istanbul University, Capa-Istanbul, Turkey; **Department of Chemistry, Faculty of Engineering, Istanbul University, Avcilar-Istanbul, Turkey; ***Department of Biology, Faculty of Science, Istanbul University, Beyazit-Istanbul, Turkey.

Introduction: Thiosemicarbazones have a wide range of biological activity. Metal complexes of thiosemicarbazones are a class of compounds presenting some biological applications as antiviral, antibacterial and antitumor, depending on the parent aldehyde, ketone and metal ion. 3-aminopyridine-2-carboxaldehyde thiosemicarbazone (Triapine) that was shown antitumor effects in preclinical experiments is being evaluated in phase I and II clinical trials. **Objective:** In this study, we investigated the apoptotic effect mechanisms of the compounds which we defined their cytotoxic effects. That is why we determined Caspase-3 and cytochrome-C which are molecules have a role in apoptotic pathway, and also we verify with DNA fragmentation.

Material and Methods: We synthesized the iron (III) chelates of some hydroxy or methoxy-substituted N1,N4-diarylidene-S-methylthiosemicarbazones which are in the $[Fe(L)Cl]$ general formula. Thiosemicarbazones were characterized by elemental analysis and magnetic measurements, ^1H-NMR , UV-Vis, IR and mass spectroscopy. Cytotoxicity experiments were done using K562 chronic myeloid leukemia cells by [3-(4,5-dimethylthiazol-2-yl)-2,5-diphenyltetrazolium bromide] (MTT) assay. The concentration of compounds that provides 50% inhibition cell growth (IC_{50}) were calculated from dose-response curve. Cytotoxic effect of thiosemicarbazones was evaluated by comparing the IC_{50} values. Caspase-3 and cytochrome-C activation were determined in cells treated in IC_{50} of thiosemicarbazones. In addition DNA fragmentation was determined by reaction diphenylamine method. **Results:** All of iron(III) chelates of N1,N4-diarylidene-S-methylthiosemicarbazones have selective anti-leukemic effects in K562 cells. Caspase-3 and cytochrome-C activity of K562 cells were significantly increased by compari-

son with controls without compounds. DNA fragmentation was produced 65-84 % by all compounds. **Conclusions:** Our results indicate that selective cytotoxic potential of the chelates depends on metal ion, hydroxy and methoxy substituents, and also substituent locations. We are thinking possessing a drug potential of the new N1N4-diarylidene-thiosemicarbazone chelates which have shown antileukemic effect. This study is supported by TUBITAK. (with the project #109S188)

The features of new synthesized iron (III) chelates of S-methylthiosemicarbazones and IC_{50} values.

Compounds	R_1	R_2	Metal	MW	$IC_{50}(ug/ml)$
1	3-OCH3	4-OCH3	Fe	462.3	3.5
2	4-OCH3	3-OCH3	Fe	462.3	5
3	4-OCH3	4-OCH3	Fe	462.3	0.53
4	3-OCH3	4-OH	Fe	448.35	4.5
5	3-OH	4-OCH3	Fe	448.35	0.61
6	3-OCH3	3-OH	Fe	448.35	3.5
7	4-OH	4-OCH3	Fe	448.35	0.01

A1036

CLONAL CHROMOSOMAL ABNORMALITIES IN PHILADELPHIA NEGATIVE CELLS (PH-) OF CHRONIC MYELOID LEUKEMIA (CML) PATIENTS DURING TYROSINE KINASE INHIBITORS (TKIS) TREATMENT: ARGENTINE EXPERIENCE. Labarta JD*, Varela AI**, Figueroa MF**, Zarate T**, Pavlovsky C***, Giere I***, Lombardi V***, Verri V****, Gonzalez J****, Flores GM****, Larripa IB⁵, Mercado-Guzman ZV⁵, Otero IS**, Lluesma-Goñalons M**, Ardaiz M del C**, Moiraghi EB**. *Genoma, Buenos Aires, Argentina; **División Hematología del Hospital JM Ramos Mejia, Buenos Aires, Argentina; ***Fundaleu, Buenos Aires, Argentina; ****División Hematología del Hospital CG Durand, Buenos Aires, Argentina; ⁵Departamento de Genética de la Academia Nacional de Medicina, Buenos Aires, Argentina.

Introduction: Clonal cytogenetic abnormalities in Ph- cells of CML patients during α -Interferon and TKI treatment have been previously described. These are evaluable only in patients who achieve some type of cytogenetic response. This is different from clonal evolution that refers to additional chromosomal changes in Ph+ cells. **Objective:** To evaluate the frequency and significance of Ph- chromosomal abnormalities in a cohort of Argentinian CML patients one during first or second line TKI therapy: Imatinib (IM), Dasatinib (D) and Nilotinib (N). **Material and Methods:** A retrospective analysis was performed in order to calculate the frequency of Ph- abnormalities. Cytogenetics: Chromosome analysis of unstimulated Bone marrow (BM) or Peripheral blood (PB) cultures and PHA-stimulated PB cultures (constitutional karyotype) was performed by GTG-banding. FISH: CEP 8, cytocell probe and double fusion bcr/abl, LIVE-LEXEL. **Results:** 193 patients were analysed: 79.8% (154/193) treated with IM; 15% (29/193) with Dasatinib and 5.2% (10/193) with Nilotinib, with a median follow up of 28 months (range: 13-41) Data for each ITK are shown in table 1. Total frequency of Ph- abnormalities was: 5.7% (11/193). Some of these abnormalities disappeared during the patients' follow up. Some patients presented cytopenia and/or evolution to advanced phases CML. **Conclusions:** -Some of the abnormalities described in this cohort have already been reported (Trisomies 6, 8, XYY and monosomy 7), being T8 and -7 the most frequent. Trisomy 6 has been reported in one patient under IM treatment but our patient received N. This is the first report of Trisomy 6 in patients receiving second generation ITKs. We found not reports of Trisomy 11 and 13 in the literature. - Monosomy 7, as previously reported was related poor prognosis. Trisomy 11 and 13 as sole abnormalities are indicators of poor prognosis in other hematologic malignancies. Our patient with Trisomy 11 had a poor outcome. - All this indicates the importance that patients on TKIs, that have achieved some grade of cytogenetic response, should be regularly followed with cytogenetic techniques besides of RT-PCR for bcr/abl, not only to evaluate the decrease of Ph+ cells, but also to look for Ph- chromosomal abnormalities.

A1044

FLOW CYTOMETRIC ANALYSIS OF THE LYMPHOMA-SPECIFIC ANTIGEN ROR1 SUBSTANTIALLY INCREASES DETECTION SENSITIVITY AND SPECIFICITY

OF NEOPLASTIC B CELLS IN CHRONIC LYMPHOCYTIC LEUKAEMIA (CLL). Uhrmacher S, Schmidt C, Praulich I, Rocha CK, Gehrke I, Poll-Wolbeck SJ, Hallek M, Kreuzer KA. Department I of Internal Medicine, University at Cologne, Cologne, Germany

Introduction: Flow cytometry is commonly used to establish the diagnosis of chronic lymphocytic leukemia (CLL) using a defined combination of antibodies to discriminate between normal B cells and CLL cells (CD5, CD19, and CD23). Unfortunately none of these markers are exclusively expressed on CLL cells, but also on other cells in peripheral blood or bone marrow. The receptor tyrosine-like orphan receptor 1 (ROR1) is an embryonic glycoprotein involved in several developmental processes. It was shown to be highly expressed on CLL cells, but not on normal B cells. **Objective:** Due to this fact we examined the potential of ROR1 as a diagnostic marker in initial and follow-up diagnostics of clinical heterogeneous patients with CLL. **Material and Methods:** Peripheral blood of 177 CLL patients in different clinical stages as well as of healthy volunteers was subjected to flow cytometric analysis of ROR1 surface expression. The study included 105 untreated patients with or without relevant comorbidities and 72 patients which were analysed at different time points of treatment. A fluorescently labelled anti-ROR1 antibody was used. We also examined ROR1 expression in 12 patients with other B- non- Hodgkin- Lymphomas (B-NHL). **Results:** ROR1 was expressed uniformly at high levels on the surface of CLL cells (mean 98.1%) of untreated patients, independent of clinical stage or other comorbidities. ROR1 expression was also consistently high on CLL cells from treated patients, independent of received chemotherapeutic substances and treatment cycle. In contrast, in healthy volunteers ROR1 expression on different cell subgroups was only marginal (mean 1.9%) and significant lower than on CLL cells ($p=0.01$). Also in B-NHL patients we could detect ROR1 surface expression, albeit in varying intensities. Further, in CLL patients no correlation between the ROR1 expression level and prognostic markers could be detected. **Conclusions:** Due to the uniform ROR1 expression on CLL cells in treated as well as untreated patients we conclude that ROR1 might be a valuable additional marker for initial and follow up diagnostics, but can not detect CLL specifically, because of its expression in other B- NHL. With regard to our former results

Table 1. Frequency of Ph- abnormalities.

TKI	n Patients	Median Follow up, months (range)	Chromosomal Ph'(-) Abnormalities	% Ph'(-) Abnormalities (n/nn)
IMATINIB	154	27 (13-44)	Trisomies: 8, 11 and 13; XYY; del (12p); hexaploidy (6n=138)	3.9 (6/154)
DASATINIB	29	28 (24-31)	Monosomy 7; Trisomies: 8 and 13	10.3 (3/29)
NILOTINIB	10	24 (11-24)	Trisomies: 6 and 8	20.0 (2/10)

we also conclude that ROR1 might be able to improve flow cytometry based MRD diagnostic which is currently equally sensitive than laborious RQ-IgH-PCR.

A1046

CLONAL EVOLUTION OF PROGNOSTICALLY ADVERSE MOLECULAR AND CYTOGENETIC FEATURES DURING THE DISEASE COURSE IN CHRONIC LYMPHOCYTIC LEUKEMIA (CLL). Praulich I, Rocha CK, Schiller J, Hallek M, Kreuzer KA. Department I of Internal Medicine, University at Cologne, Cologne, Germany.

Introduction: It has been shown that chronic lymphocytic leukemia (CLL) patients displaying a mutation within the p53 tumor suppressor gene or a chromosomal deletion 17p (del(17p)) have an adverse prognosis when compared to CLL patients who do not exhibit these anomalies. Furthermore, there is growing evidence that complex chromosomal aberrations, and especially translocations detected in the neoplastic clone are associated with a similar inferior outcome even if the patients otherwise exhibit prognostically favourable factors. **Objective:** During a twelve month period we identified a total of 23 patients with progressive CLL showing either a p53 mutation and a del(17p) (n=8, 35%) or a sole p53 mutation (n=7, 25%) or a sole del(17p) (n=1, 4%) or complex chromosomal abnormalities (= three structural or numerical anomalies) without del(17p) (n=7, 30%). **Material and Methods:** Eighteen patients of the total population (78%) exhibited an unmutated immunoglobulin heavy chain variable (IgHV) locus. **Results:** In 6 cases the observed lesion could not be detected in a previous investigation. Out of these cases in 3 patients a del(17p) evolved together with a p53 mutation, in one case a p53 mutation evolved without a del(17p) and in two cases complex chromosomal aberrations developed. The mean duration for the development of adverse parameters was 21 months. In most cases (n=4) those occurred after treatments with either fludarabine and/or cyclophosphamide and/or rituximab or bendamustine and/or rituximab. However, in two patients clonal evolution was detectable without any causative therapy for CLL. **Conclusions:** We conclude that clonal evolution of prognostically adverse molecular or cytogenetic lesions can occur in patients with initially favourable risk profile. It appears that this holds true not only for established parameters such as del(17p) and p53 mutations but also for newer prognostic factors such as a complex karyotype. Biologically, an overall genetic instability may account for this phenomenon as well as a mitogenic or mutagenic effect caused by cytostatic drugs. It can therefore be discussed, whether assessment of adverse risk factors including conventional karyotyping should be generally performed at an earlier point of the disease course and might be repeated if clinical signs of progression are evident.

A1048

BIDIRECTIONAL INTERACTIONS OF CLL CELLS AND THEIR MICROENVIRONMENT VIA THE ANGIOPOIETIN

2/TIE2 AXIS. Gehrke I, Poetzsch B, Poll-Wolbeck SJ, Hallek M, Kreuzer KA. Department I of Internal Medicine, University at Cologne, Cologne, Germany.

Introduction: The microenvironment crucially impacts on the pathophysiology of chronic lymphocytic leukemia (CLL). We could recently show that bone marrow stromal cells support CLL cell survival in a coculture set up via secretion of the vascular endothelial growth factor (VEGF). Further, several studies demonstrated an angiogenesis-independent function of VEGF in the apoptotic resistance of CLL cells, their most prominent pathophysiological feature. Also the pro-angiogenic protein angiopoietin (Ang)2 has been detected in plasma/serum of CLL patients and associated with advanced disease stages, shorter progression free survival and general bad prognosis. In addition, CLL cells have been described to secrete Ang2 under culture conditions. Up to date, no data is available on expression of the Ang2 receptor Tie2 in CLL. Also, it is not known whether Ang2 acts in an autocrine fashion or potentially influences the microenvironment by Ang2 secretion. We confirmed Ang2 expression (rtPCR) and secretion (ELISA) (105.2 pg/ml +/- 18.8 pg/ml) by CLL cells (n=12). Further, we could show that CLL cells do not express Tie2 receptor (flow cytometry) (n=10). Interestingly, we detected low Tie2 levels on granulocytes (CD45/side scatter) of CLL patients. Human umbilical vein endothelial cells (HUVEC) functioned as positive control for Ang2 and Tie2. Based on absence of Tie2 on CLL cells we conclude that Ang2 does not act in an autocrine fashion but the CLL cell rather influences its microenvironment by secretion of Ang2. **Objective:** To assess possible bidirectional interactions of CLL cells with the bone marrow microenvironment. **Material and Methods:** We set up cocultures of CLL cells with the bone marrow-derived stromal cell line HS5. **Results:** Preliminary results suggest upregulation of Ang2 but no induction of Tie2 expression in CLL cells in this set up. In addition, primary bone marrow-derived stromal cells will be analysed for Tie2 expression and Ang2 secretion and their impact on CLL cells. To assess the lymph node microenvironment, we generated nurse like cells (NLCs) from PBMCs of CLL patients and are currently investigating their Ang2/Tie2 status and impact on CLL cells. The same investigation will be undertaken with monocyte-derived dendritic cells and macrophages from CLL patients. **Conclusions:** Blockage of secreted Ang2 using a peptibody or soluble Tie2 protein and blockage of Tie2 using an antibody shall give further insight into the interactions of CLL cells with their microenvironment and may identify the Ang2/Tie2 axis as target for therapeutic interventions directed towards the CLL cells within their protective niches.

A1052

IMATINIB FOR CHRONIC MYELOID LEUKEMIA AS FIRST LINE THERAPY. RESULTS IN CUBA. Pavón-Morán V, Jaime-Fagundo JC, Hernández-Ramírez P, Gómez-Baute R, Arencibia-Núñez A, Carnot-Uría J, Hernández-Padrón C,

Espinosa-Estrada E, Ávila-Cabrera OM, González-Otero A, Dorcós-Balea E, Lam-Díaz RM, Baganet-Cobas A, Machín-García S, Menéndez-Veitia A, Nordet-Carreras I, Núñez-Quintana A, Serrano-Mirabal J, Agramonte-Llames O, Amor-Vigil AM, Bencomo-Hernández A, Lavaut-Sánchez K, Hernández-Cabezas A, Fernández-Nodarse R, Cristo-Pérez V, Díaz-Durán CV, Fernández-Aguila J, Goldman J. Instituto de Hematología e Inmunología, La Habana, Cuba. On Behalf of the National Program Group for Leukemia.

Introduction: The introduction of imatinib has changed the treatment of chronic myeloid leukemia (CML). The drug reduces the leukemic cells burden in chronic phase of the disease and promises to prolong survival very substantially in comparison to earlier drugs. It is recommended as first line therapy for newly diagnosed patients with CML in chronic phase (CP). **Objective:** We evaluated the efficacy and tolerance of this drug and survival in our patient population. **Material and Methods:** Patients were eligible for the study if they were between 5 and 65 years of age with a confirmed diagnosis of Ph+ CML in CP, within six months before study entry and a previous treatment for CP-CML limited to hydroxyurea. Imatinib (400 mg per day) was administered orally. Patients were followed with complete blood counts every 4 weeks. Bone marrow aspiration and cytogenetic studies were performed every 6 months. The study was conducted in 14 hospitals throughout the country and 61 patients with newly diagnosed CML were enrolled. **Results:** Fifty-seven patients achieved haematological response. Major cytogenetic response (MCyR) was achieved in 97,6% and complete cytogenetic response (CCyR) was achieved in 77,04 %. Of the 47 patients who achieved CCyR, 16 (26,22%) obtained molecular remission. With a median follow-up of 41 months, the estimated rate of event free survival at 60 months was 89,6 %. All events were mild and resolved after a temporary suspension of the drug. **Conclusions:** Imatinib treatment was resumed in each case without further problems.

A1057

STANDARDIZATION OF A MULTIPLEX RT-PCR FOR DETECTION OF CHRONIC MYELOPROLIFERATIVE DISORDERS (MPDS) BASED ON TAGUCHI METHOD. Pérez-Contreras VA, Alonso-Muñoz C, Cortés-Penagos C. Laboratorio de Marcadores Moleculares de Neoplasias Hemato-Oncológicas, Facultad de Químico-Farmacobiología, UMSNH. Laboratorio de Embriología, Facultad de Ciencias Medicas y Biológicas "Dr. Ignacio Chávez" UMSNH. Laboratorios Mendel. Morelia Michoacán México.

Introduction: Analysis of BCR-ABL transcripts by RT-PCR is routinely used to assess both diagnostic and treatment response in patients with chronic myeloid leukaemia (CML). Other Myeloproliferative Disorders (MPD) including polycythaemia vera (PV), essential thrombocythaemia (ET), and primary my-

elofibrosis (PMF) has been recently characterized at a molecular level showing the prevalence of a single point mutation in the cytoplasmic tyrosine kinase JAK2 (JAK2V617F), introducing a new excellent diagnostic molecular marker that has been adopted by the World Health Organization (WHO) as a diagnostic criteria for this myeloid neoplasms. These discoveries have changed the landscape for diagnosis and classification of chronic myeloproliferative disorders. **Objective:** The objective of the present study is the use of Taguchi method as an alternative approach to standardized a multiplex RT-PCR to detect both BCR-ABL and JAK2V617F markers in a simple test usefull in a routine molecular diagnostic practice. **Material and Methods:** RNA from positive samples was extracted by standard methods. PCR reaction was carried out after Reverse Transcriptase reaction with variable amounts of specific primers depending on the experiment being investigated (JAK2 V617F and BCR-ABL). We carried out all reactions in duplicate with specific controls to avoid non specific products. **Results:** The development and standardization of the RT-PCR test to detect JAK2 V617F mutation and BCR-ABL translocation variants b2a2 and b3a2 was carried out using an orthogonal array including major factors affecting the PCR reaction (primer and Mg+2 concentration, Taq polymerase, annealing time and temperatures and PCR cycles) and their associated levels. Using this array through 36 independent assays we successfully optimized a simple test for both molecular markers. Additionally, the optimized test distinguishes homozygous from heterozygous mutations. The test was also design to analyze the presence of additional mutations on JAK2 exons 12 and 13 by sequencing analysis. **Conclusions:** Current results suggest that orthogonal arrays are useful tools for optimization molecular techniques leading to determine the most significant variables during the test. The Taguchi method allowed us to standardized a rapid and robust method for detection of the BCR-ABL transcripts and JAK2 V617F mutation with favorable performance characteristics that make it advantageous for clinical diagnosis.

A1060

TREATMENT OF HAIRY CELL LEUKEMIA: LONG-TERM RESULTS IN A DEVELOPING COUNTRY. Ruiz-Delgado GJ^{*.5}, Tarín-Arzaga LC^{****}, Alarcón-Urdaneta C^{*.5}, Calderón-García J^{*.6}, Gómez-Almaguer D^{****}, Ruiz-Argüelles GJ^{*,**.*.***}. ^{*}Centro de Hematología y Medicina Interna. Clínica Ruiz; ^{**}Laboratorios Clínicos de Puebla. Clínica Ruiz; ^{***}Universidad Popular Autónoma del Estado de Puebla; ^{****}Hospital Universitario de Nuevo León; ⁵Benemérita Universidad Autónoma de Puebla; ⁶Facultad de Medicina. Universidad la Salle. México.

Introduction: Splenectomy, the treatment of choice of hairy cell leukemia (HCL) for many years, leads to normalization of the peripheral blood counts in approximately one half of all cases. Interferon-alpha (IFN) renders a high overall response but most responses are partial. Cladribine, (2-CDA) produces remarkably high remission rates with a single cycle of therapy and the

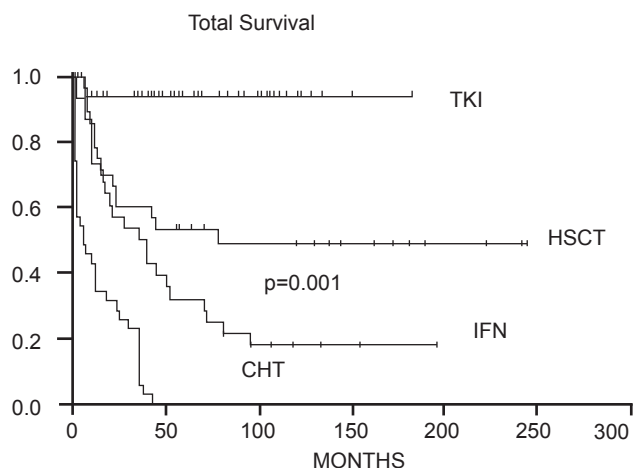
excellent results with this agent delivered as a single course of therapy led to 2-CDA being the primary therapy selected by many hematologists. **Objective:** To assess the response of patients with HCL in México, treated with either IFN or 2-CDA, taking into account the affordability of the two drugs in two institutions: Centro de Hematología y Medicina Interna de Puebla y Hospital Universitario de Nuevo León, between July 1987 and May 2011. **Material and Methods:** Twenty nine consecutive patients with hairy cell leukemia were treated in the two institutions with either IFN (n = 18) or cladribine (n = 11). Between 1987 and 1993, all patients were treated with IFN. After 1993, patients were advised to be treated with 2-CDA but the final selection of the treatment was done by the patients themselves, specifically by their ability to obtain the drug abroad, since it has never been commercially available in México. **Results:** Median age was 62 (range 29 to 83) years; there were 21 males and eight females. Seven of the 18 patients in the IFN group (39%) achieved a complete remission (CR), whereas all the patients in the 2-CDA group entered a CR. Three patients in the 2-CDA group relapsed and needed an additional course of the drug, 2, 3 and 6 years after the initial one. The median overall survival (OS) of the whole group has not been reached, being above 217 months, the 217-month OS being 91%. The survival of patients treated with either IFN or 2-CDA was not statistically different (94% OS at 217 months versus 91% OS at 133 months, respectively). **Conclusions:** These data suggest that treatment of HCL with either 2-CDA or IFN is equally effective; treatment costs with IFN are substantially lower than those of the purine analog. These observations may be critical in developing countries, where costs are a major obstacle.

A1067

THE THERAPEUTIC RESULTS IN CHRONIC MYELOID LEUKEMIA PH1+. 1986 TO 2011. CMN "20 DE NOVIEMBRE". López-Hernández MA, Banda-García LI, Alvarado-Ibarra M, Alvarez-Vera JL, González-López M. The Hematology Department. CMN "20 de Noviembre". ISSSTE.

Introduction: Introduction. Thirty years ago, patients with chronic myeloid leukemia Ph1+ hardly reached but five years of life. Since then their probabilities of curing are real, with hematopoietic stem cell allogeneic transplantation, or to live indefinitely with inhibitors on tyrosin quinasa. **Objective:** To present the experience in the treatment of chronic myeloid leukemia Ph1+, in a single hospital, in the past 25 years. **Material and Methods:** Were seen 205 patients. Diagnosis was made with known clinical, hematological and myeloid data. All patients had Ph1+ identified through karyotype. The accelerated and blast phases were considered for this analysis according to WHO criteria. The different treatments were chemotherapy (CHT), interferon (IFN), hematopoietic stem cell allogeneic transplantation (HSCT) or tyrosine kinase inhibitors (TKI). Remission criteria included hematologic remission, complete cytogenetic (CCG) and major molecular remission (MMR). **Results:** The mean age was 43 years (15 to 86). The

gender frequency was 101:104 (F:M). The most frequent clinical and laboratory findings were: weight loss (37%), splenomegaly (52%), mean white blood cells $198 \times 10^9/L$ ($36-999 \times 10^9/L$). The Sokal index was low/medium/high: 77/66/62. On diagnosis 191 patients were in chronic phase and 14 in a blast phase. CHT (busulfan or hydroxyurea) was used from 1986 to 1999 in 66 cases; only (74%) reached hematological remission. IFN associated to hydroxyurea was used from 1991 to 2001 and was indicated in 42 patients; there was hematological remission in 95% and CCG remission in 2%. 34 patients went on to HSCT as of 1992; 44% still survive; all of them have cytogenetic remission. 63 cases received TKI as of 2001; in 65% it was the initial option; most of them (38 patients) with imatinib and three with nilotinib; finally 43 were treated with imatinib, 9 with nilotinib and 12 with dasatinib. There was hematological remission in 98%; CCG remission in 57% and MMR in 32%. Total survival is shown in the chart below. **Conclusions:** Conclusions. TKI's have a clear advantage as a therapeutic option. The maximum efficacy is obtained with HSCT, with regards to achievement of CG remission and absence of progression, but the high mortality, restrictions related to age and difficulty in finding a compatible donor limit its use.



A1082

EPIDEMIOLOGY AND OUTCOME OF TREATMENT OF CHRONIC PHASE CHRONIC MYELOID LEUKEMIA (CML) WITH 400 MG IMATINIB IN MEXICO. Cervera-Ceballos E*, Lopez-Navarro OG*, Candelaria-M*, Gordillo-D*, Best-C**, Urtado-R***, Kassak-J****, Aguayo-A⁵, Vargas-Viveros JP***, Corrales- Alfaro CN*, Martinez-Balbuena M*, Mendez-Labardini JR*. *National Cancer Institute, Mexic; **Western General Hospital "Zoquepan"; ***Hospital Angeles of Pedregal; ****General Hospital of Mexico; ⁵National Institute of Medical Science and Nutrition Salvador Zubiran.

Introduction: Chronic myeloid leukemia (CML) is a clonal disorder secondary to the chromosomal translocation t (9; 22)

(q34, q11). The product of this translocation is a chimeric gene (BCR/ABL) encoding a protein tyrosine kinase, which promotes cell proliferation. Imatinib was developed by Nicholas B. Lydon, Brian J. Druker and Charles L. Sawyers, revolutionizing the treatment of CML and has demonstrated through clinical trials the efficacy in CML. In Mexico, access to treatment with imatinib has been available by the support of the "Glivec International Patient Assistance Program" (GIPAP). Here we present data on chronic phase CML response to treatment with imatinib (400 mg) in a mestizo population in Mexico. **Objective:** To evaluate the clinical characteristics of individuals from Mexico with chronic phase CML treated with 400 mg of Imatinib treated under the GIPAP program and to analyze cytogenetic response at 18 months, disease-free survival and overall survival. **Material and Methods:** We conducted a retrospective study including all consecutive patients during the period of January 2003 to January 2008. Statistical was made with Kaplan and Meier, X², Student T-test. We performed a logistic regression analysis. **Results:** We studied a total of 238 patients with CP-CML, with a mean age of 41.4 years (12-82 years), 130 men (54.6%) and 108 women (45.4%). The grade 3 or 4 toxicities that occurred were: nausea (n = 2, 0.8%), diarrhea (n = 1, 0.4%), anemia (n = 1, 0.4%), thrombocytopenia (n = 4, 1.7%), leucopenia (n = 2, 0.8%) and neutropenia (n = 1, 0.4%). The most common grade 1 and 2 toxicities were nausea (n = 29, 12.2%), facial edema (n = 33, 13.9%), anemia (n = 37, 15.5%), diarrhea (n = 13, 5.5%), fatigue (n = 20, 8.4%), headache (n = 11, 4.6%) and neutropenia (n = 19, 18%). The variables associated by logistic regression with cytogenetic response at 18 months were: the use of imatinib before 6 months of symptom onset, and a low EUTOS. **Conclusions:** The median age is lower than that reported in Anglosaxon series. The variables associated with a cytogenetic response at 18 months were: the use of imatinib before 6 months of onset of symptoms, and a low EUTOS. Overall survival at 5 years was 89.8%, similar to the IRIS study, and the disease-free interval was 73%, which is lower than reported in other studies, this may be due to a lack of attachment to treatment in these patients.

A1088

PREVALENCE OF IMMUNOPHENOTYPES OF CHRONIC LYMPHOPROLIFERATIVE DISEASES AT CLINICA RUIZ-LABORATORIOS. Perez-Romano B, Neira JC, Ruiz-Arguelles GJ, Ruiz-Arguelles A. Laboratorios Clinicos de Puebla and Centro de Hematología y Medicina Interna, Puebla, Puebla, Mexico.

Introduction: A retrospective analysis of the results of the immunophenotyping of cases of chronic lymphoproliferative disease, performed at Laboratorios Clinicos de Puebla (Clinica Ruiz) from 2002 to 2011, was performed. Classification was performed according to the 2nd Latin American Consensus Conference for Flow Cytometric Immunophenotyping of Hematological Malignancies, and those recommended by the World Health Organization. **Objective:** To determine the prevalence

of types and subtypes of chronic lymphoproliferative diseases in Laboratorios Clinicos de Puebla. **Material and Methods:** A total of 4979 immunophenotyping studies in newly diagnosed cases were performed in the aforementioned period. Of these, 856 corresponded to chronic lymphoproliferative diseases. **Results:** Table 1 shows the prevalence of different types of chronic lymphoproliferative diseases that were classified in Laboratorios Clinicos de Puebla from 2002 to 2011. **Conclusions:** The relative low number of cases of chronic lymphoproliferative diseases classified in our institutions, as compared to those of acute leukemia, by no means depicts the prevalence of the disease in our country. This finding is more related to the fact that most lymphomas diagnosed in Mexico are considered as "solid tumors" and referred to the oncologist, rather than the hematologist for treatment. Our laboratory receives patients primarily from hematologists, hence the small numbers of lymphoma cases. The same is true for the proportions of types of disease, for it is very likely that certain diseases are more commonly treated by the oncologist, while others are referred to the hematologist.

Chronic lymphocytic B cell leukemia	547
B lineage chronic lymphoproliferative diseases	229
Hairy cell leukemia	34
Chronic lymphocytic T cell leukemia	20
Marginal zone lymphoma	8
NK cell leukemia	7
Mantle cell lymphoma	5
Cleaved cell lymphoma	1
Hairy cell leukemia variant	3
Masked lymphocytes splenic lymphoma	1
Lymphoplasmocytoid leukemia	1

A1110

CLINICAL AND DEMOGRAPHIC CHARACTERISTICS OF MYELOFIBROSIS PATIENTS REFRACTORY TO CURRENT AVAILABLE THERAPIES IN MEXICO. Alvarado-Ibarra M*; Vargas-Viveros P**; Ovilla-Martinez R***; López-Hernández M*; Hurtado-Monroy R**; Solís-Poblano JC****; Limón-Flores A****; Agreda-Vasquez G⁵; Silva-López S⁶; Kassack-Ipiña J⁷; Zavala-Cervantes S⁸; Aguilar-Luna C⁹; Baez-De la Fuente E¹⁰; Nacho K¹¹; Mucius I¹¹; Iniguez L¹¹; Muniz T¹¹; Romero-Salas T¹¹. * ISSSTE Centro Médico Nacional 20 de Noviembre, México DF; **Hospital Ángeles del Pedregal, México DF; ***Hospital Ángeles Lomas, EdoMex; ****UMAE HE CMN Gral. de Div. Manuel Ávila Camacho IMSS Puebla; ⁵Instituto Nacional de Nutrición Salvador Zubirán, México DF; ⁶ISSSTE Hospital 10 de Octubre, México DF; ⁷Hospital General de México O.D., México DF; ⁸Hospital Star Médica, Mérida; ⁹Hospital Regional "Dr. Valentín Gómez Farías" ISSSTE, Zapopán; ¹⁰UMAE # 25 IMSS Monterrey; ¹¹Novartis Oncology, México DF.

Introduction: Primary Myelofibrosis (PMF) post – polycythemia vera MF (PPV MF), and post – essential thrombocythemia MF

(PET MF) are Myeloproliferative Neoplasms (MPN), characterized by bone marrow fibrosis. Data from USA have estimated the annual incidence of BCRABL1-negative diseases to be 2.1 per 100,000. Current incidence in Mexico is unknown; and available therapies include hydroxyurea, interferon (IFN), and phlebotomy for PV and ET, as well as androgens, steroids, chemotherapy, IFN, thalidomide, EPO and radiotherapy for PMF. A potentially curative therapy is allogeneic hematopoietic stem cell transplantation. This report will focus on the characteristics of 22 MPN patients refractory to current available therapies in Mexico. **Objective:** To explore clinical demographic characteristics of MPN patients in Mexico. **Material and Methods:** We collected data from 22 patients including age, gender, institution (public or private); liver and renal function, complete blood cell count (CBC); subtype of disease; risk group, ECOG performance status, spleen size, and bone marrow fibrosis grade. **Results:** The median age was 63.6 years; 47.4% were female, 58% of patients proceeded from Private Health Institutions (PHI). Liver and renal function were adequate in 100% of patients, defined as direct bilirubin = 2.0 x ULN, and serum creatinine = 2 x ULN; CBC analysis presented a peripheral blood blast count < 10%. 42.1% of patients had PMF; 21% had PPV-MF; and 36% had PET-MF. 11% had Low risk; 53% intermediate-1 risk; 26% intermediate-2 risk; and 10% high risk. ECOG 0 was present in 21% of patients; ECOG 1, 63.16%; ECOG 2, 10.53%; and ECOG 3 in 5.3%. Splenomegaly was found in 100% of cases; with 63% of splenomegaly less than 10cm; 26.36% between 10 and 20cm; and 10.53% more than 20cm. Spleen medium size was 10.3 cm; BLCM. Bone marrow biopsy showed: No Fibrosis, 5%; Grade I, 42%; Grade II, 21%; Grade III. 32%. (Fig 1). **Conclusions:** Characteristics of MF patients of this multi-institutional cohort in Mexico, remark the disease's severity. The median age of patients was according with the literature reported (66 y.o.). In this cohort, most patients were from PHI. PMF was more frequent than PPV-MF and PET-MF. There is a need for improving diagnosis and therapy of this very rare disease.

A1119

ADHERENCE TO IMATINIB THERAPY: SINGLE CENTER EXPERIENCE. Beyan C*, Kaptan K*, Eriksi A**. *Gulhane Military Medical Academy, Etlik, Ankara Turkey; ** GATA Haydarpaşa Training Hospital, Uskudar, Istanbul, Turkey

Introduction: The classical therapy for newly diagnosed chronic myeloid leukemia (CML) is imatinib which is a tyrosine kinase inhibitor. Some of the previously untreated cases have imatinib resistance or intolerance at the time of the diagnosis. Additionally, gained response to imatinib might be lost in time. **Objectives:** Another reason for loss of response is decreased adherence to therapy. The aim of this study is to investigate the compliance of our patients to imatinib therapy. **Material and Methods:** We included 39 CML patients between July 2007-June 2011. According to regulations of Health Ministry drug reports for imatinib

have to be renewed every six months. Renewal periods over and equal to 1 month is considered as nonadherence to therapy. The median age of 39 CML cases are $42,6 \pm 17,4$ years (20-85) and 23 are males. Control periods are median 4(1-7), control gap is median 31 months (4-45). Five of the patients have no response (5.1%), two of them have intolerance (5.1%). **Results:** Nonadherence to therapy is 9.6% (16/166). At least once nonadherence to therapy is 25,6% (10/39). In comparison according to sex have no difference between males (6/23) and females (4/16) ($p=0,620$). Patients =50 years of age have nonadherence to therapy 20,0 % (5/25) whereas patients =50 years have nonadherence to therapy 35,7% (5/14) ($p=0,486$). Nonadherence to therapy in previously treated patients (interferon alpha, cytosine arabinoside, hydroxyurea) is 12,5 % (1/8), previously untreated patients is 29,0% (9/31) ($p=0,323$). Second generation TKI usage because of nonadherence to therapy is 10% (1/10), and 13,8 % (4/29) in other cases. **Conclusions:** Adherence to therapy must be included as an important evaluation parameter in future studies of CML. Patients and families should be educated for the probable bad outcomes of the disease in case of nonadherence to therapy. Counselling programmes could be effective in persisting imatinib medication, resulting in the improvement of overall compliance.

A1130

INITIAL EXPLORATION OF EFFECT OF K562-DERIVED MICROVESICLES ON THEIR PARENTAL CELL. Juanjuan L, Huiyu L, Shiang H, Yan Y, Xin L. Center for Stem Cell Research and Application, Union Hospital, Tongji Medical College, Huazhong University of Science and Technology, Wuhan 430022, China.

Introduction: Recently microvesicles have been widely detected in various biological fluids including peripheral blood, urine and ascitic fluids. They play an important role in cell signaling and the process of molecular communication between cells. **Objective:** The research is to investigate the effect of microvesicles (MVs) which are derived from K562 cell on the proliferation and apoptosis of their parental cell and analyse the mechanism. **Material and Methods:** K562 -MVs were obtained by gradient centrifugation, and labeled with PKH26, a red fluorescent lipophilic dye. Human umbilical vein endothelial cells (HUVEC) were incubated with the PKH26-labeled K562-MVs and then subjected to fluorescence microscope and photographed. After treated with different concentrations of K562-MVs, the proliferation of K562 cells was determined by CCK8 kit and cells apoptosis was analysed by Flow cytometry with a combination of annexin V and propidium iodide (PI). Furthermore the expression levels of bcl-2 gene were examined by semi-quantitative RT-PCR. **Results:** We found that K562-MVs could integrate with HUVEC cells, and the event was visualized using fluorescence microscope (figure 1). This result gave us a message that K562-MVs perhaps could influence the function of target cells by delivering active substances. According to CCK-8 assay, 0.12,

0.20, 0.30, and 0.40mg / ml K562-MVs could promoted growth of K562 cells except the 0.06mg/ml group which had no statistical significance when compared to the control, and the proliferation rates that were (16.39±1.17)%, (33.50±1.44)%, (45.18±2.45)%, and (59.52±6.92)% respectively became higher when the concentrations of K562-MVs increased. This suggested K562-MVs could promote the viability of K562 cells. As compared to the control group, the apoptosis rate of K562 cells was decreased, and the expression level of bcl-2 gene was increased in the 0.12mg /ml K562-MVs group, which indicated that K562-MVs could improve survival and reduce apoptosis in K562 cells and bcl-2 gene may be one of the mechanisms involved in. **Conclusions:** In conclusion, K562-MVs are not meaningless or useless cell debris, but the information carrier with bioactive molecules. K562-MVs can promote their parental cells proliferation and inhibit their apoptosis by increasing expression levels of bcl-2 gene.

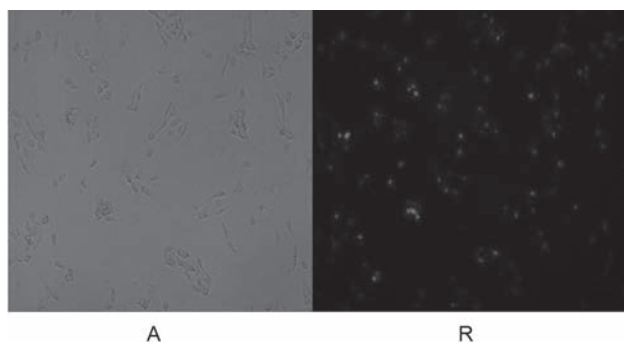


Figure 1. K562-MVs can integrate with HUVEC. Serum-starved HUVEC were incubated with the PKH26-labeled K562-MVs for 10 hours and then subjected to fluorescence microscope. A. The adherent HUVEC can be seen under the visible light. B. The PKH26-labeled K562-MVs can be seen at the place where the HUVEC adhere under Green excitation light.

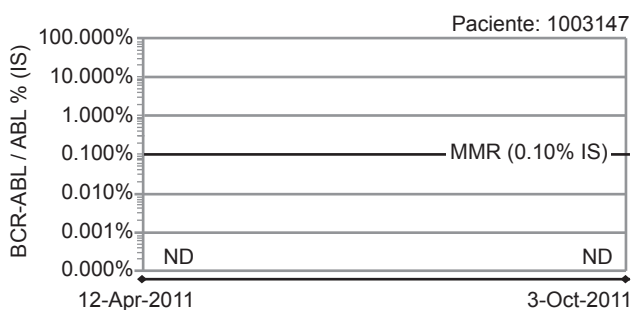
A1133

EARLY AND COMPLETE MOLECULAR RESPONSE IN PH+ CHRONIC-PHASE (CP) CHRONIC MYELOID LEUKEMIA (CML) TREATED WITH NILOTINIB. Aguilar Luna JC. Hematology Department. Hospital Dr. Valentín Gómez Fariás ISSSTE Zapopan, Jalisco MX.

Introduction: Nilotinib is a selective inhibitor of Bcr-Abl recently approved for the treatment of newly diagnosed patients with chronic myeloid leukemia. Nilotinib induces high molecular remission rates measured by qRT-PCR. **Objective:** To report the achievement of early and complete molecular response in a Mexican patient with newly diagnosed Ph+ CP CML treated with nilotinib 600 mg/day. **Material and Methods:** We diagnosed a 49 year old (y.o) woman with CP-CML in february 2010. She had a clinical picture of 3 months of evolution with lypothimia and Thrombocytosis. There was no palpable splenomegaly. The initial CBC count showed: Hb 13.1 g/dL; leukocytes 31.1x 10

9; Platelets 1.008.000/mL and normal DHL. The initial bone marrow karyotyping showed 46, XX, t(9:22)(q34:q11) and the FISH analysis was positive for bcr-abl. In april 2010 we began treatment with nilotinib 600 mg/d. **Results:** After six months of treatment we performed a bone marrow karyotyping showing complete cytogenetic response and also we performed qRT-PCR for bcr-abl showing complete molecular response. The subsequent controls (april 2011 and oct 2011) were reported as undetectable bcr-abl, showing durability of this response. The patient tolerated well nilotinib without any adverse events and without lab abnormalities. **Conclusions:** Nilotinib as front line therapy in our center is a very interesting new therapy since it can induce early and deep responses including CCyR and CMR. The durability of these responses will be monitored in a sixth monthly basis

Resultados acumulados muestran el más reciente 6 visitas con resultados válidos. Por favor, consulte los informes anteriores de los resultados de las anteriores visitas no se muestra.



A1135

EXPERIENCE WITH TYROSINE KINASE INHIBITORS IN CHRONIC MYELOID LEUKEMIA: A ANALYSIS OF ISSSTE MERIDA REGIONAL HOSPITAL. Herrera-Aguilar A, Morales-Adrian J, May-Magaña. Department of Hematology, ISSSTE Mérida Regional Hospital, Mérida, Yucatán, México.

Introduction: Mérida Regional Hospital (MRH) of ISSSTE in Yucatán since 1993 has made the diagnosis of 37 cases of chronic myeloid leukemia (CML) with 92% of those detected in the chronic phase of disease, of which 57% are male, age ranges are between 14 and 84 years with a mean of 54 and our incidence is 1 to 4 cases per year. In our hospital we have 3 tyrosine kinase inhibitors (TKI): imatinib, nilotinib and dasatinib. **Objective:** Report the response obtained with TKI in patients with CML the ISSSTE MRH in Yucatan. **Material and Methods:** Was assessed hematologic and molecular responses measured by polymerase chain reaction (PCR), was used response criteria of European LeukemiaNet Group and the National Comprehensive Cancer Network. **Results:** Of the 37 patients there are currently alive and in treatment 18 managed entirely with TKI, Imatinib: 8 patients, Nilotinib 6 patients and Dasatinib 4 patients. In the

table 1 shows the response to TKI. All our patients are in CHR and patients who failed to show the MMR is because we do not have the results of its monitoring PCR studies, so far only one case showed a suboptimal response to MMR handled in a patient with imatinib was modified so that a second generation TKI with adequate results. It is important to note that the last 19 cases detected since 2003, 17 were undergoing treatment and only one patient who debuted in blastic phase of the disease has died, the other case no longer follows the controls are presented in chronic phase and subsequently lost of the medical consultation. **Conclusions:** The response found on the TKI has been good, and patients who have not tolerated any treatment they have adapted to some of the other options TKI. We are pending completion of PCR studies on follow-up of our cases and mutational studies in patients who develop resistance to a TKI.

	Beginning of treatment	CHR	MMR	CMR
Imatinib	1	4	3	0
Nilotinib		5	1	0
Dasatinib	1	1	1	1

CHR: complete hematologic response; MMR: major molecular response; CMR: complete molecular response.

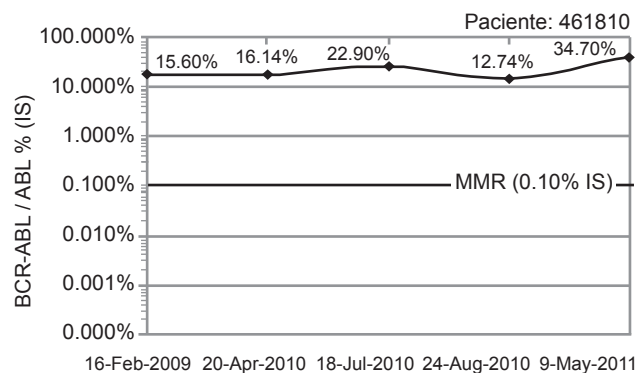
A1171

NILOTINIB IN PH+ CML PATIENTS RESISTANT TO IMATINIB (IM) IN A TERTIARY CARE CENTER IN GUA-DALAJARA, MEXICO. Aguilar-Luna JC. Hospital "Valentín Gómez Farías" ISSSTE Zapopan Jal. MX.

Introduction: Nilotinib is a second generation TKI inhibitor that is highly selective for Bcr-Abl. Nilotinib is approved as second line therapy in Mexico since 2007. Lack of achievement of Complete Cytogenetic response (CCyR) and Major Molecular Response (MMR) with IM is associated to a poor prognosis. Second generation TKIs including nilotinib are capable of reverting this dismal prognosis and delay progression in IM resistant patients. **Objectives:** We report 4 clinical cases of Ph+ CML resistant to IM that started therapy with nilotinib at our institution. **Material and Methods:** Case 1: a 44 year old female, with CML diagnosis since January 2005, she had received IM including high doses (800 mg/day). She developed clonal evolution with and additional traslocation 1;20 in Ph+ cells. In March 2009 we began treatment with nilotinib 800 mg/day, the additional chromosomal abnormalities disappeared and % of BCR-ABL also declined. Case 2: A 34 year old male with CML since October 2002. He was treated during 9 years with IM 600 mg/day. The molecular monitoring showed a increase of Bcr-Abl transcripts and for this reason we changed therapy to nilotinib 800 mg/day in April 2011. Case 3: A 57 year old male with CML since september 2006, treated with IM 800 mg/day until october 2010, when we started therapy with nilotinib 800 mg/day, despite this treatment the previously progressive increase

of %Bcr-Abl did not show a decrease suggesting probably the presence of a Bcr-Abl mutation. Case 4: A 29 year old female with CML since April 2006. Treated with high doses of IM (600 mg/day) until April 2011. She had loss of major molecular response despite high doses of IM, then she was treated with nilotinib 800 mg/day since April 2011. The last level of Bcr-Abl was 16.3% in the international scale. **Results:** 1. The patient tolerates well nilotinib after 2 years of therapy (fig1), 2. We will monitor the response with qRT-PCR for bcr-abl. 3. This patient did not have an optimal response to nilotinib. 4. We are expecting the report of the follow-up in order to know if remission was achieved. **Conclusions:** There is a considerable proportion of patients that are primary resistant or have loss of response to IM. Nilotinib in our experience is an effective therapy in most cases treated in our center. It is very important to have a optimized monitoring with qRT-PCR.

Resultados acumulados muestran el más reciente 6 visitas con resultados válidos. Por favor, consulte los informes anteriores de los resultados de las anteriores visitas no se muestra.



A1178

CHRONIC MYELOID LEUKEMIA: MOLECULAR MONITORING AT THE NATIONAL CANCER INSTITUTE. Limon-Rivas DA, Arcos-Fonseca DM, Cruz-Velazquez J, Chavez-Jacal MS, Cervera-Ceballos EE. Instituto Nacional de Cancerología (INCan).

Introduction: Chronic Myeloid Leukemia (CML) represents 15% of adulthood leukemias with an incidence rate of 1-2 per 100.000 persons. The main goal of treatment is to reduce white blood cells and the elimination of Philadelphia chromosome Ph (juxtapositioning the Abl1 gene on chromosome 9 to a part of the BCR gene on chromosome 22) using tyrosin-kinase inhibitors such as imatinib mesylate (Gleevec). The criteria of treatment efficiency are based on: a complete cytogenetic response (Ph chromosome undetectable) follow by quantification of BCR/ABL transcripts using reverse transcription polymerase chain reaction (RT-PCR). Recent studies realized after Imatinib appearance, have demonstrated the need of a molecular monitoring by

quantitative methods once a cytogenetic response. **Objectives:** In Mexico, The National Cancer Institute is one of the main centers with a CML population, thus the standardization and Implementation of a molecular monitoring method is necessary to give a follow-up of those patients with a complete cytogenetic response. **Material and Methods:** Obtention of blood samples in 4 mL Vacutainer tubes with 7.2 mg EDTA. Isolation of Mononuclear cells from human peripheral blood using Ficoll Nucleic Acids extraction using TRIZOL, RNA quantification with Nano Drop (THERMO), Real time PCR for quantitative RT-PCR Analysis of BCR-ABL following manufacturer's instructions (MolecularMD). Analysis and results report following the recommendations of Molecular MD Kit for International Scale (IS) report. Switch Samples with the reference laboratory in Australia for the validation and certification of this laboratory in Mexico for the incorporation to the IS. **Results:** The standardization of cells isolation, nucleic acids extraction and qRT-PCR. The analysis and report of 100 samples of CML divided in: 34% patients without BCR/ABL transcripts detection, 33 % patients in remission with a mayor molecular response (MMR, less or equal to 0.1) and 33% patients without a MMR (more than 0.1). **Conclusions:** It is important to standardize fusion genes such as PML/RARA, AML1/ETO, TEL/AML1, etc involved in major hematological disorders. It is also essential to give the patient the best care in the diagnosis.

A1179

CYTOGENETIC FINDINGS IN CHRONIC MYELOMONOCYTIC LEUKEMIA. PRACTICE TEN YEARS AT THE NATIONAL CANCER INSTITUTE. Arcos-Fonseca DM, Cruz-Velazquez J, Chavez-Jacal MS, Espinoza-Zamora JR, Diaz-Vargas G, Cervera-Ceballos EE. National Cancer Institute, Mexico.

Introduction: Chronic Myelomonocytic Leukemia (CMML) has features both of myelodysplasia and myeloproliferation. In the WHO classification it has migrated from the MDS category into the MPD/MDS disorders. To differentiate CMML from atypical CML, evaluation of the proportion of granulocytic precursors and the blood film to identify anormal monocytes are useful. Cytogenetic abnormalities are common (20-50%) but again none are specifically associated with CMML. Those patients with abnormal karyotypes are more likely to have advanced disease and to transform to acute leukemia. Monosomy 7 is common, as are +8, der(12p) presenting as a terminal deletion or translocation, -Y. Abnormalities of chromosome 5 (-5, 5q, t(5;12)(q31p13)) are less common in CMML than in myelodysplasia. And the other hand, juvenile myelomonocytic leukemia occurs in children less than 5 years old and encompasses conditions previously termed "juvenile chronic myeloid leukemia", infantile monosomy 7 syndrome, and other myelodysplastic/myeloproliferative diseases of childhood. This is particularly associated with monosomy 7. In approximately 25-33%, other cytogenetic abnormalities (+8 and abnormalities of chromosome 7) also occur. In common with

CMML, karyotypic abnormalities are frequent in atypical CML, occurring in 30-80%, +8 being the most frequent. Interestingly, a fusion gene between PDGFRB and H4 has been reported en at least one patient. **Objectives:** Cytogenetic aberrations were identified in CMML reported from 2000 to 2010 at the National Cancer Institute. **Material and Methods:** We obtained the results of requests made to the cytogenetics laboratory with a diagnosis of LMMC and epidemiological analysis was performed and the frequency of cytogenetic abnormalities reported in the group. **Results:** 12 samples were identified, of whom 33% are male, presenting an average of 55 years and an age range between 33 and 77 years. The complex karyotype is mostly found cytogenetic abnormalities (involving the (1q34), del (6p22), del (7q32) and del (11p15), and the most frequent numerical alteration monosomy of chromosome 5) occurring in 14%. **Conclusions:** Conventional cytogenetic analysis is an important tool at diagnosis and disease monitoring, however, techniques such as fluorescence in situ hybridization and reaction polymerase chain currently are indispensable for the detection, characterization and monitoring disease, especially in cases where it is not possible to obtain good quality material for conventional cytogenetic analysis.

A1182

CHRONIC LYMPHOCYTIC LEUKEMIA (CLL). A RETROSPECTIVE STUDY OF 10 YEARS AT THE NATIONAL CANCER INSTITUTE. Arcos-Fonseca DM, Cruz-Velazquez J, Chavez-Jacal MS, Espinoza-Zamora JR, Diaz-Vargas G, Cervera-Ceballos EE. National Cancer Institute, Mexico.

Introduction: Chronic lymphocytic leukemia (CLL) is characterized by accumulation of monoclonal malignant B cells in blood, lymph nodes, liver, spleen, and bone marrow. CLL is a disease of a subtype of mature B cells characterized by expression of a specific combination of cell surface molecules and surface immunoglobulin. CLL is the most common adult form of leukemia in Western society. The median age at diagnosis between 1997 and 2001 was 72 years: for males 70 years and 74 year of age for females. Despite the morphological homogeneity, the disease varies enormously in prognosis, with some patients requiring no treatment for many years, if even, while others die rapidly with chemotherapy resistant disease. Karyotypic analyses, including standard metaphase chromosome analysis and fluorescence in situ hybridization (FISH), have also been key in identifying subgroups of patients with B-Cell lymphoproliferative diseases. Within molecular cytogenetics analysis, progress has been hampered by the lack of a consistent cytogenetic lesion. The most common abnormality, involving deletion of a small region of chromosome 13q14. Similarly, patients with deletions and mutations involving either the p53 gene on chromosome 17p13.3 or the ATM gene on 11q23 fare badly. Determining the nature of the molecular events associated with these different sub-groups is now a major challenge. **Objective:** Identify the most common cytogenetic abnormalities from 2000 to 2010, and the frequency of cytogenetic abnormalities reported in the group.

Material and Methods: They obtained the results of requests made to the cytogenetics laboratory with a diagnosis of CLL and epidemiological analysis was performed and frequency of cytogenetic abnormalities. **Results:** 36 samples of which 61% are male; the average age is 53 years with an age range that moves from 25 to 84 years. The most common cytogenetic abnormalities is complex karyotype (11%), with a history of that 67% of the samples was not possible to perform chromosome analysis was performed but the FISH technique to identify key markers LLC. **Conclusions:** Although conventional cytogenetic analysis (CCA) should not be omitted in the diagnosis and disease monitoring techniques such as fluorescence in situ hybridization (FISH) and chain reaction (PCR) have become effective tools and essential in the detection, characterization and monitoring of the disease, especially in those cases where you do not get optimal cell growth.

A1189

REPORT OF THE FIRST STANDARDIZATION OF QRT-PCR FOR BCR-ABL IN CHRONIC MYELOID LEUKEMIA (CML) IN MEXICO. Meillón-García L, Delgado-Lopez N, Sandoval C, Nacho-Vargas K. Hospital de Especialidades Centro Médico Nacional SXXI IMSS

Introduction: Current therapy with tyrosine kinase inhibitors (TKIs) have improved the management of CML. The TKIs are capable of inducing deep responses including Major Molecular Response (MMR) and Complete Molecular Response (CMR). These responses should be measured through molecular techniques including the quantitative real-time polymerase chain reaction (qRT-PCR), according to the new International Scale (IS) expressed in percentage of the ratio of BCR-ABL/ABL. **Objectives:** To describe the steps required for achieving the first standardization of qRT-PCR for Bcr-Abl at the Laboratory of Hematology of the Specialties Hospital at the Instituto Mexicano del Seguro Social in Mexico City. **Material and Methods:** In 2010 and until May 2011 we collected peripheral blood samples in tubes with EDTA. A lysis buffer for red cells was used and white cells were stabilized with trizol. RNA was obtained by precipitation with phenol-chloroform. RNA quantification were performed with a kit of Molecular MD One –Step qRT-PCR BCR-ABL (1101-01 Portland OR, USA), Taqman primers for using with Bcr-Abl were directed from exon 13 of BCR to exon 2 of ABL; for internal control the interval between exon 3 and exon 4 of ABL was used. RNA samples were quantified by the spectrophotometer (Nanodrop 2000) and were processed twice in the RT-PCR equipment Step–One (Applied Biosystems). The sensitivity of the calibration curve was established by a series of dilutions of plasmids using from 10⁻⁵ to 10⁻¹. The value of IS was established by the ratio of % of BCR-ABL /ABL. Conversion factor was 0.81 that was adjusted with Molecular MD. This method allows the establishment of Major Molecular Response since detects at least 3 logs of reduction, initially reported by

IRIS investigators (0.10% of BCR-ABL in the IS). **Results:** A first proof of achievement was obtained in May 2011, since a rate of 97% of concordance was reported in the sample exchange between Molecular MD (Oregon, Portland) and Mexico city Labs. **Conclusions:** We report the first standardization of qRT-PCR for Bcr-Abl. This new standardized tool is a first step for having a harmonized report for diagnosis and monitoring in PB samples of CML patients in Mexico using the IS. We will continue with sample exchanging with Adelaida (Australia) laboratory in order to have an additional validation of the Conversion Factor.

A1211

MOLECULAR STATUS IN A COHORT OF PATIENTS WITH CHRONIC MYELOID LEUKEMIA AT THE INSTITUTO MEXICANO DEL SEGURO SOCIAL (IMSS). Delgado-Lopez N, Ayala-Sanchez M, Sandoval-Sanchez C, Meillón-García L, Lugo Y, Vega P, Romo A, Nacho-Vargas K. Hospital Especialidades CMN SXXI IMSS, Hospital de Especialidades CMN La Raza, IMSS. HGR1 Carlos McGregor Sanchez Navarro, IMSS. UMAE León, Guanajuato IMSS de Leon y Hospital de Guadalupe IMSS.

Introduction: With the recent standardization of a laboratory of reference of the Hospital de Especialidades of the National Medical Center Siglo XXI (IMSS) an increased number of tests were requested to our center in order to have the results in percentage of Bcr-Abl/AbI according to the new international scale. To have this information is critical since it has therapeutic implications (continuing with the same TKI or changing to an alternate TKI). **Objectives:** To report the molecular status of 274 patients with CML attended at the Instituto Mexicano del Seguro Social. **Material and Methods:** From August 2011 to December 2011, peripheral blood samples of 276 patients were sent to our laboratory. We extracted RNA as previously described. We asked to hospitals have at least 5 ml of blood in order to maximize optimal results. Then we put these results in our data base and classified patient samples in five groups according the percentage of Bcr-Abl/AbI in the international scale: The first group were patients with >10% of Bcr-Abl; the second group were patients with >1-10% of Bcr-Abl; the third group were patients with >0.1-1%; the fourth group were patients with 0.1% and less than 0.1 and the fifth group were patients with undetectable Bcr-Abl transcripts. **Results:** We found the following distribution: Group I (> 10% Bcr-Abl): 47 patients (17.15%); Group II (>1-10%): 36 patients (13.13%); Group III (>0.1-1% Bcr-Abl) 50 patients (18.24%); Group IV (0.1% and less than 0.1% of Bcr-Abl) 84 patients (30.65%); and Group V: undetectable Bcr-Abl: 57 patients (20.83%). **Conclusions:** At least 50% of patients treated with imatinib, nilotinib or dasatinib have reached lower levels of minimal residual disease. This information is very useful for clinician and should be interpreted individually according the time of treatment with TKIs for having adherence to the international

and ELN recommendations. While levels of more than 10% could be related to the diagnosis of the disease if this value is found after 12 months of therapy it could be related to treatment failure or relapse. By other hand; Bcr-Abl levels so low as 0.1% or less are associated with MMR and undetectable is related to a complete molecular response. To have systematized information will allow to have optimized therapies at our institution.

A1226

NILOTINIB IN PATIENTS WITH CHRONIC MYELOID LEUKEMIA (CML) THAT HAVE LOSS OF RESPONSE AND HAVE PROGRESSION WITH IMATINIB OR HAVE INTOLERANCE. REPORT OF THE CLINICAL EXPERIENCE IN A TERTIARY CENTER OF THE INSTITUTO MEXICANO DEL SEGURO SOCIAL IN MONTERREY, MEXICO. Báez de la-Fuente E. UMAE # 25 IMSS Monterrey, N.L.

Introduction: After a decade of treatment of CML with imatinib the results are impressive. Nevertheless the imatinib resistance and intolerance are frequent situations in clinical practice, that worsens the prognosis of no responders or intolerant patients to imatinib. Alternative therapies are needed for this situation. The recent introduction of nilotinib a selective Bcr-Abl inhibitor to the treatment of CML can offer to these group of patients a good therapeutic choice for controlling the disease without need of dose escalation of imatinib. **Objective:** To report the outcomes of a group of patients with CML treated with nilotinib as second line of treatment. **Material and Methods:** We reviewed clinical files of 16 patients with CML that were treated with nilotinib 800 mg/day at our institution. Ten patients were male and 6 were female. Age interval 24 to 75 years old. All of them were previously treated with imatinib 400 mg/day. Seven of 16 patients have had doses escalation to 600 mg/day of imatinib and only 1 of 16 have had dose escalation to 800 mg/day. HU and Peg-IFN was also added in some cases. The main reason for switching therapy to nilotinib was resistance and progression of the disease in 14 cases and intolerance to imatinib in 3 cases. Nilotinib have availability in our institution since August 2009. **Results:** All the patients had complete hematologic response. Two patients with intolerance to imatinib have had clinical improvement. One patient progressed to blastic phase and developed CNS bleeding and died. Four patients achieved Major Molecular Response (MMR). Two patients had only slight decrease of Bcr-ABL (2 log red) One patient had progressive increase in the % of transcripts of Bcr-Abl, subsequently a mutational analysis showed duplication of exons 4 and 9 of ABL gene.

This patient required combination of nilotinib +pegylated IFN. Three patients did not achieve MMR. Other 3 reports will be presented at the meeting. **Conclusions:** Nilotinib is a highly effective therapy in imatinib resistant and intolerant patients. Molecular monitoring is imperative in all the cases for follow-up and mutational analysis is required if patient develops resistance to a 2nd gen TKI

A1236

FREQUENCY ANALYSIS OF MULTIDRUG RESISTANCE-1 GENE (MDR-1) IN PATIENTS WITH CHRONIC MYELOID LEUKEMIA AND ACUTE LYMPHOBLASTIC. EXPERIENCE IN GENERAL HOSPITAL OF MEXICO. Martínez-Tovar A*, Olarte-Carrillo I*,***, Salazar-Terán I.V***, Ramos-Peñañiel C****,5, Mendoza-Salas I****, Zamora-Domínguez J*, Rozen Etta F⁵, Collazo-Jaloma J⁵, Gutiérrez-Romero M⁵, Kassack-Ipiña JJ⁵, Martínez-Murillo C⁵, Miranda-Peralta EI*, Nacho-Vargas KA⁶. *Laboratorio de Biología Molecular, Hospital General de México; **Benemérita Universidad Autónoma de Puebla, Facultad de Ciencias Químicas; ***Posgrado en Ciencias Biológicas UNAM; ****Escuela Nacional de Ciencias Biológicas-IPN; ⁵Servicio de Hematología U-103, ⁶Novartis Oncología México.

Introduction: In recent years the use of targeted therapies such as tyrosine kinase inhibitors (TKI) have improved the prognosis of chronic myeloid leukemia (CML) by blocking the oncogenic protein responsible for activating the process of proliferation and inhibition of apoptosis (BCR-ABL). However, in some cases patients are resistant to treatment. Resistance mechanisms described can be of two types of mutations in the catalytic region of BCR-ABL or overexpression of multidrug resistance genes (MDR-1). But unknown frequency of expression of MDR-1 gene in patients with CML and LAL General Hospital of Mexico. **Objectives:** To analyze the frequency of expression of MDR-1 gene in patients with CML and ALL. **Material and Methods:** We analyzed 149 samples of leukemia patients of which 60 were classified as novo CML, 19 CML subsequent and 70 ALL. We examined the expression of MDR-1 by RT-PCR in fresh leukemia cells. **Results.** The results show a frequency of expression of the gene MDR-1 from 43% in the case of LMC novo and 69% in the subsequent LMC. In the case of LAL the frequency of expression of the gene MDR-1 was 35.7%. **Conclusions.** Early detection of MDR-1 genes in leukemia will predict the failure to have better treatments and therapeutic strategies in these hematologic diseases. This project is supported by the Pharmaceutical Industry, Oncology Division (Novartis). Conacyt health-2011-1-162269.

LYMPHOMAS AND MYELOMAS

ORAL PRESENTATION

A1069

SERUM BIOMARKERS DETECTED BY PROTEOMIC APPROACH IN PATIENT INVOLVED BY DIFFUSE LARGE B CELL LYMPHOMA. Villela L*, Patena S*, Winkler R**, Scott S*, Bolanos-Meade J***, Fajardo O*, Rojo R*, Hernandez-Hernandez JA*, Guzman E*, Perfecto Y*, Gutierrez MA*, Chacolla R*, Baltazar S****, Hernandez R****, Delgadillo LF⁵, Encarnacion S⁵. *Instituto Tecnológico y Estudios Superiores de Monterrey Campus Monterrey, Escuela de Medicina y Ciencias de la Salud, Centro de Innovación y Transferencia en Salud. Catedra de hematología y Cancer. Tec Salud; **CINVESTAV. Unidad Irapuato; ***Sidney Kimmel Comprehensive Cancer Centre at Johns Hopkins; ****UMAE Clínica 25 del Instituto Mexicano del Seguro Social; ⁵UNAM. Centro de Ciencias Genómicas. Cuernavaca, Morelos.

Introduction: Diffuse Large B-Cell Lymphoma (DLBCL) is the most frequent subtype of Non-Hodgkin Lymphoma in the world (35%). Despite of chemotherapy with R-CHOP, patients with DLBCL continue to relapse or have refractory disease after treatment. For this reason, the search for biomarkers that could predict complete response (CR) and survival is important. Proteomics is in charge of the identification, quantification and characterization of the proteins in tumor cells. There are some descriptions about the proteomic profile of lymphomatous cells, but there are not any reports about the proteomic profile in serum in patients with DLBCL. **Objective:** We present the preliminary results from the serum profile using proteomic technology in such patients. **Material and Methods:** The pre-treatment serum from 18 patients with diagnosis of de novo DLBCL who were treated with R-CHOP and 8 healthy people (control cases) were analyzed. The technology Proteomizer® was used. Proteins were separated in double dimension gels (2D-SDS-PAGE at 12%) and stained with Coumassi/silver. Spot lecture was done by the software PDQuest. The potential spots were cut and analyzed by Liquid Chromatography Mass Spectrometry. Clinical variables were also analyzed together with the proteomic profile results. **Results:** There were 8 proteins identified with differential expression in the serum of patients with DLBCL, 2 inflammatory (amyloid A and P) and 1 antioxidant (GPX3). GPX3 was the most

frequent protein observed compared with the control group (72% vs 12%, respectively p=0.009). This protein presented a sensitivity of 72% (CI95%: 49% to 85%), specificity of 87.5% (CI95%: 63-98%) with a PPV of 93% (CI95%: 68% to 99%) and NPV of 58% (CI95%: 32 to 81%). The clinical characteristics of patients with GPX3 (+) compared with the GPX3 (-) were the followings: More frequent in males (54% vs 20%, respectively), older age, elevated LDH (64% vs 40%, respectively), >1 ECOG (23% vs 0%,), advanced IPI score (intermediate and high; 46% vs 20% and 15.4% vs 0%, respectively) all of them p=N.S. Regarding overall rate response (ORR), that was higher in patients with GPX3 (+) vs. GPX3 (-). (100% vs. 66%, respectively p=0.07). **Conclusions:** The proteomic profile at the serum level from patients with DLBCL has an inflammatory and antioxidant component in serum. The GPX3 was associated with variables of poor prognosis for DLBCL. GPX3 showed to be a potential biomarker for predicting response. Therefore, in our group we are evaluating the role of plasma GPX3 by ELISA in patients with DLBCL to attempt corroborate these data.

A1103

BAICALIN DOWN-REGULATES THE PI3K/AKT SIGNALING PATHWAY AND INDUCES APOPTOSIS IN BURKITT LYMPHOMA CELLS. Hu J, Huang Y, Zheng J, Li J, Wei T, Zheng Z, Chen Y. Fujian Institute of Hematology, Fujian Provincial Key Laboratory of Hematology, Fujian Medical University Union Hospital, Fujian, China.

Introduction: The phosphatidylinositol-3-kinase (PI3K)/serine/threonine kinase (Akt) signaling pathway is essential to the survival and proliferation of human cells, and constitutive activation of this pathway is thought to play a critical role in the progression of human hematologic malignancies. The CA46 lymphoma cell line [12], which was derived from the ascites fluid of a patient with American-type Burkitt lymphoma, carries the (8;14) translocation, overexpresses Bcl-2 and c-myc mRNAs, and has been proven a useful model of Burkitt lymphoma. Baicalin, a flavone present in *Scutellaria baicalensis* Georgi, inhibits the growth of human leukemia and myeloma cells through induction of apoptosis. **Objectives:** The present study was undertaken to ascertain whether baicalin also suppresses the growth of Burkitt lymphoma cells and to explore potential mechanisms through

which such suppression might occur. **Material and methods:** MTT and colony formation assay were used to evaluate cell proliferation. Apoptosis in response to baicalin treatment was detected with Annexin V-FITC/PI. Western blot was performed to detect the changes in the expression of the related proteins. **Results:** Treatment of cultured CA46 Burkitt lymphoma cells with baicalin for 48 h markedly decreased the rate of cell proliferation; an IC50 value of 10 μ M was obtained. Colony formation was almost fully suppressed at 10 μ M baicalin. CA46 cells underwent apoptosis in response to baicalin treatment as evidenced by an increase in the percentage of cells stained with Annexin V-FITC/PI, by increased DNA fragmentation, and by activation of the intrinsic mitochondrial pathway for cell death as characterized by increased expression of cleaved caspase-9, cleaved caspase-3, and cleaved poly(ADP-ribose) polymerase. Additionally, baicalin was found to down-regulate the phosphatidylinositol-3-kinase (PI3K)/serine/threonine kinase (Akt) signaling pathway. thought to play a critical role in the progression of human hematologic malignancies. **Conclusions:** The concentrations at which baicalin downregulate PI3/Akt pathway in CA46 cells were comparable to those that suppressed growth and induced apoptosis, supporting the hypothesis that the observed growth-inhibitory and apoptosis-inducing actions of baicalin in these cells are mediated by down-regulation of this pathway.

A1134

SECOND PRIMARY CANCERS IN PATIENTS WITH NEWLY DIAGNOSED MULTIPLE MYELOMA AND MONOCLONAL GAMMOPATHY OF UNKNOWN SIGNIFICANCE DETECTED BY 18F-FDG PET/CT. Bacovsky J*, Myslivecek M**, Koranda P**, Scudla V*, Minarik J*, Pika T*, Buriankova E**. *3rd Clinic of Internal Medicine, Medical Faculty and University Hospital Olomouc, Czech Republic; **Clinic of Nuclear Medicine, Medical Faculty and University Hospital Olomouc, Czech Republic. Medical Faculty and University Hospital Olomouc, Czech Republic.

Introduction: The occurrence of second primary cancers in myeloma patients has been recognized for several years. Newer imaging modalities, such as 18F-FDG PET/CT, have been recently introduced for staging to assess the activity and extent of disease in patients with multiple myeloma (MM) and gammopathy of undetermined significance (MGUS). **Objective:** The exact frequency of PET/CT incidental detection of additional primary malignancies during evaluation of MM is not well known. Purpose of this study was to evaluate the value of integrated whole-body positron emission tomography and computed tomography (PET/CT) in detecting of second primary cancer at the time of diagnosis and initial staging of Multiple myeloma and MGUS. **Material and methods:** A total of 170 patients with multiple myeloma (MM) and 66 patients with MGUS were enrolled in the study. Median age of MM patient was 68 years (32–86) and M/F ratio 1,0. In I. st. according Durie Salmon

staging system were 12 %, in II st. 35 %, in III st. 53 %, IPI I st. 40%, II.st. 30%, III.st. 30%. Median age of MGUS patients was 67 years (31–84) and M/F ratio 0,8. All patients underwent 18F-FDG PET/CT examination at the time of initial staging for MM or MGUS before any therapy. Routine diagnostic tests including biochemistry, conventional radiography and ultrasound examination were done. Patients were without clinical signs indicating other malignancy. **Results:** A total of 9 second primary malignant tumors were identified in group of 170 MM patients (5 %). 18F-FDG PET/CT found focal lesions indicative of a second primary cancer, which were not detected by routine examination and staging of MM. Second primary cancers included: 3 patients with carcinoma of thyroid gland, 3 patients with carcinoma of colon, 1 patient with carcinoma of breast, 1 patient with carcinoma of lung, 1 patient with lymphoma. In group of 66 patients with MGUS were no other primary cancers detected. All patients were successfully treated and because their disease was diagnosed early. **Conclusions:** 18F-FDG PET/CT at the time of the initial staging is useful for screening a second primary cancer with a high sensitivity. Second clinically asymptomatic cancers were detected in 5% of MM patients. All these unexpectedly detected cancers were successfully treated. Supported by grant IGA MZ CR NT 12215-4/2011.

A1150

PX-171-003-A1, AN OPEN-LABEL, SINGLE-ARM PHASE 2 STUDY OF CARFILZOMIB IN PATIENTS WITH RELAPSED AND REFRACTORY MULTIPLE MYELOMA: LONG-TERM FOLLOW-UP AND SUBGROUP ANALYSIS. Martin T*, Singhal S**, Wang M***, Vij R****, Jakubowiak⁵ A, Jagannath S⁶, Lonial S⁷, Kukreti V⁸, Bahlis N⁹, Alsina M¹⁰, Chanan-Khan A¹¹, Buadi F¹², Reu F¹³, Somlo G¹⁴, Zonder J¹⁵, Song K¹⁶, Stewart K¹⁷, Stadtmayer E¹⁸, Kunkel L¹⁹, Rajangam K²⁰, Wear S²¹, Orłowski R²², Siegel D²³. *University of California San Francisco, San Francisco, CA; **Northwestern University School of Medicine, Chicago, IL; ***MD Anderson Cancer Center, Houston, TX; ****Washington University School of Medicine, St. Louis, MO; ⁵University of Chicago Medical Center, Chicago, IL; ⁶Mount Sinai Medical Center, New York, NY; ⁷Winship Cancer Institute, Emory University School of Medicine, Atlanta, GA; ⁸Princess Margaret Hospital, Toronto, ON, Canada; ⁹Tom Baker Cancer Centre, University of Calgary, Calgary, AB, Canada; ¹⁰H. Lee Moffitt Cancer Center, University of South Florida, Tampa, FL; ¹¹Roswell Park Cancer Institute, Buffalo, NY; ¹²Mayo Clinic, Rochester, MN; ¹³Taussig Cancer Center, Cleveland Clinic; ¹⁴City of Hope National Medical Center; Karmanos Cancer Institute/Wayne State University, Detroit, MI; ¹⁵University of British Columbia, Vancouver, Canada; Mayo Clinic, Scottsdale, AZ; ¹⁶University of Pennsylvania, Philadelphia, PA; ¹⁷Independent Consultant, San Francisco, CA; ¹⁸Onyx Pharmaceuticals, South San Francisco, CA; ¹⁹The Multiple Myeloma Research Consortium, Norwalk, CT; ²⁰MD Anderson Cancer Center, Houston, TX; ²¹John Theurer Cancer Center, Hackensack, NJ.

Introduction: Carfilzomib is a next-generation, selective proteasome inhibitor in development for treatment of multiple myeloma (MM). Single-agent carfilzomib has shown durable activity in patients with relapsed and/or refractory MM. PX-171-003-A1 was an open-label, single-arm phase 2b trial in patients with advanced refractory MM. **Objective:** ---. **Material and methods:** Patients received greater than = 2 prior therapies (bortezomib, either thalidomide or lenalidomide, an alkylator, and an anthracycline), were responsive to at least 1 prior therapy, and were refractory to their most recent regimen. Carfilzomib was given on days 1, 2, 8, 9, 15, 16 of 28-day cycles (C), (20 mg/m² in C1; 27 mg/m² in C2–12). Primary endpoint was overall response rate (ORR). Secondary endpoints included clinical benefit response (CBR), duration of response (DOR), overall survival (OS), and safety. Cytogenetics, peripheral neuropathy (PN), International Staging System (ISS), and prior treatment data were collected for subset analyses. Responses were assessed per IMWG and EBMT criteria and adjudicated by an Independent Review Committee. **Results:** The ORR was 23.7% with median DOR of 7.8 months. 71 of 229 pts had greater than or = 1 cytogenetic abnormality and achieved an ORR of 29.6%, with median DOR of 6.9 months. Median OS for all patients was 15.5 months. The most common treatment-emergent adverse events (AEs) greater than or = to G3 regardless of relationship to study drug were predominantly hematologic and included thrombocytopenia (28.9%), anemia (23.7%), lymphopenia (19.5%), and neutropenia (10.9%). New-onset PN was infrequent (12.4%) and PN greater than or = to G3 was reported in only 2 patients (0.8%). 38 patients completed 12C and continued on extension protocol PX-171-010. **Conclusions:** Single-agent carfilzomib achieved significant durable responses in patients with R/R MM relapsing after all available therapies including bortezomib and immunomodulatory agents, including those with unfavorable cytogenetics and advanced disease. Carfilzomib was well-tolerated, and AEs were clinically manageable with no new, unexpected, or cumulative toxicities, allowing prolonged dosing for disease control. Importantly, exacerbation of pre-existing PN was uncommon. The CBR and median DOR achieved with this steroid-sparing regimen establish the potential of carfilzomib to offer substantial clinical benefit to patients with relapsed or refractory disease. These results provided the rationale for 2 subsequent phase 3 trials, ASPIRE (evaluating CRd vs Rd) and FOCUS (examining carfilzomib vs best supportive care).

Baseline	Total n	≥PR, %	≥MR, %
Overall	257	23.7	37.0
Prior therapies			
<5	110	24.5	35.5
≥5	147	23.1	38.1
Cytogenetics			
Normal/favorable	158	22.8	39.9
Unfavorable	71	29.6	33.8
ISS stage			
I	76	31.6	48.7
II	96	24.0	36.5
III	78	17.9	28.2

A1154

LYMPHOCYTES SUB-POPULATION DETERMINATION IN DE NOVO MULTIPLE MYELOMA PATIENTS. Rodriguez-Gonzalez MG*, Vela-Ojeda J*, Reyes-Maldonado E**, Montiel-Cervantes L*, Xolotl-Castillo M*, Tripp-Villanueva JF*, Perez-Retigun F*, Padilla-Guerrero G*, Romero-Salas T***. *UMAE HE CMN La Raza IMSS; **Escuela de Ciencias Quimico-Biologicas, Instituto Politecnico Nacional; ***Novartis Oncology, Mexico DF.

Introduction: Multiple myeloma (MM) is a monoclonal gammopathy characterized by an antitumoral immune dysfunction. **Objectives:** To determine lymphocyte subsets (B, T, NK, NKT, dendritic cells, gamma delta T cells, and regulatory T cells) in newly diagnosed patients with MM and to correlate the number of these cells with overall survival. **Material and Methods:** This prospective, descriptive, and comparative study was performed in 68 patients with newly diagnosed MM and 113 healthy donors. Diagnosis of MM was based on International criteria. A 5 ml peripheral blood sample was obtained from patients and healthy donors for flow cytometry analysis. Mononuclear cells were quantified and adjusted to 1x10⁶/100 microL. Lymphocyte subsets were quantified using a FACScalibur cytometer with specific antibodies directed against CD3, CD4, CD8, CD19, HLA-DR, Lyn, CD11C, CD123, CD16-56, CD25, FoxP3, va24vbeta11TCR, and pan-gamma-delta. Because the low numbers of some lymphocytes, 20 000 to 500 000 events were acquired during flow cytometry. The same determinations were performed after six months of treatment in patients with MM. The number of lymphocyte subsets were compared between healthy donors and de novo MM patients, and between basal and six months treatment in MM patients using an independent samples T test and paired T test, respectively. Overall survival and disease free survival was evaluated by Kaplan-Meier curves. **Results:** The median age of MM patients was 59.5 years, 35 (51.5%) were male and 33 (48.5%) female. IgG myeloma was present in 53% of the patients and 82% of them were in Durie-Salmon stage III. Pre-treatment values of lymphocyte subsets in MM patients were lower comparing with healthy donors (total lymphocytes p<0.001, CD3+ p<0.006, CD4+ p<0.00001, NK cells p<0.00001, type 2 dendritic cells p<0.007, and CD19 p<0.01). After six months of treatment, there was a decrease in total lymphocyte counts (p<0.0002), CD4+ (p<0.0004), CD8+ (p<0.001), CD3+ (p<0.001), and NKT cells (p<0.02). By contrast, we observed an increase in type 1 dendritic cells (p<0.009), type 2 dendritic cells (p<0.01), and Treg cells (p<0.008) after six months of treatment. Median overall survival was 22.4 months. **Conclusions:** There is a lower number of CD4+ lymphocytes, NK cells, type 2 dendritic cells and B cells in MM patients when compared with healthy donors. After six months of treatment, there is an increase in dendritic cells and Treg cells, but some lymphocyte subsets (CD4, CD8, and NKT cells) remained at lower levels, a phenomenon that can be of clinical interest.

POSTERS

A1001

CARDIOTOXICITY FOLLOWING CYCLOPHOSPHAMIDE THERAPY. Atalay F*, Gulmez O**, Ozsanca A***, Kizilkilic E*. Baskent University Istanbul Medical and Research Center * Department of Hematology; ** Department of Cardiology; ***Department of Pneumology

Introduction: Cardiac toxicity is one of the life-threatening complications of the cancer therapy. Cyclophosphamide-induced cardiotoxicity is a rare complication and symptoms occur usually within 1-2 weeks. **Objective:** A- 66 year old woman with a previous history of hypertension was admitted to our hospital with the recent diagnosis of Burkitt lymphoma with mediastinal mass. **Material and Methods:** Rituximab and Hyper CVAD chemotherapy protocol was applied to her. **Results:** On the 7 day of the chemotherapy she developed dyspnea. The electrocardiogram showed on that day showed low voltage in limb and precordial leads and echocardiogram showed diffusely increased myocardial echogenicity, mild pericardial effusion, generally impaired biventricular systolic functions with a LVEF 31% and right ventricular mid-apical akinesia, and manifest pleural effusion even though she had both normal biventricular functions before chemotherapy. **Conclusions:** Her medical therapy for the congestive heart failure was planned. Her dyspnea decreased and she was discharged on the 10 day with a LVEF of 37%, and normal right ventricular functions. After one month the second control echocardiography showed normal biventricular functions with a LVEF of %60. Drug induced cardiotoxicity therefore be taken into consideration when using cyclophosphamide therapy, especially when antacyclines coadministered. Close communication between hematologist and cardiologists is required.

A1011

ADHESION OF MULTIPLE MYELOMA CELLS TO FIBRONECTIN IS INFLUENCED BY TOLL-LIKE RECEPTOR-1 TRIGGERING ON THEIR SURFACE. Abdi J*, Mutis T**, Garssen J*, Engels F*, Redegeld F*. *Division of Pharmacology, UIPS, Faculty of Science, Utrecht University, Utrecht, Nederland; ** Dept. of Clinical Chemistry & Hematology, Utrecht Medical Center, Utrecht, Nederland

Introduction: Multiple myeloma (MM) is a fatal lymphoid neoplasm characterized by infiltration in the bone marrow of malignant plasma cells. Adhesion of malignant cells to bone marrow stromal cells and fibronectin (FN) contributes largely to pathogenesis. Although many factors, derived from MM cells or their microenvironment, have been implicated to support and maintain this interaction, no study has to date addressed the effects of external factors including those derived from infection or inflammation. Furthermore, it is well established that myeloma

patients are vulnerable to a variety of infections, and a history of infectious or chronic inflammatory diseases has been reported in some MM patients. Recently, Toll-like receptors (TLRs) have been addressed as candidates that could explain the possible contribution of infection or inflammation to malignancy. In MM, these receptors have displayed a heterogeneous (and higher) expression compared to normal plasma cells, however, their potential role in MM pathogenesis and biology needs further mechanistic research. **Objective:** In an in vitro model system, we sought to explore the effects of TLR-1 activation on MM cells adhesion to fibronectin, so as to gain an insight into the possible contribution of TLR activation to MM pathogenesis. **Material and Methods:** The human myeloma cell lines, Fravel, OPM-1, OPM-2 and NCI-H929, were selected for the study. Expression of TLR-1 and integrins (alpha4, alpha5, beta7, alphaVbeta3) was analyzed by using PCR or FACS. The TLR-1 synthetic ligand, Pam3CSK4, was used for cell stimulation. Adhesion to fibronectin was assessed with fluorometric adhesion assay. To determine the adhesion molecules involved in adhesion to fibronectin, blocking antibodies against relevant integrins were used. Additionally, MyD88 gene knocking down with siRNA and also NFkB pathway blocking (using Bay 11-7082) were performed as confirmatory and complementary experiments. **Results:** All cell lines strongly expressed TLR-1 both at mRNA and protein levels. Alpha4 and alphaVbeta3 (weakly) were detected on all cells and their expression was dose-dependently increased. On the other hand, adhesion to fibronectin decreased for all cell lines in a dose-dependent manner. Interestingly, only beta7 integrin displayed down-regulation in FACS analysis and this effect was found to be TLR-1 and NFkB-mediated. Additionally, blocking experiments showed that beta7 is the mostly involved integrin in adhesion to fibronectin. **Conclusions:** Our results indicate that TLR-1 triggering on MM cells down-regulates their adhesion to FN mediated mostly by beta 7 integrin. This might provide us with a therapeutical targeting implication, on the condition that further research on MM primary cells are performed.

A1022

REGISTRY OF PATIENS WITH MYELOYDYSPLASTIC SYNDROME (MYDYS) IN SLOVAKIA. Mistrík M*, Richterová K*, Valeková L*, Markuljak I**, Guman T***, Palašthy S****, Brabec P⁵. University hospital Bratislava, Slovakia; *University hospital Martin, Slovakia; **Faculty hospital F.D. Roosevelta, Banská Bystrica, Slovakia; ***University hospital Košice, Slovakia; ****Faculty hospital J.A. Reimana, Prešov, Slovakia; ⁵Masaryk University, Brno, Czech Republic.

Introduction: Clinical studies are moving us forward with know how and are transcribed into the guidelines, but mostly they work with more or less selected groups of patients. On the other hand, population based studies are giving us information about

complex process of guideline implementation in daily praxis. They give real data how effectively is health care provided to nonselected group of patients. The purpose of MyDyS project is to collect data of nonselected patients with myelodysplastic syndrome in the region of Slovakia. **Objective:** The purpose of MyDyS project is to collect data of nonselected patients with myelodysplastic syndrome in the region of Slovakia. **Material and Methods:** Material and methods: 5 centers participated on creation and implementation of a simple database covering findings at diagnosis and data about management of patients with diagnosis of myelodysplastic syndrome based on FAB and WHO MDS classification system. It implements IPSS risk estimation as well and enables on line patient registration. **Results:** Results: 130 patients were registered In the years 2009 – 2011 to MyDyS and for this paper 128 patients could be analyzed; male vs female gender: 53,1% vs. 46,9%; average age 64,8 yrs (range 22,0-89,4 yrs). The most frequent diagnosis according to FAB MDS classification was RA (53,9%) and RAEB (21,9%), and according to WHO classification RCMD (27,3%) and RAEB II. (22,7%). Majority of patients suffered from de novo MDS (89,8%). **Conclusions:** Growth factor and transfusion therapy was the most frequent treatment modality, transfusions of red blood cells were given to 35 patients, average 20,7 (range 2-80 RBC units). Deferasirox (mostly 1500mg/ d) was given to 25 patients, and serum ferritin level response was as expected decreased approximately after 8 months in 27,3% patients, after 12 months in 57,4% patients. Chelation therapy was associated with GIT intolerance in 1 case, and skin reaction in 2 patients.

A1028

IN VITRO ANTINEOPLASTIC ACTIVITY OF ALKYLPHOSPHOCHOLINES AND CURCUMIN IN CUTANEOUS T-CELL LYMPHOMAS (CTCL). Yosifov D, Kaloyanov K, Konstantinov S. Laboratory for Experimental Chemotherapy, Dept. of Pharmacology, Pharmacotherapy and Toxicology, Faculty of Pharmacy at the Medical University of Sofia, Bulgaria.

Introduction: CTCLs are a heterogeneous group of non-Hodgkin's lymphomas thought to represent malignancies of skin-homing T cells. The most frequent forms are mycosis fungoides (MF) and Sezary syndrome (SS). Malignant cells in MF reside primarily in skin lesions, while SS is a leukemic form and has an aggressive clinical course. Treatment options have recently expanded with the approval of new agents, but no therapy has been shown to be curative so far. Alkylphosphocholines are a class of membrane-targeted drugs that are actively being developed for treatment of multiple myeloma, chronic lymphocytic leukemia and other hematologic malignancies. Their activity against CTCL has not been studied in detail. Curcumin is a natural substance that is a strong NF- κ B inhibitor and has shown effects in various cancers. **Objective:** The aim of the study was to determine the activity of two alkylphosphocholines (miltefosine and erufosine) and curcumin in CTCL cells (MJ,

MyLa CD4+ and MyLa CD8+, originating from patients with MF, as well as Hut-78 and HH, originating from SS). **Material and Methods:** Cytotoxicity was determined by the MTT-dye reduction assay and oligonucleosomal DNA fragmentation was detected by ELISA. Signal transduction changes were analyzed by Western blotting. **Results:** The IC50 ranged from 0.9 (Hut-78) to 86.4 mcM (HH) for miltefosine and from 5.7 (Hut-78) to 50.0 mcM (HH) for erufosine. The efficacy of curcumin was relatively uniform and the IC50 values ranged from 12.8 (HH) to 21.9 mcM (MJ). Curcumin and both alkylphosphocholines induced apoptosis as demonstrated by activation of caspases, PARP cleavage and oligonucleosomal DNA fragmentation. Altogether, Hut-78 and MJ cells were most responsive to alkylphosphocholines. Interestingly, the same cells had the highest expression of Bcl-XL. Despite a low Bcl-XL level, HH cells were most resistant. Higher level of phosphorylated Akt was found in HH and MyLa CD8+ cells. The alkylphosphocholines induced dephosphorylation of Akt and phosphorylation of JNK. They also caused accumulation of LC3B, indicative of autophagy stimulation. 3-methyladenine, an inhibitor of autophagy initiation, potentiated the cytotoxic effect of alkylphosphocholines. Chloroquine and NH4Cl, inhibitors of autophagosome maturation, inhibited the cytotoxic effects of miltefosine and erufosine in Hut-78 cells. **Conclusions:** Taken together, our data show that alkylphosphocholines and curcumin hold promise as future therapeutics for CTCL. Alkylphosphocholines modulate signal transduction and result in induction of autophagy and apoptosis. Their therapeutic potential is strengthened by excellent safety profiles: lack of myelotoxicity and already approved skin application for miltefosine. Curcumin has the status of well tolerated non-toxic food additive.

A1032

T-CELL RICH NHL, A NEW DISEASE OR AN EMERGING UNDERSTANDING- A CASE REPORT AND REVIEW OF LITERATURE. Asuquo MI,* Jibrin PG**. *Department of Hematology, University of Calabar, Calabar, Cross River State, Nigeria; **Department of Anatomical Pathology, National Hospital, Abuja, Nigeria.

Introduction: A 50year old male referred to the hematology outpatient on account bilateral neck swellings. Has been on treatment for Chronic lymphocytic leukemia with a chlorambucil and prednisone monthly for 15 months. Diagnosis and treatment was not by a hematologist. Patient had a bullnecked appearance with lymph node enlargement involving cervical, submental, right supraclavicular and bilateral axillary regions. Histology and immunohistochemistry showed strong reactivity for Leucocyte common antigen, CD20, and CD3. Cells were small round cells with effacement of follicular arrangement. Treatment was Rituximab- CHOP with excellent clinical and hematologic remission. **Objective:** T -cell rich Non Hodgkins Lymphoma is not

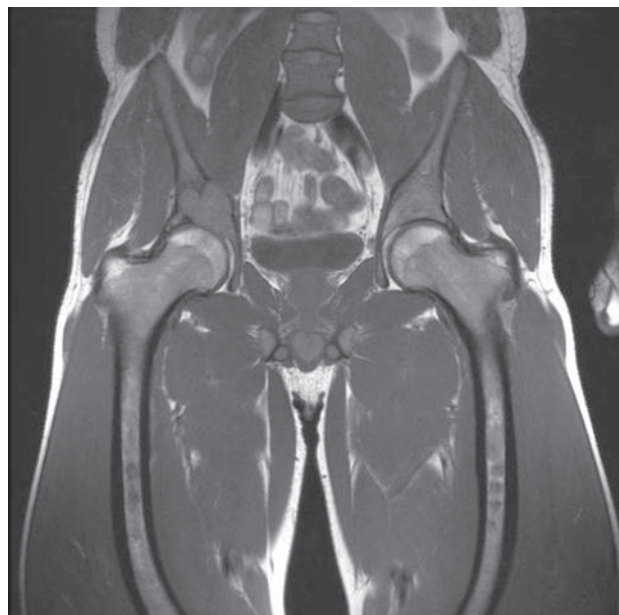
very common in our practice. What is the place for this clinical expression in the current classification of lymphoid malignancies? What are the management modalities? Literature review of current thinking on T-cell lymphoma was embarked upon to find answers to these questions. **Material and Methods.** Medline, Pubmed, online data bases were employed in the search. **Results:** T-cell rich Lymphoma has been encountered by many pathologist and hematopathologist. **Conclusions:** There is no consensus as to the classification of T-cell rich lymphoma. More work needs to be done in order to place these findings in its proper place in lymphoma classification and best line of management.

A1035

A COMPLICATED DIAGNOSIS OF MYELOMA. Jones RE, Loizou E, Lee E. Countess of Chester NHS Foundation Trust, UK. Countess of Chester NHS Foundation Trust Liverpool Road Chester CH2 1UL UK.

Introduction. This case documents the complicated diagnosis of myeloma in a 41 year old Polish man who presented to his GP with hip pain following physical exercise. A plain radiograph of the right hip revealed a lucency, and subsequent magnetic resonance imaging showed appearances consistent with marrow replacement. **Objective:** Further enquiries uncovered a history of splenomegaly and hip fracture, and a recent diagnosis of lymphoma in his mother. Blood tests showed raised IgM with no evidence of a monoclonal band, but urine was positive for Bence Jones protein. Biopsy of the acetabular lesion showed reactive sclerosis, necrosis and no evidence of plasmacytoma, however marrow aspirate, which did not show any disease, contained a clot in which there was a deposit of plasmacytoma. **Material and Methods:** Following haematology review, myeloma was excluded in the absence of paraprotein, lack of marrow involvement, and its inability to explain splenomegaly. Diagnosis of solitary plasmacytoma was made. Bone marrow aspirate was repeated which showed increased cellularity and lipid-containing macrophages with morphological features of Gaucher's cells. Repeat biopsy of the lytic lesion showed atypical plasma cells. A revised diagnosis of Gaucher's disease with a solitary plasmacytoma was made. A query was raised as to whether his mother had been misdiagnosed with lymphoma, but may actually be suffering from Gaucher's disease. **Results.** The patient was treated with a course of radiotherapy to the plasmacytoma and was commenced on enzyme replacement therapy for Gaucher's disease. He was well for several months but unfortunately developed a further plasmacytoma in the right distal femoral condyle, again treated with a course of radiotherapy. Repeat investigations were still consistent with a diagnosis of Gaucher's disease and there was still no evidence of myeloma. However after the development of lytic lesions in the spine, ribs, sternum and skull, a diagnosis of non-secretory myeloma with concurrent Gaucher's disease was made. The patient went on to have further radiotherapy, chemotherapy and autologous stem cell transplant. He unfortunately continues to

develop new lytic lesions and is awaiting review for possible allogenic stem cell transplant. **Conclusions:** Gaucher's Disease is a lysosomal storage disorder resulting from glucocerebrosidase deficiency. Consequently there is deposition and accumulation of glucocerebroside in tissue macrophages, forming Gaucher's cells. Links have been made between Gaucher's and myeloma, possibly due to a combination of macrophage activation which leads to chronic stimulation of the immune system, and immune system dysregulation due to glucocerebrosidase accumulation. However, pathology is still unclear.



A1061

RITUXIMAB DOES NOT IMPROVE THE OVERALL SURVIVAL OF PATIENTS WITH B CELL NON-HODGKIN'S LYMPHOMA: EXPERIENCE IN A DEVELOPING COUNTRY. Ruiz-Delgado GJ*.*.*.*, Gomez-Almaguer D****, Cantu-Rodriguez OG****, Tarin-Arzaga LC****, Alarcon-Urdaneta C*.⁵, Rodriguez-Morales U***, Calderon-Garcia J*.⁶, Fernandez-Vargas O*.⁷, Montes-Montiel M*, Sanchez-Cardenas M****, Ruiz-Arguelles GJ*.*.*.*. *Centro de Hematología y Medicina Interna. Clínica Ruiz; **Laboratorios Clínicos de Puebla. Clínica Ruiz; ***Universidad Popular Autónoma del Estado de Puebla; ****Hospital Universitario de Nuevo León; ⁵Benemérita Universidad Autónoma de Puebla; ⁶Facultad de Medicina. Universidad La Salle. México; ⁷Centro Universitario de Ciencias de la Salud. Universidad de Guadalajara.

Introduction: Rituximab (R) has changed the prognosis of patients with non-Hodgkin's lymphoma (NHL) in developed countries, but its role has not been analyzed in underprivileged circumstances. Treatment with R as a single agent has resulted in significant responses in patients with almost every subtype of B-cell NHL lymphoma. However, its biggest benefit is seen

when it is combined with chemotherapy regimens for patients with indolent as well as aggressive B-cell NHL. **Objective:** To analyze the impact of the addition of R to conventional chemotherapy in the outcome of treatment of patients living in a developing country, afflicted by two types of B-cell NHL: Follicular lymphoma (FL) and diffuse large B-cell lymphoma (DLCL), in two institutions: Centro de Hematología y Medicina Interna de Puebla and Hospital Universitario de Nuevo León.

Material and Methods: One hundred and two previously untreated patients with NHL treated in two institutions in a developing country were analyzed: 28 patients with FL and 74 with DLCL. Patients in both groups were treated upfront with either CHOP (cyclophosphamide, adriamycin, vincristine and prednisone) or R-CHOP (CHOP and rituximab); the decision to employ one or the other schedule was dependent on the ability of the patients to defray the expense of the monoclonal antibody.

Results: In the group of 74 patients with DLCL, 42 were given CHOP and 32 R-CHOP, whereas in the 28 patients with FL 19 were given CHOP and 9 R-CHOP. The impact of the addition of R to the therapeutic schedule was analyzed and found to be more clear in FL than in DLCL. In patients with DLCL, the overall survival (OS) was 87% at 80 months for those treated with R-CHOP and 84% at 145 months for those treated with CHOP (p NS). On the other hand, in patients with FL, the OS was 89% at 88 months for those treated with R-CHOP and 73% at 92 months for those treated with CHOP (p < .05). In a multivariate analysis, other variables were identified to be associated with the OS were IPI and number of cycles in DLCL. **Conclusions:** The addition of rituximab produced a minor positive impact in the OS of patients with FL, but not in those with DLCL. Since the addition of rituximab results in a 36-fold increase in treatment costs, these observations may be important to decide therapeutic approaches in these patients living in underprivileged circumstances.

A1065

COSTS ASSOCIATED WITH TREATMENT OF MEXICAN PATIENTS WITH NON HODGKIN LYMPHOMA AT DIFFERENT STAGES. Balderas-Peña LMA*, Rubio-Jurado B**, Solano-Murillo P**, Sat-Muñoz D**, Contreras-Hernández I**, Garcés-Ruiz OM**. *Unidad de Investigación Médica en Epidemiología Clínica; **División de Oncología-Hematología y Servicio Nacional de Radioneurocirugía, UMAE Hospital de Especialidades del Centro Médico Nacional de Occidente, Guadalajara, Jalisco, Mexico; ***Medicia Interna, Hospital General de Zona 1-A "Venados", México D. F., Instituto Mexicano del Seguro Social.

Introduction: Non-Hodgkin lymphoma is a heterogeneous lymphoproliferative disease. Regression of disease is related to prognostic factor included: age, clinical stage, extra-nodal disease, ECOG status and some biomarkers as lactic deshydrogenase. Currently used treatment with cyclofosfamide, doxorubicine, vincristin (CHOP) and prednisone show late com-

plete response no higher than 40%; with monoclonal antibodies (rituximab) enhance rate response to 75% (R-CHOP), enhancing costs of treatment derived from chemotherapy. **Objective:** To describe costs associated with treatment of different stages of patients with non-Hodgkin lymphoma at the Mexican Institute of Social Security (IMSS) from the healthcare payer's perspective. **Material and Methods:** A costs study was elaborated. Resource use and cost data were obtained from hospital (second and tertiary levels) records of 70 patients treated during the period of July 2008 to February 2009 using the following inclusion criteria: women >16 years with histological diagnosis of non-Hodgkin lymphoma who accepted to be included in the protocol with written informed consent, although excluded patients were those who showed a second malignant neoplasm or incomplete information. We calculate mean, median, standard deviation (SD) and 25th and 75th percentiles for each clinical stage, and statistical differences were estimated through ANOVA tests; p value < .05 was considered significant for demonstrating differences. **Results:** Median total cost (all costs are in U.S. dollars) per patient was \$4,683.11 (\$2,857.20-\$10,031.70); mean cost per chemotherapy was \$3,595.96 (SD: \$5,800.27); and mean cost of radiotherapy was \$439.05 (SD: \$554.51). Mean cost per patient in each clinical stage with chemotherapy was as follows: I) \$3,167.01 (SD: \$3,912.16); II) \$4,484.05 (SD: \$3,546.32); III) \$5,524.38 (SD: \$4,424.02); IV) \$2,500.44 (SD: \$3,502.07), and for non-classified patients: \$2,482.52 (SD: \$3,502.07) (p=0.499). **Conclusions:** Results show that in Mexico, the most expensive treatment was for patients with Non-Hodgkin lymphoma at clinical stage III.

A1080

CONCENTRATION OF THE RECEPTOR OF ACTIVATOR PLASMINOGENO-UROKINASA IN HEMATOLOGICAL NEOPLASIAS. Rubio-Jurado B**, Tello-González A*, Bustamante-Chávez L**, de la-Peña A****, Riebeling-Navarro C⁵, Arnulfo Nava A**⁵. *Servicio Hematología, UMAE, HE, CMNO; **Unidad de Investigación en Epidemiología Clínica, UMAE, HE, CMNO; ***Lab Hematología, UMAE, HE; ****Lab. Clínico. Inst Nal. Cardiología, Ignacio Chávez; ⁵UIEC UMAE HP CMN-SXXI IMSS/ UNAM, 6DICS, Universidad Autónoma de Guadalajara.

Introduction: The receptor of the Activator of Plasminogen type urocinasa (uPAR) is a molecule related to the invasiveness and metastasis of the cancer, being overexpressed in the surface of the neoplastic cells and possibly conditioning detectable serum levels. It participates in the adhesion and migration of inflammatory cells. In this ambisoectivo work the serum levels of uPAR in hematological neoplasies are identified, especially in lymphomas. **Objectives:** To identify the serum levels of uPAR in patients with hematological neoplasies. **Material and methods:** Incident and prevalent consecutive cases of patients and 10 controls clinically healthy, were included. Site: UMAE Hospital of Specialties CMNO, IMSS. Clinical-demographic

data were registered of the file, it determined u-PAR in serum by ELISA (pg/L) and D-Dimer by nefelometría (mg/dl). **Results:** We included 42 cases (median age, minimum value to maximum value) 55, 25 to 88 years. 15 (36%) women and 27 (64%) men. Eight cases are new cases. Lymphomas, 35 cases (83%), LAM 5 (12%), LAL and LCG one case (2.3%). Histopathology of cases of lymphoma: Diffuse Cel-G 19 (54%) Diffuse Small Cel 4 (11%) Follicular 4 (11%), Cel-T 4 (11%), Mantle Cel 2 (5.7%). D-dimer, elevated in 28 cases (67%), range: 570 to 9191 ug /dL. Median serum level of uPAR 785 pg / L, ranging 190-1950pg / L. UPAR in 10 controls with a median 600 pg / L, ranging 360-750 pg / L ($p < 0.01$). Novo patients are 8 cases with lymphoma cel-B, with median serum level of 852 pg uPAR / L (360-1650 pg / L). **Conclusions:** In this case series we found a high frequency of positive D-D, no thrombotic events present clinically, shows that there is activation of coagulation and fibrinolysis in these patients. Compared with controls soluble uPAR levels in patients with cancer are high. These biomarkers should be related in our population with treatment response and survival.

A1081

HODGKIN LYMPHOMA (HL) IN PATIENTS WITH HUMAN IMMUNODEFICIENCY VIRUS (HIV). Labarta JD*, Varela AI**, Figueroa MF**, Zarate T**, Pavlovsky C***, Giere I***, Lombardi V***, Verri V****, Gonzalez J****, Flores GM****, Larripa IB⁵, Mercado-Guzman ZV⁵, Otero IS**, Lluesma-Goñalons M**, Ardaiz M del C**, Moiraghi EB**. *Genoma, Buenos Aires, Argentina; ** División Hematología del Hospital JM Ramos Mejia, Buenos Aires, Argentina; ***Fundaleu, Buenos Aires, Argentina; **** División Hematología del Hospital CG Durand, Buenos Aires, Argentina; ⁵Departamento de Genética de la Academia Nacional de Medicina, Buenos Aires, Argentina.

Introduction: HL is not considered a marker disease for AIDS, however, in patients with HIV, it becomes evident in advanced stages, with involvement of the bone marrow (BM), presence of B symptoms, aggressive histological patterns and worse prognosis than in patients without evidence of immunocompromise. A report published by the National Cancer Institute (NIH), pointed out that these patients evidenced unfavorable histologic features (mixed cellularity and lymphocytic depletion), extranodal compromise, B symptoms, and a CD4 count of 300cells/mcL. Those patients that have received conventional chemotherapy plus Highly Active Anti Retroviral Therapy (HAART) revealed an overall survival of 74%, at 2 years as compared to 30% to those that have not received HAART. **Objective:** to describe the characteristics of this disease entity, its evolution, disease free survival and overall survival in patient with HL and HIV. **Material and methods:** retrospective review of 21 patients with HL and HIV managed and treated in our department during a 10 year period (2000-2010). **Results:** we reviewed 21 patient cases, 18 males/3 females, with a median age of 34 years. Ninety percent showed

evidence of histologic features of mixed cellularity and 10% evidence for nodular sclerosis and a median CD4 count of 176cells/mcL.; 62% were IVB stage at diagnosis with BM involvement, 24% were stage IIIB and 14% were IIB. Seventy six percent were receiving HAART at initial diagnosis and 67% were treated with chemotherapy with Adriamycin, bleomicin, vinblastine and dacarbazine (ABVD protocol) achieving a complete remission in 56% of the cases. These patients also received HAART. The overall survival was 67 months and the disease free survival was 24 months. The overall mortality was 24%. **Conclusions:** in our patient group there was a prevalence of males over females, a high proportion were diagnosed in advanced stages of the disease with aggressive histological patterns and the median CD4 counts was lower. The majority of the patient received HAART and had an overall survival of 5,5 years. Only one patient relapsed. Three of the patients that were not evaluable received 1-2 cycles of ABVD and died due to infectious complications.

A1093

ADVERSE EVENTS ASSOCIATED WITH THALIDOMIDE IN MULTIPLE MYELOMA PATIENTS: EXPERIENCE OF A UNIVERSITY HOSPITAL. Tarín-Arzaga L, Díaz-Osuna V, Pequeño Luévano M, Flores-Jiménez J, Saldaña-Vazquez R, Ceballos-López A, Velasco-Ruiz I, Gómez-Almaguer D. Servicio de Hematología del Hospital Universitario "Dr. José Eleuterio González", UANL. Monterrey N.L. México.

Introduction: Treatment for multiple myeloma (MM) has changed in recent years. Bortezomib and lenalidomide are effective new high-cost drugs not affordable for the majority of our patients, therefore, thalidomide continues being an excellent drug for MM, unfortunately, due to adverse events is necessary to discontinue the drug or reduce the dose. **Objectives:** To assess the frequency of adverse events associated to thalidomide. **Material and methods:** We analyzed a cohort of patients with a diagnosis of multiple myeloma treated with a thalidomide, dexamethasone and cyclophosphamide recruited in the Hematology service of the University Hospital, UANL, to assess the frequency of adverse events associated with thalidomide, and the clinical impact of the dose modification. To evaluate adverse event severity CTCAE-4 for each event was used. **Results:** We included 67 patients. Thirty three (49.3%) females. The median age was 65 (25-82) years. Forty nine patients were evaluated for adverse events: 45 (91.8%) of them presented at least one adverse event; 18 (40%) patients who presented any adverse event initiated with 100 mg and 27(60%) with 200mg. Of 40 evaluated patients for sensory neuropathy (SN), 33(82.5%) developed some grade of SN, 15(45.5%) of them had been initiated with 100mg and 18 (54.5%) with 200mg. In 15 (45.4%) patients the dose was reduced: 2 patients with grade 1 and 13 with grade 2 SN; seven patients (21.2%) discontinued treatment due to grade 3 SN. In 11(33.3%) with grade 1 SN the dose remained unchanged. Thirty one patients were evaluated for constipation, 30 (96.8%)

of them developed grade 2 constipation; 20 (66.7%) was started with 200 mg and 10 (33.3%) with 100 mg, no dose changes were done. Ten patients of 45 evaluated patients developed grade-1 drowsiness, 8 (80.0%) of these patients at initial dose of 200 mg and 2(20.0%) with 100mg; two (2.9%) of 67 patients developed a thrombotic event, in one patient initial dose was 100mg and in the other patient was 200mg; in both patients the treatment was interrupted. **Conclusions:** Although low doses of thalidomide were used in our patients, we found a higher frequency of adverse events than previously reported. It is necessary to optimize the thalidomide dose in our mainly uninsured patients who have very few treatment options.

A1094

REPORT OF A PLASMA BLASTIC LYMPHOMA (PBL) IN HIV NEGATIVE PATIENT. Labarta JD*, Varela AI**, Figueroa MF**, Zarate T**, Pavlovsky C***, Giere I**, Lombardi V***, Verri V****, Gonzalez J****, Flores GM****, Larripa IB⁵, Mercado- Guzman ZV⁵, Otero IS**, Lluesma-Goñalons M**, Ardaiz M del C**, Moiraghi EB**. *Genoma, Buenos Aires, Argentina;** División Hematología del Hospital JM Ramos Mejia, Buenos Aires, Argentina; ***Fundaleu, Buenos Aires, Argentina; **** División Hematología del Hospital CG Durand, Buenos Aires, Argentina; ⁵Departamento de Genética de la Academia Nacional de Medicina, Buenos Aires, Argentina.

Introduction: PBL is a neoplastic process, rather infrequent with aggressive clinical evolution. The cells of PBL are characterized by the absence of B markers (CD20). In some cases CD79a is identified, partially. They show definite expression of VS38c and CD138. Originally, the PBL was described as a lesion in the oral cavity of HIV positive patients, associated with Epstein - Barr virus. Lately, the report of these types of cases underwent an increment, mostly in cases of patient without any definable and overt immunodeficiency. The median age at diagnosis is 58 years with predominance among males. This disease entity is mainly identified in the oral cavity and GI tract in HIV negative patients, with infrequent involvement of lymph nodes. **Objective:** We are reporting a case studied and managed at our center. **Material and Methods:** 49 y/o male, was referred in September 2011 with an exophytic tumor lesion of rapid growth present in the right lower maxilla, identified following a dental extraction. Serology studies which included HIV, HBV and HCV were all negative. By CAT scan it is evident that there were lytic lesions in the ascending branch of the right superior maxilla, right costal arch (second and tenth rib) and 10th left rib, without any evidence for lymph node involvement. Protein electrophoreses studies failed to reveal any abnormalities. Biopsy of the lesion in the oral cavity was diagnosed as PBL (WHO 2008). Immunophenotypic studies revealed: CD20 (-), bcl-6(-), CD38 (+), kappa (+), co-expression of MUM-1 and partial expression of CD45, Ki67 (85%). Bone marrow biopsy: infiltrated with PBL. **Results:** The patient was given 4 cycles of chemotherapy (bortezomid, dexamethasone

and cyclophosphamide). In January 2012 bone marrow biopsy, Cat scans and immunophenotyping studies failed to reveal the presence of abnormal elements. The patient is now in complete hematologic remission scheduled for autologous bone marrow transplant. **Conclusions:** We report a case of PBL diagnosed in a HIV negative patient with a negative history of secondary immunodeficiency induced by organ transplant, steroidal therapy or autoimmune disease process. The therapy included Bortezomid, Cyclophosphamide and dexamethasone. The patient achieved complete hematologic remission. The adverse effects of the chemotherapy were Grade 1-2.

A1105

PRIMARY BONE LYMPHOMA. CASE REPORT. Guerrero-Martínez AI*, Garcés- Ruiz OM*, Aguilar-López LB*, Rincón-García I*, Díaz-Ruiz R*, Ibarra-Hernández A*, González-Moncada AI*, Caballero-Jasso J**, Tavares J***, Rubio-Jurado B*. *Hematology Service, UMAE, CMNO; **Surgical Oncology Service, UMAE, CMNO; ***Departament of Pathology, UMAE, CMNO. Instituto Mexicano del Seguro Social. Guadalajara, Jalisco. México.

Introduction: Primary bone lymphoma represents 3% of malignant bone tumors, accounting for 4-7% of primary extranodal lymphomas. It is essential to the staging and diagnosis of the tumor, including immunohistochemistry and image study to design comprehensive treatment decision. Based on this information an interdisciplinary team determines the individual therapeutic approach. There is no standard treatment strategy. This condition presents diagnostic difficulties delaying the start of treatment. We present three cases of chronic bone pain with a final diagnosis of primary bone lymphoma. **Objective:** To report the clinical characteristics of patients with primary bone lymphoma. **Material and methods:** We describe prevalent cases with primary bone lymphoma, were attended in the period 2009 -2011, at the Hospital of Specialties IMSS CMNO. Clinical-demographic data were registered of the file. **Results:** We included 3 cases are female. 18,28 and 44 years old. Clinically present with chronic pain, located in the affected site. No B symptoms, increased local volume. Anatomical location: tibia, iliac and femur. Histopathology, the 3 patients with diffuse large cell B immunophenotype, CD20 (+). No bone marrow infiltration. Laboratory, anemia normo / normo in two cases (Hb 9.4 and 10.4 g / dL). Normal lactic deshidrogenasa. Treatment. R-CHOP in 3 cases, one case received local radiotherapy. One patient received surgical treatment. Clinical and radiographic are in remission. 4,7 and 25 months follow. **Conclusions:** DISCUSSION. The primary bone lymphoma is a rare condition. Early diagnosis is based on adequate histopathological supported immunohistochemistry. The association with pathological fracture or spinal cord compression at diagnosis complicates their management. Staging with radiodiagnostic methods as MRI or PET identified a higher proportion of patients with advanced disease who require treatment intensification. Patients with limited disease

treated with radiation therapy alone has poor results (5-year OS <45%). Protocols chemo-radiotherapy improves disease-free survival and overall survival. It is difficult to determine the best method for staging and treatment given the low prevalence of this disease in our population.

A1108

CASE REPORT OF A CD4+/CD56+ HEMATODERMIC NEOPLASM. Prisdashka K*, Balabanova M*, Genova M**, Konstantinov S***. *Department of Dermatology and Venereology, Medical University in Sofia, Bulgaria; **National Specialized Hospital for Active Treatment of Hematological Diseases - Sofia, Bulgaria; ***Department of Pharmacology, Pharmacotherapy and Toxicology-Medical University in Sofia, Bulgaria.

Introduction: The CD4+/CD56+ hematodermic neoplasm, formerly known as blast NK-cell lymphoma, is a rare and aggressive neoplasm with a high incidence of cutaneous involvement, risk of leukemic dissemination and poor prognosis. The characteristic features are an expression of the T-helper inducer cell marker CD4 and the NK-cell marker CD56 in the absence of other T-cell or NK-cell specific markers. It usually presents as either solitary or multiple nodules or systemic involvement of lymph nodes and bone marrow. The disease primarily affects elderly patients with male/female ratio of 3 to 1 but can occur in patients younger than 50 years in 30% of the cases, with only occasional paediatric cases reported. **Objective:** To describe a rare case of aggressive hematodermic neoplasm with lethal outcome. **Material and Method:** We present a 75-year old female patient. In September 2010 the patient was admitted in our clinic with six month history of solitary tumor formation on the right arm. Multiple well-demarcated, bruise-like hardened nodules over her face, trunk, arm and legs were seen. The diagnosis of a CD4+/CD56+ hematodermic neoplasm (Blast plasmacytoid dendritic cell neoplasm) was based on the histological and immuno-histochemical exploration of skin lesion samples and bone marrow biopsy, as well as on the morphology and flow cytometry of peripheral blood neoplastic cells. **Results:** The patient received 10 cycles of polychemotherapy (cyclophosphamide, epirubicin, vincristine and prednisone) with little response. Following the fourth cycle the lesions achieved near complete resolution with reduction of the abnormal population and loss of expression of the CD56 from baseline flow cytometry. After the eighth cycle the patient came with relapse of the skin lesions. The disease progressed. One month after the 10th chemotherapy cycle the patient died. **Conclusion:** This case illustrates the importance of recognizing the clinical, pathological and immuno-histochemical features of the disease. It is aggressive with median survival period of only 12 to 14 months. An early start of polychemotherapy is crucial and usually induces remissions, although relapse and resistance development are common. We discuss further studies and trials which are needed in order to find some better treatment options for this rare, but aggressive and often lethal neoplasm.

A1111

CLINICAL FEATURES AT DIAGNOSIS OF CHILDHOOD LYMPHOMA AT A UNIVERSITY HOSPITAL IN NORTH-EAST MEXICO. Jaime-Pérez JC*, Olguin-Ramírez LA*, Rodríguez-Martínez M*, Marfil-Rivera LJ*, González-Llano O*, Gómez-Almaguer D*. *Hospital Universitario, Dr. José Eleuterio González, Universidad Autónoma de Nuevo León, Monterrey, Nuevo León, México

Introduction: There is scarce information regarding the incidence of lymphoma during childhood from developing countries. **Objectives:** We aimed to document the epidemiologic characteristics, clinical presentation and treatment modalities of lymphoma in pediatric patients from northeast Mexico referred to the Hematology Service of the "Dr. José E. González" Cancer Center of the School of Medicine of the Autonomous University of Nuevo León. **Material and Methods:** We conducted a retrospective observational study covering the period from January 1, 2005 to December 31, 2010. The study included all patients younger than 16 years of age with a histopathological diagnosis confirmed by biopsy of Hodgkin Lymphoma (HL) or Non-Hodgkin Lymphoma (NHL), from northeast Mexico. Clinical files and electronic records were reviewed, and a database built using the statistical package SPSS v. 17 for the analysis of collected data. The clinical characteristics and individual frequencies for each type of pediatric lymphoma were analyzed. Patients that did not fulfill the age range, had incomplete data, or a non-confirmatory biopsy were excluded. **Results:** Data from 34 children were analyzed; 10 (29.4%) had a diagnosis of HL and 24 (70.5%) NHL. Three (30%) patients with HL were females and 7 (70%) men, while NHL was the diagnosis in 6 (25%) girls and 18 (75%) boys. The median age at presentation for the whole group of childhood lymphoma was 9.5 years (1-16), while separate medians were 10.5 years for HL, and 8.5 years for NHL. The histopathological diagnosis in 70% of the cases of HL was nodular sclerosis, while in the remaining 30% a specific subtype was not identified. The histopathological types for NHL included B cell (12.5%), lymphoblastic (45.8%), Burkitt (20.8%), anaplastic (4.1%), large cell (4.1%) and in the remaining 12.5% a subtype could not be specified. **Conclusions:** We describe a small group of children with the diagnosis of HL, 70% were classified to the nodular sclerosis subtype, which coincides with the reported worldwide; in 30% of the cases a specific subtype was not identified. For NHL we found a higher incidence in children at 70.5%, compared to 55% documented in the U.S. and Europe, and a male to female gender rate of 4:1. While in other populations a 3:1 ratio is reported. The commonest type of NHL in our group was the lymphoblastic variety, while other reports refer a predominance of Burkitt's lymphoma. 33% of our patients with NHL died during the study, which is higher than expected and reflects a low cure rate.

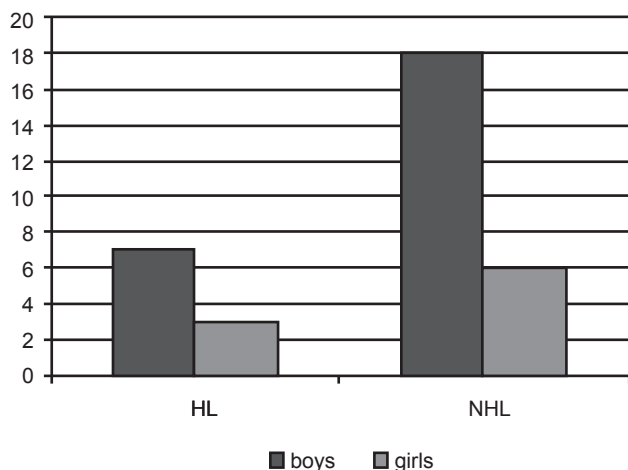


Figure 1. Distribution according to gender for Hodgkin and Non-Hodgkin Lymphoma of childhood in northeast México.

A1115

EPOCH CHEMOTHERAPY IN HIV-ASSOCIATED NON-HODGKIN LYMPHOMA: EXPERIENCE OF AN INSTITUTION. Amador-MF, Vargas-Ruiz A, Sherling-Ocampo A, Deffis-Court M, Dell J, Rosales D, González G, Espinoza G, Zamora E. Instituto Nacional de Ciencias Medicas y Nutrición Salvador Zubiran.

Introduction: Patients with HIV-associated non Hodgkin lymphoma have a poor prognosis. Unfortunately, the poor overall survival in such patients, combined with the toxicity associated with aggressive chemotherapy, led to the use of less intensive therapy to reduce complications. Infusional administration of chemotherapy has been explored as a potential strategy in patients with HIV-associated non Hodgkin lymphoma. Several trials have evaluated a continuous intravenous infusional regimen of etoposide, vincristine, doxorubicin, cyclophosphamide and prednisone (EPOCH) in such patients, shown good results.

Objective: Evaluate the efficacy and safety of infusional EPOCH chemotherapy in patients with HIV-associated non-Hodgkin lymphoma. **Material and Methods:** The patients were enrolled between 2005 and 2011. Eligible patients were HIV seropositive associated non-Hodgkin lymphoma. EPOCH chemotherapy was administered for 6 cycles over day 1 through day 5 as a 96-hour continuous infusion of etoposide, doxorubicin and vincristine and oral prednisone with cyclophosphamide on day 5 as outlined in (table 1). The probabilities of overall survival and progression free survival were calculated using the Kaplan-Meier method beginning from the date of diagnosis HIV-associated non-Hodgkin lymphoma until death, progression due to lymphoma or last follow-up as appropriate. **Results:** Of 23 patients enrolled, 16 were analyzed. The median patient age was 38 años, 87% were men. The principal histologic subtypes were DLBCL and plasmablastic lymphoma. The majority of patients had Interna-

Table 1. EPOCH chemotherapy

Drug	Dose	Route	Treatment day
Etoposide	50mg/m ² /day	CIV	1,2,3,4 (96 hours)
Doxorubicin	10mg/m ² /day	CIV	1,2,3,4 (96 hours)
Vincristine	0.4mg/m ² /day	CIV	1,2,3,4 (96 hours)
Cyclophosphamide (cycle 1)	375mg/m ² /day	IV	5
CD4+ cells >100/mm ³	187mg/m ² /day	IV	5
CD4+ cells <100/mm ³			
Cyclophosphamide (after cycle 1)	é187mg above previous cycle	IV	5
Nadir absolute neutrophil count >500	é187mg below previous cycle	IV	5
Nadir absolute neutrophil count <500			
Prednisone	60mg/m ²	PO	1,2,3,4,5

tional Prognostic Index (IPI) low, however the most patients had advanced clinical stage at diagnosis of non-Hodgkin lymphoma. The median CD4+ cells was less than 100/mm³. Antiretroviral treatment at study entry included HAART in 37.5%. Complete remission was achieved in 50% of patients. The median follow-up time was of 13 months. The progression-free survival (PFS) and overall survival (OS) rates were 56% y 75%, respectively. (figure 1 y 2). the major side effect was neutropenia and 37.5% developed severe neutropenia and fever. **Conclusions:** We conclude that infusional EPOCH is an effective regimen for HIV-associated lymphoma with tolerable toxicity, however the efficiency was lower than previously reported. We should look at other strategies to improve response rates in patients with HIV-non Hodgkin lymphoma.

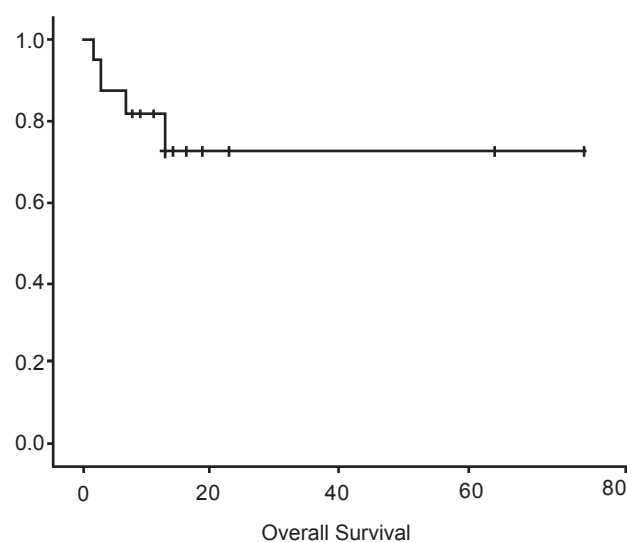


Figure 1. Overall Survival (OS)

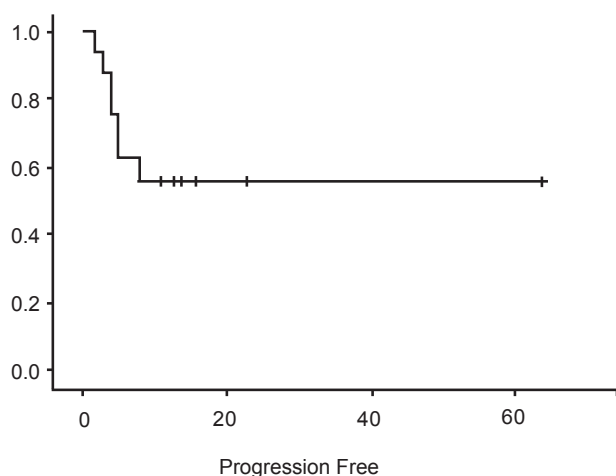


Figure 2. Progression Free Survival (PFS)

A1138

ALK-POSITIVE DIFFUSE LARGE B-CELL LYMPHOMA PRESENTING WITH MASSIVE SPLENOMEGALY AND CERVICAL LYMPHADENOPATHY: A CASE REPORT WITH PARTICULAR IMMUNOPHENOTYPIC FEATURES. Deffis-Court M*, Llamas-Gutiérrez FJ**, González-Rodríguez K*, Vargas-Ruiz AG*, Lome-Maldonado C**. *Departamento de Hematología; **Departamento de Patología. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubiran, México D.F., México.

Introduction: Anaplastic lymphoma kinase (ALK)-positive diffuse large B-cell lymphoma is a rare and distinct variant of diffuse large B-cell lymphoma with characteristic morphologic, immunophenotypic, and cytogenetic features. **Objectives:** We report a case of ALK-positive diffuse large B-cell lymphoma in a 31-year-old male with progressively worsening splenomegaly and cervical lymphadenopathy. **Material and Methods:** Review the clinical chart of the patient, medical facts, laboratory data and histology reports. Online literature search keywords: Anaplastic lymphoma kinase (ALK)-positive diffuse large B-cell lymphoma. **Results:** Histologically, the tumor cells exhibited plasmablastic morphology with expression of EMA +, IgA +, Mb2, OCT-1, BCL-6 but negative for CD20, CD10, CD79a, CD30, HHV8, CD138 and CD38. More importantly, the neoplastic cells showed positive immunoreactivity in a granular cytoplasmic distribution sparing the nucleus and nucleolus. **Conclusion:** This is the first reported case that showed histology ALK-positive diffuse large B-cell lymphoma in the spleen.

A1139

GEMCITABINE, VINORELBINE AND DEXAMETHASONE IN PATIENTS WITH RELAPSING OR REFRACTORY HODGKIN'S AND NON-HODGKIN'S LYMPHOMA. Rosas-Cabral

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Introduction: The treatment of patients with relapsed and refractory Hodgkin lymphoma (HL) or non-Hodgkin Lymphoma (NHL) remains challenging. New treatment strategies that are based on targeting oncogenic signaling pathways are currently explored. This review will focus on new treatment modality for patients with relapsed. **Objectives:** Evaluate the efficacy of gemcitabine/vinorelbine/dexamethasone in the treatment of patients with relapsed or refractory HL and NHL. Assess the safety of chemotherapy regimen. **Material and Methods:** Retrospective and retrospective study. We review patient records with relapsed and refractory HL or NHL, who were treated with GVD, of either gender, aged 18 years or more, in the period between January 2009 and June 2011, in hospitals in the central region of Mexico. **Results:** Seven patients (six male and 2 female) were treated with gemcitabine/ vinorelbine/dexamethasone (GVD) from May 2009 through March 2011, for refractory or recurrent Non-Hodgkin's or Hodgkin's lymphomas (4 in relapse, 5 in primary refractory disease). The median age, the median time from diagnosis and the median duration of previous treatments were respectively 25 years (range 18-72), 29 months (range 10-99) and 6 months. Histology was as follows: 3 nodular sclerosing, 1 mixed cellularity, 1 lymphocyte depleted and 2 diffuse large cell lymphomas. Patients were stage IIB (3), IVB (3) and IV-X (1). Three patients were previously treated with ABVD plus radiotherapy, one with ABVD alone, two with CHOP, and one with CHOP-R. Five patients had bulky disease at diagnosis and received involved-field radiotherapy for those sites, three had bulky disease at relapse. Four patients had extranodal disease at relapse. All of patients were treated with gemcitabine 1000mg/m² days 1 and 8, vinorelbine 40 mg/m² day 1, and dexamethasone 40 mg days 1-4, 8-11 each 21-28 days. A median of 6 (range 6-8) courses of GVD were administered. Following treatment, complete response was achieved in 4/7 patients, 3/7 achieved partial response. 86% of patients remain free of disease 8-25 months (median 9 months). One patient died of progressive disease (response duration 8 months). Myelosuppression was the major toxicity with 1/7 febrile neutropenia (1 documented bacteremia). The rest of patients not experienced alopecia, severe neutropenia or delayed of programmed treatment, and use of growth factors was not necessary. Overall survival is 86% to 25 months of following. **Conclusions:** Our data suggest that gemcitabine/vinorelbine/dexamethasone is an effective and well tolerated regimen in previously treated patients with Hodgkin's or Non-Hodgkin's lymphomas, and deserves further evaluation.

A1146

MULTIPLE EXTRAMEDULLARY PLASMACYTOMA - THE CLINICAL PICTURE OF RELAPSE IN A PATIENT WITH IGA MULTIPLE MYELOMA. Popov VM***, Vladareanu AM*****,

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Introduction: Multiple plasmacytomas are an uncommon presentation of extramedullary relapse of multiple myeloma (MM). **Objective:** We describe a case of relapsed IgA MM with three sites of extramedullary plasmacytoma with no evidence of marrow involvement. This is the second reported case of a patient with rapid extramedullary relapse of disease. **Material and Methods:** Woman, 42 years old, was admitted for back pain in the thoraco-lumbar region of the spine; pain radiates down the right sciatic nerve, L5-S1 characteristics, and incomplete paresis of the right tibial and common peroneal nerve. **Results:** Surprisingly, although CT diagnosis was herniated lumbar disk, there was an epidural tumor that infiltrates L4 vertebral body and was totally ablated. Because after surgery the low back pain persists and within 5-6 days develops strong paraparesis, urine retention, a thoracolumbar junction MRI is made emphasizing a giant tumor that infiltrates T12-L1 vertebral corpus, with epidural extension and compression of the dural sac, marked tumor extension being in intimate adherence and anterior to inferior vena cava and aorta. Surgical intervention has performed. The histopathologic diagnosis was plasmacytoma. The bone marrow biopsy indicated 80% plasmocytic infiltration. Protein electrophoresis and immunofixation was Ig A Lambda positive. The patient received 4 courses of VAD chemotherapy without remission, followed by 8 Bortezomib Dexamethasone courses. Checks after the 8 courses and after 6 months showed complete hematologic remission and partial clinical remission (sensibility and sphincterian control). After one year, the patient presented severe anemia, increase in abdominal volume and light upper abdominal pain. Ultrasound examination revealed bilateral giant ovarian tumor. The bone marrow biopsy did not show plasmocytic infiltration. The surgical exam showed associated ovarian tumor, an inflamed gallbladder which was surgically removed. Histopathological and immunohistochemical examination of removed samples (ovary and gallbladder) established the diagnosis of plasmacytoma. After surgical treatment, the patient accused right facial paresthesia with complete anesthesia after two weeks and associated right hipoacusia. MRI revealed pontocerebelous angle infiltrates. **Conclusions:** The clinical onset with spinal tumor mass extended into the spinal canal as well as the paravertebral is less characteristic. Column scans show uncharacteristic appearance in its early stages, determining the obligation of performing MRI in cases with intense pain treatment to rebel surgical interventions, before the onset of spinal cord compression syndrome. Chemotherapy associated with kinetotherapy allowed partial motor recovery of the patient. Extramedullary plasmacytoma can mimic the appearance of solid tumor of thoracic or abdominal organs, seldom there are multiple localizations without bone marrow involvement.

A1147

SINGLE-AGENT CARFILZOMIB VERSUS BEST SUPPORTIVE CARE REGIMEN IN PATIENTS WITH RELAPSED AND REFRACTORY MULTIPLE MYELOMA: FOCUS (PX-171-011), A RANDOMIZED, OPEN-LABEL, PHASE 3 STUDY. Hájek R*, Masszi T**, Palumbo A***, Minarik J****, Oriol A⁵, Rosinol⁶, Maisnar V⁷, Bryce R⁸, Ro S⁸, Powne A⁹, Ludwig H¹⁰. *University Hospital Brno, Czech Republic; **St. Istvan and St. Laszlo Hospital, Hungary; ***Molinette Hospital, Italy; ****University Hospital Olomouc, Czech Republic; ⁵Hospital Universitari Germans Trias i Pujol, Spain; ⁶Hospital Clinic Barcelona, Spain; ⁷University Hospital Hradec Kralove, Czech Republic; ⁸Onyx Pharmaceuticals, South San Francisco, CA; ⁹Wilhelminen Hospital, Vienna, Austria.

Introduction: The proteasome inhibitor carfilzomib has demonstrated single-agent activity in phase 2 studies in patients with relapsed and refractory multiple myeloma (R/R MM). In PX-171-003-A1—a single-arm study of carfilzomib monotherapy in heavily pretreated refractory patients—37% of patients achieved greater than or equal to minimal response (MR) and median time to progression was 3.9 months. Carfilzomib is currently under review by the US Food and Drug Administration for approval to treat R/R MM. **Objectives:** To support European registration, a phase 3 randomized study—FOCUS (CarFilzOmib for AdvanCed Refractory Multiple Myeloma European Study; EudraCT No. 2009-016840-38)—is being conducted comparing overall survival (OS) with single-agent carfilzomib to a “best supportive care” (BSC) regimen (low-dose glucocorticoids with optional cyclophosphamide and comfort and palliative care). **Material and Methods:** Patients must have received greater than or equal to 3 prior regimens including bortezomib treatment (defined as greater than or equal to 4 cycles at full dose as tolerated). Patients must be responsive to greater than or equal to 1 line of therapy and be either non-responsive (less than or equal to stable disease) or refractory to their most recent therapy. Eligible patients are randomized 1:1 (stratified by number of previous therapies and geographical region) to carfilzomib or BSC regimen. Target enrollment is 302 patients. Treatment schemes are: Treatment will continue until disease progression or unacceptable toxicity. Following confirmation of disease progression or discontinuation from study treatment, all patients will enter long-term follow-up for survival. Crossover is not allowed upon progression. The primary endpoint is OS and secondary endpoints include progression-free survival, overall response rate, clinical benefit rate, disease control rate, duration of response, and safety. Disease assessments will be determined by study investigators and an independent review committee according to the International Myeloma Working Group Uniform Response Criteria (MR per European Blood and Marrow Transplantation Group criteria). The FOCUS study began accruing in September 2010 and 135 patients are enrolled as of January 2012. **Conclusions:** FOCUS will provide more rigorous data for carfilzomib, as this is the first carfilzomib study with OS as the primary endpoint and will not

Cycle	Carfilzomib	BSC
1	20 mg/m ² IV D 1,2 27 mg/m ² IV D 8,9,15,16	Prednisolone 30 mg PO QOD OR
2-9	27 mg/m ² IV D 1,2,8,9,15,16	Dexamethasone 6 mg PO QOD; plus optional
≥10	27 mg/m ² IV D 1,2,15,16	Cyclophosphamide 50 mg PO QD (max 1400 mg/cycle)

be confounded by crossover. The larger study group will also provide more robust secondary response data and additional safety data.

A1148

PERIPHERAL T-CELL LYMPHOMA NON SPECIFIC (PTCL NOS) WITH REGULATORY T-CELL PHENOTYPE. CASE REPORT. Espinoza-Macías GA*, Agreda-Vásquez GP*, Llamas-Gutiérrez FJ**, Lome-Maldonado C**. *Hematology Department; **Pathology Department. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán

Introduction: Peripheral T cell Lymphoma non specific (PTCL NOS) is the most common PTCL and represent almost the 30% of the cases. This category involves mature T cell neoplasia that do not fix in anyone else. The T regulator lymphocytes phenotype (Treg) seems to represent a different lymphoma subtype, which is not recognized by the World Health Organization (WHO), but with a very aggressive clinical and worst behavior. There are just 3 reported cases in the world literature. **Objective:** To present a clinical case of PTCL NOS with Treg phenotype at the Instituto Nacional de Ciencias Medicas y Nutrición Salvador Zubiran. **Material and Methods:** We recollected clinical and laboratory dates from the expedient's patients. The diagnosis was confirmed by the hematopathology, based on the strong CD25 and FOXP3 positivity. **Results:** A 61 years-old woman, with medical history of Hypertension and Diabetes of 5 years of evolution in treatment. Starts with clinical picture characterized by edema of lower limbs and back pain, abdominal pain patient has subsequently located, accompanied by weight loss of 18 kg and data from partial bowel obstruction, presence of abdominal mass. CT evaluation revealed the presence of wall thickening at jejunal and multiple abdominal lymph nodes at the paraaortic. It takes intestinal biopsies by enteroscopy with a diagnosis pathologic of PTCL NOS Treg (CD3 +, FOXP3 +, CD4 +, CD20-, MUM1-, CD10-, BCL6-IGA-, CD8-, CD7-, CD56-, CD30-, CD25 +, CD5 - LPM1-cytokeratin AE1/AE3-) without bone marrow infiltration. Staged as IIIB low-intermediate International Prognostic Index and starts handling scheme with CHOEP chemotherapy for 6 cycles, achieving complete remission. A 12 month follow-up showed relapse, with a picture of diarrheal stools and abdominal colic pain, showing colonoscopy, infiltrating ascending and transverse colon with a report

of the same PTCL NOS Treg (CD25 + / FOXP3), thus ending 1 st relapse and ICE decides to rescue scheme. The patient had multiple complications: pneumonia, CNS toxicity by Ifosfamide, this one with nonconvulsive status epileptics. The patient died 21 months of diagnosis. **Conclusions:** The differential diagnosis includes adult T-cell leukemia/lymphoma, which proposed cell of orange has a Treg phenotype. The proposal of a new subtype of lymphoma where the expression of FOXP3 can be present.

A1152

INCIDENCE OF HIV (HUMAN IMMUNODEFICIENCY VIRUS)-ASSOCIATED LYMPHOMAS AT INSTITUTO NACIONAL DE CANCEROLOGÍA (INCAN), IN A PERIOD OF SIX YEARS. Barbosa-Ibarra AA, Rivas-Vera MS. Hematology Department Instituto Nacional de Cancerología, Mexico, DF

Introduction: Patients with HIV infection have a high risk of developing neoplasias. Lymphomas are considered to be the second most frequent neoplasia after Kaposi's sarcoma and a late manifestation of HIV infection. Lymphomas are commonly diagnosed in advanced stages. The most frequent histological type is the diffuse large-B cell lymphoma (DLBCL). The schemes of chemotherapy offer the best results in survival in the age of HAART.

Objectives: To describe the clinical characteristics and response to treatment of the patients with HIV-associated lymphomas diagnosed in the INCAN from January, 2005 to December, 2010.

Material and methods: Descriptive, retrospective, longitudinal and observational study of patients with diagnosis of lymphoma and HIV infection at the INCAN. **Results:** We identified ninety-five patients 70.3 % of them between 30 and 49 years, with a male predominance (M:F 10:1) and advance stage in 72 %. The median CD4 cell count was of 141/dl. One third of patients (31.2 %) had extranodal primary disease. A trend to increase in case detection was observed in the last 3 years. There non-Hodgkin's lymphomas (87.4 %) predominated over Hodgkin's. DLBCL were the most frequent histological subtype (54/83, 65 %). The majority of the patients with non-Hodgkin's lymphoma received CHOP (40/83). Only 19/83 patients received DA-EPOCH, The global response (RC+RP) was 48 %; 38 % obtained CR: 26 % with CHOP and 42 % with DA-EPOCH. **Conclusion:** The characteristics of the Mexican population of patients HIV-associated lymphoma are similar to the world population. This one is the biggest series reported in Mexico. There were no complications for the use of chemotherapy and anti-retroviral therapy together. DA-EPOCH seems to be better than CHOP. In our hospital the economic resources of the patients limit the treatment of more patients with DA-EPOCH, even the use of rituximab. A cost-benefit study should be undertaken to prospectively compare between these principal schemes of chemotherapy

A1155

INCIDENCE OF DIFFUSE LARGE B CELL LYMPHOMA PRIMARY CUTANEOUS LEG TYPE IN A STATE OPER-

ATED ONCOLOGY CENTER IN ESTADO DE MÉXICO. Armenta-San Sebastián JA, Gutierrez-Serdán R, Gómez-Plata E, Díaz-Vargas G. Centro Oncológico Estatal I.S.S.E.M.Y.M.

Introduction: Diffuse large B cell lymphoma primary cutaneous leg type is a rare sub type of lymphoma compromising 4% of all primary cutaneous lymphoma and 20% of all primary cutaneous lymphomas. These lymphomas preferentially affect the lower legs, but 10-15% arise at other sites. **Objectives:** Establish the incidence of the Primary cutaneous DLBCL leg type in the overall population of lymphomas in a third level hospital in Estado de México. **Material and Methods:** We presented three cases of Primary Cutaneous DLBC leg type, one of them appeared in the right forearm, the other two appeared in the lower limbs: one in the right leg, and the other in the left inguinal zones. The one in the forearm was treated with surgical excision and then radiotherapy, the other two with chemo-immunotherapy (R-CHOP). **Results:** The two patients that receive R-CHOP 21 for 8 cycles obtain complete and durable response. The one with the surgical excision and radiotherapy was lost from follow up. **Conclusion:** We present three rare cases of DLBC lymphoma their treatment, outcomes and clinical photos.

A1158

PRIMARY ESOPHAGEAL KI-POSITIVE ALK+ ANAPLASTIC LARGE CELL LYMPHOMA OF T-CELL PHENOTYPE: A CASE REPORT AND LITERATURE REVIEW. Barbosa-Ibarra AA*, Ojeda-Tovar J*, Siñani-Cárdenas M*, Vázquez-Mellado A**, Rivas-Vera S*. * Hematology Department, Instituto Nacional de Cancerología. Mexico, DF. **Tecnológico de Monterrey, Facultad de Monterrey, Monterrey, N.L.

Introduction: Patients with lymphoma and gastrointestinal tract involvement (10-20 %) secondary to local invasion (stomach or mediastinum). The primary presentation of esophagus is rare (<1 %), the incident brought of 0.8-1.2 cases/100,000. More often in males and diffuse large B cells the most common. **Objectives:** To describe clinical and pathological characteristics of a patient with primary esophageal CD 30 positive ALK positive anaplastic large cell lymphoma. **Material and Methods:** Review of clinical record. **Results:** A 29 years old man, was referred to us for further management of dysphagia, he had difficulty in swallowing solids and over the past two months, without symptoms B. Hb: 14.6 g/dL, Ht:45.1 %, leucocyte count: 19 000/mm³, platelets:388 000/mm³, DHL:135UI/L, β2M:1.9 mg/L, The hepatitis and HIV tests were negatives. Upper endoscopic examination was realized by digestive hemorrhage and new episode of dysphagia. It revealed a multilobed, exofitic, infiltrating ulcerate lesion with circumferential diminishing 95 % of the esophagic diameter at 30 cm from the incisors, without involvement of the esophagus-gastric union, stomach and duodenum. The biopsy shows Anaplastic large cell Non Hodgkyn Lymphoma (NHL) of T-cell phenotype, positive for CD-30, MUM-1, ALK-1 (nucleolar, membrane and

granular), Ki-67(95%), CD-4, CD-45 and granzyme at atypical cells but negative for CD-20, CD-3, CD 138, BCL-6, CD-5, CD-8, CKAE/AE3, EBV, CE10, Ber-EP 4,CEA. Bone marrow without infiltration, PET scan showed mass in esophagus (SUVmax 26.6), peri-gastric adenopathies (SUV 14max), pancreatic-duodenal adenopathies (SUVmax 13.7). Stage IIaX (esophagus), IPI intermediate-low and t(2:5) (p23; q35) positive. He received CHOP-R improving ingestion but after 2nd cycle he progresses needing gastrostomy. He received second line with ICE presenting clinical improvement so we retire gastrostomy after the first cycle. Nowadays he had finished 4° cycle, he is asymptomatic and waiting extension studies to evaluate response. **Conclusions:** Our case corresponds to a 29 years old man with Anaplastic large cell NHL ALK-positive, subtype common, IPI intermediate-low (survival 5 years of 69 %). This is the first case reported in Mexico. There is not a treatment that demonstrates superiority and multidisciplinary approach is recommended with chemotherapy and radiotherapy.

A1160

INCIDENCE OF SECONDARY NEOPLASIA ASSOCIATED WITH MULTIPLE MIELOMA IN A STATE OPERATED ONCOLOGY CENTER. Armenta-San-Sebastián JA, Gutierrez-Serdan R, Gomez-Plata E, Lara-Torres C, Aguirre-Quezada D. Centro Oncológico Estatal I.S.S.E.M.Y.M.

Introduction: Of all the primary hematological neoplasm, multiple myeloma is the one associated with increased risk of a second malignancie. The incidence of this disease is 3% of all the types of multiple myeloma. This incidence is increasing with the better outcome in multiple myeloma. **Objective:** Establish the incidence of multiple myeloma and secondary neoplasm. **Material and methods:** Of 54 total cases of multiple myeloma in 6 years of the Centro Oncológico Estatal 5 patients have a secondary neoplasm three of them begin with the diagnosis of multiple myeloma and two of them have multiple myeloma as a secondary neoplasm. This corresponds to 9.25 % of all the patients. Multiple myeloma to a secondary neoplasm is 5.5%, and from a another neoplasm to multiple myeloma 3.7%. **Results:** The overall incidence of secondary neoplasm associated with multiple myeloma is 9.25% in our center, having fist multiple myeloma and then a secondary neoplasm is 5.5%, and primary neoplasm with secondary multiple myeloma 3.7%. **Conclusion** In literature the risk of secondary neoplasm in multiple myeloma is 1.19 times fold, the principal neoplasm are involving gastrointestinal tract and skin. In our series we have two breast cancers, one penial cancer, one mesothelioma and one gastric cancer. There still no evidence of increase risks by myeloma cell by itself neither therapy.

A1170

PRIMARY MEDIASTINAL LARGE B-CELL LYMPHOMA – RESULTS OF OUR CENTE Miljkovic E, Marjanovic G,

Markovic D, Nikolic V, Macukanovic-Golubovic L, Govedarovic N. Clinic of Hematology-Clinical Center Nis Serbia.

Introduction: Primary mediastinal large B-cell lymphoma (PMBCL) is a distinct clinico-pathological subtype of diffuse large B-cell lymphoma (DLBCL) and represents less than 3% of all non-Hodgkin lymphoma cases. The tumor appears to be derived from medullary B cells within the thymus gland. Phenotypically tumor cells are positive for CD45, B cell markers (Pax-5, CD19, CD20, CD22, CD79a) and bcl-2, often positive for CD30, MUM1 and bcl-6. and negative for CD10 and CD21. The optimal treatment is unknown, with some studies suggesting a superior outcome with dose-intensive chemotherapy regimens. The role of mediastinal radiotherapy upon completion of chemotherapy remains unclear. **Objectives:** Aim was to evaluate response in patients treating with R-MACOP-B regimen. **Material and methods:** We performed a retrospective review of 5 patients with PMBCL, who were treated at our clinic during the period between January 2009 and December 2010. **Results:** Among 5 patients there were 4 women and 1 man. Median age was 31 years (24-33). Most patients were in stage II-80% (4) and one in stage III-20%, all of them with B symptoms. Bulky mediastinal mass (>5cm in diameter) was present in all of our patients. Superior vena cava syndrome with facial edema, neck vein distention and upper extremity swelling was seen in 60% of the patients (3). After treating with R-MACOP-B regimen (rituximab, methotrexate, doxorubicin, cyclophosphamide, vincristine, prednisone, beomycin) complete response was accomplished in 40% (2), partial response in 20% (1). Two patients (40%) died, one due to progression of disease and one due to the toxicity of the treatment. Two patients who completed R-MACOP-B had involved-field radiation therapy (IFRT) following chemotherapy and complete remission was confirmed by performing PET scan. **Conclusions:** PMBCL, an uncommon disease affects young people with a female prevalence and frequent bulky mediastinal mass. Studies have demonstrated advantage of R-MACOP B compared to CHOP, so we treated our patients with this regimen. Two patients who completed R-MACOP B and had IFRT are still in complete remission after two years of follow up.

A1175

CLINICAL FEATURES OF 25 PATIENTS WITH HODGKIN LYMPHOMA. Ibarra-Hernández A***, Guerrero-Martínez AI***, Díaz-Ruiz R***, González-Moncada A***, Aguilar López LB****, Vega-Ruiz A****, Rubio-Jurado B****, García-Rincón IF****, Garcés-Ruiz OM****. *Servicio Hematología, UMAE, CMNO, IMSS

Introduction: Hodgkin lymphoma (HL) is less than 1% of all malignancies and 14% of lymphomas with an incidence of 4 cases per hundred thousand people a year, is more common in men. Limited-stage treatment has improved with the adoption of combination therapy, despite this, up to 10% of these patients may

have treatment failure. However, in advanced stages, up to 10% does not reach complete remission and 20-30% of patients will have relapsing response. **Objectives:** To determine, in patients with HL, its epidemiological and clinical characteristics and response to standard treatment. **Material and Methods:** It is a retrospective study with information obtained from clinical files. Patients with diagnosis of HL hospitalized in the hematology department of the CMNO IMSS Hospital the period 2009-2011. We recorded demographic data, histology, clinical stage and time of first relapse. **Results:** We included 25 patients, 15 male, mean age of 38 years, 10 women with mean age of 37 years. The predominant histological type of classical Hodgkin Lymphoma was mixed cellularity in almost half of patients. Response was observed for 44% of the population and mortality of 12%. **Conclusions:** Despite treatment with chemotherapy and radiation, we observed a mortality of 12% of the population, mainly due to advanced stages of disease. It is well known that the performance of PET imaging scan during surveillance allows early detection in relapsed and refractory Hodgkin Lymphoma.

A1180

PEMPHIGUS AND OBLITERATING BRONCHIOLITIS: MULTIORGANIC AUTOIMMUNE PARANEOPLASTIC SYNDROME IN A PATIENT WITH LYMPHOMA. De la Torre-Lujan AH*, Zapata-Canto NP*, Vega-González T**, Candelaria-Hernández M*, Rivas-Vera S*. *Department of Hematology, Instituto Nacional de Cancerología México DF; **Department of Dermatology, Instituto Nacional de Cancerología México DF.

Introduction: Paraneoplastic Pemphigus was first described in 1990. This disorder is characterized by mucocutaneous lesions similar to those of multiforme erythema. It has been related to lymphoproliferative diseases and with a high mortality rate. **Objective:** To describe the clinical, pathological and immunohistochemical characteristics of a patient with paraneoplastic autoimmune paraneoplastic syndrome and follicular lymphoma. **Material and Methods:** Review of Clinical record. **Results:** A 48 year old male who presented a mass in the right parotid in 2008, a biopsy was done outside our hospital: chronic follicular adenitis in the parotid gland. He notices a tumor despite the resection. CT showed a lesion of the parotid gland and a 95% pleural effusion in the left side, he had lost weight (10 kg), so he was referred to our institution but he abandoned treatment due to symptom improvement. In May 2011, he presented exacerbation of symptoms and oral ulcers that persisted despite medical care and dysphagia, the patient return to our hospital on May 2011. At that time he had an ECOG 1, cervical adenopathies and disseminated dermatitis in trunk, upper and lower extremities, formed by crusts, denuded areas and some vesicles of clear content predominantly in oral mucosa and genital area. A lymph node biopsy and new studies corroborate Follicular Lymphoma grade 2, stage IIIB. A biopsy of cutaneous lesions reported pemphigus vulgaris and he was treated with prednisone and drying soaks,

showed mild improvement. After 1st cycle of R-CHOP, lesion disappeared and a diagnosis of paraneoplastic pemphigus was considered. Five months later he developed dyspnea at rest and cough. CT showed heterogenous bilateral infiltrate that did not respond to medical treatment, becoming oxygen dependent. In January 2012 he was reviewed by neumologist, who diagnosed obliterating bronchiolitis. The evidence of bronchial epithelium affection has made us consider a multiorganic autoimmune paraneoplastic syndrome. **Conclusions:** Paraneoplastic pemphigus can appear in patients with lymphoproliferative diseases. Recently had been proposed diagnostic criteria for this disease: lymphoproliferative pathology, painful stomatitis, antiplaquin antibodies and acantolisis. Paraneoplastic pemphigus is the only form of pemphigus in which there is visceral affection due to an autoimmune process that can develop acantolisis in the bronchial epithelium. Pulmonary disease with obliterating bronchiolitis can cause more than 30% mortality in these patients. The knowledge of clinical manifestations in this autoimmune multiorganic paraneoplastic syndrome and its confirmation by simple laboratory techniques can lead to early detection of a hidden neoplasia, avoiding the development of respiratory disease.

A1181

SYNCHRONOUS NEOPLASMS IN A PATIENT WITH AIDS. REPORT OF A CASE. AT THE INSTITUTO NACIONAL DE CANCEROLOGÍA MÉXICO. Zapata-Canto NP*, De la Torre Lujan A*, Rivas-Vera S*, Pérez-Jiménez C**, Avilés-Salas A***. *Department of Hematology, Instituto Nacional de Cancerología, México; **Department of Infectology, Instituto Nacional de Cancerología México; ***Department of Pathology, Instituto Nacional de Cancerología México.

Introduction: Plasmablastic lymphoma (PBL) is a diffuse proliferation of large neoplastic cells most which resemble B immunoblasts, with a plasma cell-like immunophenotype. Immunodeficiency secondary to HIV predispose to the development of PBL. Tumor cells are EBV- infected in the majority of patients. The clinical course is very aggressive with most of the patients dying in the first year after diagnosis, although outcome may be improved with better management of HIV infection.

Objectives: To present a case of AIDS-related concomitant NHL and cervical cancer with a literature review. **Material and**

Methods: Review of the clinical records. **Results:** 27 year old heterosexual woman (2 sexual partners). She presented with abdominal pain and a left ovarian mass. Pathology report of the resected mass and cervical cone showed dysgerminoma with necrosis and hemorrhage and in situ cervical cancer. After six days she presented with fever, malaise, dyspnea, and a rapidly growing tumor mass on her left thigh. She was first seen at out institution a month latter with B symptoms and a left thigh 8x8x4 cm exophitic, verroucus and friable tumor. Revision of her outside pathology was reported as PBL expressing CD10, CD138 and MUM-1. Biopsy of the thigh mass was also reported

as PBL. Bone marrow aspiration corroborated infiltration. CT scan showed gastric fundus and retroperitoneal lymph node infiltration. She was found to be HIV Labs showed low CD4 (84) and high viral copy load (2,116,630). Epidermoid cervical cancer with glandular extension was corroborated, CKAE/AE3 positive. Interestingly, the cervix was also infiltrated by PBL. One cycle of CHOP chemotherapy was started with no improvement. Once HIV results were received she received second line chemotherapy with EPOCH and HAART with poor attachment to therapy. A week after the second EPOCH cycle she presented with rapid progression of the tumor on her left thigh and was admitted with septic shock from which she recovered. The tumor was refractory to treatment with a third EPOCH cycle and was voluntarily discharged. **Conclusions:** There are three AIDS-defining neoplasms, NHL, Kaposi's sarcoma and cervical cancer. The risk of development non-Hogkin lymphoma depends on the grade and duration of the immunodeficiency state (CD4 counts frequently bellow 200). Extranodal disease is frequent and may affect ovaries and soft tissue. This case shows concomitant stage IVBX NHL colocalized with cervical cancer.

A1184

A RETROSPECTIVE ANALYSIS OF PATIENTS WITH FOLLICULAR LYMPHOMA: OUTCOMES ACCORDING TO FLIPI AND TREATMENT. Zamora-Perez E*, Agreda-Vasquez GP*, Buganza-Torio E**, Gonzalez J***. *Department of Hematology INCMNSZ; **Department of Internal Medicine Hospital Angeles; ***Department of Internal Medicine Universidad Autonoma de Baja California.

Introduction: Follicular lymphoma (FL) is the most common indolent nodal lymphoma. Based on the number of centroblasts, the World Health Organization (WHO) classification divides FL into grades 1, 2, and 3. Patients with FL grade 1–2 and grade 3A do not have different outcome, but grade 3B would be a disease clinically more similar to Diffuse Large B Cell Lymphoma (DLBCL) –aggressive but curable with anthracyclines. The general status of the patient with FL is usually preserved, with few patients presenting with B symptoms or an altered performance status, although most of the patients do relapse after their treatment. **Objective:** To determine the clinical and biochemical characteristics of patients with FL and the outcomes according to score FLIPI and type of treatment. **Material and Methods:** This is a retrospective and retrolective analysis of patients with FL at the Instituto Nacional de Ciencias Medicas y Nutricion Salvador Zubiran, between January 2001 – January 2011. All cases were pathologically confirmed. We used the FLIPI score as prognostic index. Therapy was at the treating physician's discretion. Overall Survival were per Kaplan-Meier method. The statistical analysis was performed by SPSS 15.0. **Results:** Among 34 patients, the median age at diagnosis was 58 years (range 20–84); 16 female and 18 male. According to Berard classification, 32.35% were Grade 1(n=11); 55.88% Grade 2 (n=19) and 11.76% Grade 3

(n=4). We had no patient 3B Berard. According to FLIPI score, 26.47% (n=9) were Low, 50% (n=17) Intermediate and 23.52% (n=8) high. The bone marrow was involved in 67.64% (n=23). Fifty percent of patients were > 60years, 44.11% had =4 nodal sites involved, 23.52% had increased LDH, 8.8% Hb <12g/dl and 82.3% had advanced stage (= III). Twenty-six patients received treatment, there was a response rate of 88.46% (n=22) with first line treatment, the most used was CHOP 72.72% (16/22). Eight patients had no treatment at diagnosis, 5 patients continue without tumoral activity. Three had disease progression (37.5%). Twelve patients needed second line treatment(12/26: 46.15%), 3 with previous vigilance. The FLIPI score was the same as diagnosis in 8/9 patients with reactivation of the disease. The overall survival was of 91.2%, with a median follow up of 3134 days. **Conclusions:** The FLIPI score is related with age and biochemical characteristics (LDH and b2m). We had a median old age at diagnosis of patients with FL, most of them had advanced stage. Neither FLIPI nor Berard grade affected the good overall survival of these patients

Table 1. Characteristics of patients with Follicular Lymphoma and FLIPI

	Low FLIPI (n=9)	Int FLIPI (n=17)	High FLI- PI (n=8)	Total
Age	49.5	52.4	70.12	
>60y	2	8	7	17(50%)
Gender				
Female	5	6	5	16
Male	4	11	3	18
B symptoms	55%	64.7%	62.5%	
Bone marrow	22%	58.8%	37.5%	
Hb <12g/dl			3/8	
LDH >197		3/17	5/8	
B2m >2.5		7/17	4/8	
Treatment (n=26)				
CHOP	1/9	7/17	4/8	12
RCHOP	3/9		1/8	4
CHOP like		4/17	1/8	5
Other	0/9	4/17	1/8	5
No treatment	5/9	2/17	1/8	8

A1192

CLINICAL AND BIOCHEMICAL CHARACTERISTIC OF PATIENTS WITH NK/T-CELL LYMPHOMA, NASAL TYPE TREATED AT THE NATIONAL CANCER INSTITUTE (IN-CAN). Corrales-Alfaro CN*, Medina-Pérez C*, Barbosa-Ibarra AA*, Ojeda-Tovar J*, Zapata-Canto N*, Ramírez- Ibargüen AF*, Siñani-Cárdenas M*, Martínez- Balbuena M*, Gómez-Pérdomo G*, De la Torre – AHF*, Urbalejo-Ceniceros V*, Redondo-H*, Acosta-Maldonado B*, Cervera- Ceballos-EE*, Candelaria-Hernández M*, López-Navarro OG*, Toledano-Cuevas D**, Rivas-Vera S*, Labardini-Méndez JR*. *Department of Hematology, National Cancer Institute, Mexico; **Department of Radiotherapy, National Cancer Institute, Mexico

Introduction: NK/ T- cell lymphoma is predominant extranodal. It is characterized by vascular damage and necrosis, cytotoxic phenotype and very strong association with Epstein Barr virus (EBV) and geographic predilection (Asia, South America and Mexico). **Objectives:** To report the clinical, biochemical and treatment of patients with NK/ T-cell Lymphoma seen at the INCan in the last 7 years and determine the overall survival. **Material and Methods:** A retrospective cohort analysis of patients with NK/T-cell Lymphoma, between January 2005 to December 2011. **Results:** mWe identified 71 patients with NK/T-cell Lymphoma; the median age was 36 years (16-81), although the incidence was highest between 26 and 35 years by 32%. Male gender predominated (2.2:1). Just over half had good physical performance (ECOG: 1). The nasal cavity was the most frequent initial site (80%), followed by nasopharynx and palate. In 11% of patients documented infiltration of the central nervous system, B symptoms appeared in 48% of cases. Twelve patients were with advanced disease. According to IPI, 70% (n = 50) corresponds to IPI low. Among the biochemical characteristics emerged: nearly half of patients present with B2M level elevation, only a quarter some degree of hypoalbuminemia, while 42% had elevation of DHL and 24% with lymphopenia. To evaluate the outcome of chemotherapy and radiotherapy nasal natural killer (NK)/T-cell lymphoma: 87% of the patients received chemotherapy combined with radiotherapy (RT). The scheme used was CHOP in 87% of cases. The median radiation dose to the tumor bed was 45 Gy. The global response was 59%, 10% remained of the disease and 18% progressed. Relapse was documented in 18% of patients, local recurrence was 61.5%. **Conclusions:** In our population confirms the predominance of males and age of onset reported. It is noteworthy that in our country, the disease has been found located at the beginning. The overall response is similar to that reported in other studies including CHOP scheme and / or RT, however, is lower than reported when using patterns that include L-Asparaginase.

A1196

CLINICAL AND DEMOGRAPHIC CHARACTERISTICS OF PATIENTS DIAGNOSED WITH NON-HODGKIN DIFFUSE LARGE B-CELL AND FOLLICULAR LYMPHOMA SINCE JANUARY 2006 TO DECEMBER 2008 AT THE INSTITUTO NACIONAL DE CANCEROLOGIA, MEXICO. Ramírez-Ibargüen AF, Martínez-Balbuena MM, Siñani-Cárdenas M, Zapata-Canto NP, Barbosa-Ibarra AA, Medina-Pérez C, Corrales-Alfaro C, Ojeda-Tovar J, Acosta-Maldonado BL, De la Torre-Luján AH, Gómez-Perdomo G, Hernández-Redondo H, Urbalejo-Ceniceros VI, Candelaria-Hernández M, López-Navarro GO, Lozano-Zabaleta V, Rivas-Vera S, Labardini-Méndez JR. Department of Hematology, Instituto Nacional de Cancerología, Mexico, DF.

Introduction: Non-Hodgkin lymphomas are a heterogeneous group of malignant lymphoproliferative diseases and they are among the ten leading causes of death by cancer. The most

common is the diffuse large B-cell lymphoma (DLBCL) in 30-40%, followed follicular lymphoma (FL) in 20-30%. In the last decade have been described clinical, histological and molecular prognostic factors; as the identification of B cells germinal-center (GCB) or activated B-cell (ABC), which are associated with distinct genetic alterations and significantly different survival rates; also the expression of markers such as Bcl-2, Ki-67 were associated with different responses to treatment. **Objectives:** To describe the clinical and demographic characteristics of patients diagnosed with diffuse large B-cell and follicular lymphoma at the Instituto Nacional de Cancerología, Mexico. **Material and Methods:** We analyzed 338 patients records with diffuse large B-cell and follicular lymphoma from January 2006 to December 2008, with description of clinical and demographic characteristics. **Results:** A cohort of 338 files was analyzed, 259 (77%) patients had DLBCL and 78 (23%) FL, the median age was 59 years (20-90), nevertheless 55% of the patients was = 56 years, without predominance of gender (49% women and 51% men). We found at diagnosis 42% of ECOG 0 and only 10% with ECOG =3; 59% of the patients had B symptoms and up to 40% bulky disease. The advanced stage was the most common (43% of the patients had an EC IV and 20% EC III) and only 17% an EC I; with IPI intermediate high and high in 43%. Diffuse large B-cell lymphoma was not otherwise specified, was the predominant histology in 74%, follicular B-Cell lymphoma in 23%. Forty-two percent of the samples shows positive determination of Bcl-2, nevertheless in the rest 40% it was not processed. In FL's incident were no differences, 31% were first degree, 33% second degree and 36% third degree. Only 63% (215) of patients received as first line of treatment CHOP regimen, with overall response 84%, complete response in 66% and partial response in 18%. **Conclusions:** The LDCGB had a major incident that the LF; appearing mainly in patients of 6^a decade of the life, which coincides with the description of literature. Our patients appeared in advanced stages, with IPI intermediate-high or high in near 50%, with histological factors of bad prognostic as the expression of Bcl-2. We had overall responses of 66% with regimen CHOP.

A1214

AUTOLOGOUS STEM CELL TRANSPLANTATION IN PATIENTS WITH MULTIPLE MYELOMA AT THE INSTITUTO NACIONAL DE CANCEROLOGÍA, MEXICO. Calderon-Flores E, Medina-Perez C, Barbosa-Ibarra AA, Corrales-Alfaro C, Ojeda-Tovar J, Martínez-Balbuena MM, Siñani-Cárdenas M, Ramírez-Ibargüen AF, Zapata-Canto NP, Acosta-Maldonado BL, De la Torre-Luján AH, Gómez-Perdomo G, Hernández-Redondo H, Urbalejo-Ceniceros VI. Department of Hematology, Instituto Nacional de Cancerología. México, DF.

Introduction: Multiple myeloma (MM) is a clonal B-cell disorder characterized by abnormal accumulation of plasma cells and abnormal production of immunoglobulins, with the resulting cytopenias and bone resorption. This represent 10% of hematologic malignancies, the mean age at diagnosis is 65-70 years

old. In recent decades, different treatments have been reported with response rates of 60-80%, but disease-free survival still being short. The autologous stem cells transplantation is a treatment described with favorable responses; there are reports that described improve in response with tandem transplant. **Objectives:** To describe the clinical and demographic characteristics of patients with MM treated with autologous stem cells transplantation in the National Cancer Institute, Mexico. **Material and methods:** We reviewed records of patients diagnosed with MM who were transplanted from April 2001 to March 2011. **Results:** Thirty-nine patients were transplanted from April, 01 to March, 11: 41% were male and 59% female, mean age was 51 years old (37-64). Immunoglobulin G was reported in 51% and IgA 28%, with kappa light chain expression in 59% and lambda in 31%. The presence of Bence-Jones protein was found in 36% of patients. For the determination of prognostic index we found that 31% belonged to ISS-I, 41% to ISS-II and 18% ISS-III, the tumor burden was determined by Durie-Salmon score 56% of patients with DS -III. In 23% found at least one cytogenetic abnormality, the most frequent was the 13q deletion. The treatment used in 69% of the patients was thalidomide/dexamethasone, followed by 23% treated with VAD (vincristine, adriamycin and dexamethasone). Added to these 46% received radiotherapy. Overall response (OR) previously transplant was 82%, 23% had complete response (CR), 41% very good response (VGRP). Were performed 36 (92%) autologous transplants with melphalan-based conditioning and only 3 (8%) allogeneic transplant. The OR was 77%, 44% CR and 26% VGRP. Subsequently performed in 28 (71%) patients a second transplant in tandem achieving an OR of 89%, 54% CR and 21% VGRP. There were no reported deaths associated with any of the transplant. **Conclusions:** The mean age our patients was 51 years. MM IgG and kappa chain expression was the most common. Tumor burden corresponded to advanced stages with a DS III in 56%, however only 18% with an ISS-III. Prior to transplantation the majority had a VGRP and only 23% CR, after the first transplant achieved a higher number of CR (44%). With tandem transplants improved CR (54%), which reflects even better results.

A1215

HODGKIN LYMPHOMA: A DEMOGRAPHIC, CLINICAL, AND THERAPEUTIC DESCRIPTION OF A SINGLE INSTITUTION IN MÉXICO STATE. Armenta-San- Sebastian J, Gutiérrez-Serdán R, Díaz-Vargas G. Centro Oncológico estatal ISSEMYM.

Introduction: Hodgkin lymphoma is an uncommon malignancy involving lymph nodes and the lymphatic system, most patients are diagnosed between 15 and 30 years of years, followed by another peak in adults of 55 years old, It is now curable in at least 80%, with the standard therapy ABVD. **Objectives:** we report the results of a 5-year retrospective analysis in patients with Hodgkin Lymphoma who were treated with ABVD at our institute. **Material and Methods:** Our Institute is a third-level public

hospital which gives attention to patients older than 16 years of age requiring assistance in oncology areas. The data collected from the records of patients admitted between January 2006 and December 2011 with the diagnosis of Hodgkin's Lymphoma included: age, gender, pre-treatment full blood count values, type of therapy, response to therapy, status at latest follow-up, and causes of death. Follow-up was completed on December 31, 2011. **Results:** 51 cases were diagnosed as having Hodgkin lymphoma at diagnosis, 66% males, median age was 35 years, there were 56% nodular sclerosis, 39% mixed cellularity and 3.9% lymphocyte rich, according to the stage there were 5.88% early stage favorable and 21% were early stage unfavorable, and the remainder were advanced stage. All patients received ABVD 4 - 6 cycles, except 2 patients who received either 2 cycles of ABVD and one radiotherapy only, 6 patients died (11,76%) the cause of death are sepsis in nadir in 3 and 2 refractory disease. Median survival in all patients was 936 (50 - 1600) 4 patients relapsed. **Conclusions:** Hodgkin disease involves initial therapy with chemotherapy or combined modality therapy, we use the standard therapy ABVD and in our experience. In our experience is adequate standard therapy in advanced cases but more than 4 risk factors may try more aggressive therapies. The overall survival of our patients is similar to that reported in the literature. 3 patients died at nadir we did add to the schema colony stimulating factor.

A1216

MULTIPLE MYELOMA AND AMYLOIDOSIS AL. CASE REPORT AT THE INSTITUTO NACIONAL DE CANCEROLOGÍA MÉXICO (INCAN). Zapata -Canto NP*, Espinoza-Zamora R*, Candelaria-Hernández M*, Labardini-Méndez JR*, Avilés-Salas A**. *Department of Hematology, Instituto Nacional de Cancerología México; **Department of Pathology, Instituto Nacional de Cancerología México

Introduction Light chain amyloidosis (AL) is a low tumor burden plasma cell disorder characterized by deposition in the tissue of insoluble fibrils composed of immunoglobulin. The clinical manifestations depend of the organ involved. Congo red dye shows the amyloid deposits in affected tissues.

It can be found in the 85% of the cases in a bone biopsy or tissue. Amyloidosis has an inexorable progressive course due to uncontrolled tissue damage without treatment. The major predictor of overall survival has been the presence and extent of cardiac involvement. **Objective:** Differential diagnosis of a patient with Multiple Myeloma vs AL. **Material and methods:** Review the clinical record of a patient at the INCAN. **Results:** Women 59 years began on November 2010 with widespread bone pain. She attended to a physician who requested a protein electrophoresis test: positive to lambda light chain and a metastatic bone series without lytic bone lesions. He started treatment with cyclophosphamide, thalidomide, dexamethasone and melphalan for 9 months. On November 2011 she was hospitalized at the INCAN due to a skin infection on her right foot. Studies reported: hemoglobin:11.3g, Hematocrit:34.7%, VCM:102.2, Platelets:467, Albumin:2.5 gr/L, Globulins:2.5 gr/L, DHL:115IU/L, FA:174, protein electrophoresis: normal with no monoclonal peak, urine and serum immunofixation: lambda light chains. Bone marrow aspirate was normal but bone marrow biopsy reported: plasmacytic plasmacytoma, CD-138+ and lambda light chains positive. FISH negative for 13q deletion. At the Institute decided to continue with thalidomide and dexamethasone and discharging her from the hospital, after she improved. On June 2011, she began with tongue swelling, face edema, decided to hospitalize her, to be studied for amyloidosis, several biopsies were taken from bladder, tongue, kidney, all of them were positive for red Congo dye. No lytic lesions were observed at the metastatic bone series. Concluded Plasmacytoma in the bone marrow and AL, but the diagnosis of multiple myeloma could not be discharged because the patient had treatment when she came to the Institute. It was decided to continue on thalidomide and dexamethasone. **Conclusions:** Despite the treatment, she improved the symptoms and disease is more like an amyloidosis instead of myeloma. So we did not know how was made the diagnosis of multiple myeloma outside the Institute. The patient continues with thalidomide and dexamethasone with partial response. Primary amyloidosis represents 85% of the systemic amyloidosis in the U.S.A. but the differential diagnosis with multiple myeloma has to be made.

HEMOSTASIS AND THROMBOSIS

ORAL PRESENTATION

A1004

VTE DESPITE PROPHYLAXIS: WHATS THE NEXT STEP?

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Introduction: Venous thromboembolism (VTE) is a major cause of morbidity and mortality that affects more than one million patients every year. A number of global registries have documented poor compliance to thromboprophylaxis guidelines. Our hospital has adopted a systematic electronic admission policy to ensure compliance to guidelines. There still remains a subset of patients who develop VTE in hospital despite appropriate prophylaxis. To our knowledge, there has been no study that details this high risk group failing prophylaxis. **Objective:** Our aim was to characterize the population failing thromboprophylaxis and scrutinize our practice. **Material and Methods.** This was a retrospective cohort study reviewing charts beginning from 2007 to 2010, of all patients who developed VTE after 3 days of hospitalization. We analyzed the incidence of thromboembolic events, characterized the risk factor profiles of these high risk patients in addition to the choice of prophylaxis, treatments used and their outcomes.

Results: The incidence of VTE in patients despite prophylaxis was 0.15% (95% CI: 0.12% - 0.19%). Seventy-two patients developed VTE during this period, of which 58 had received appropriate prophylaxis. Enoxaparin was used for prophylaxis in 35 patients, while heparin was used in 23 patients. Of the 14 patients who did not receive heparin based prophylaxis, 13 had a relative or absolute contraindication for the use of heparin products. The mean age of patients was 65 years; and 57% of them were male. Thirty patients had a DVT, 17 had a PE while 11 were diagnosed with both. The average time for the development of a DVT or PE was 7 days. There was no significant difference between the patients in the heparin and enoxaparin groups. Patients with cancer, post orthopedic or surgical intervention, or sepsis/severe inflammatory response syndrome were at a high risk to develop VTE with an incidence of 88.9% (95% CI: 79.3% - 95.1%, p value < 0.000001). The incidence of treatment failure was 0.59% (95% CI: 0.45% - 0.76%). Implementation of

electronic records improved the rate of documentation of VTE prophylaxis from 55% to 80% (p=0.11) and risk stratification from 20% to 72% (p=0.0008). **Conclusions:** Our study details characteristics of this high risk patients failing prophylaxis. VTE during hospitalization is likely to worsen outcomes and thus must be prevented. Further studies are needed to determine the optimal dose of thromboprophylaxis in this high risk group, perhaps higher doses of heparin or enoxaparin. Electronic record keeping is likely to improve compliance to local VTE guidelines.

A1008

INCREASED LEVELS OF ADAMTS13 ACTIVITY IN PATIENTS WITH VENOUS THROMBOEMBOLISM. Mazetto BM, Orsi FLA, Barnabe A, Flores-Nascimento MC, de Paula EV, Annichino-Bizzacchi JM. Hematology-Hemotherapy Center, University of Campinas (UNICAMP), Campinas, SP, Brazil.

Introduction: Increased levels of inflammatory markers and clotting factors are related to the pathogenesis of VTE. The inverse relation between VWF and ADAMTS13 activity was previously described in patients with arterial thrombosis. VWF levels are known to be increased during inflammatory processes and could play a role linking the inflammatory and coagulation systems activities in VTE patients. **Objective:** To evaluate the activity of ADAMTS13 and VWF in VTE patients and its association with inflammatory markers and clinical outcome of post-thrombotic syndrome. **Material and Methods:** Seventy-seven VTE patients, 7 months to six years after the acute episode, attended at the Hemocentro de Campinas/UNICAMP, were included and 77 healthy controls matched by gender, age, ethnicity and blood group. The activity of ADAMTS13 was performed by VWF collagen binding, D-dimer by turbidimetry, CRP by nephelometry, and TNF- α , IL-6 and IL-8, VWF and ADAMTS13 antigen by ELISA. The presence of RVO was investigated by duplex examination and PTS by Villalta scale. **Results:** Thirty patients (39%) had VTE caused by transient risk factors, mainly the use of oral contraceptives, and 47 patients had spontaneous VTE. Serum levels of TNF- α and IL-6 were significantly increased in patients when compared to controls (median= 2.25 vs 1.59pg/mL, P=0.001; 1.16 vs 0.98pg/ml, P=0.013, respectively) whereas levels of IL-8 and CRP were similar among the groups (median= 18.3 vs 18.27pg/mL, p=0.47; 0.21 vs 0.17mg/dL, P=0.29, respectively).

Thirty-two patients (42,8%) had D-dimer > 0.55 mg/L and were defined as having increased coagulation activity. Inflammatory markers, such as TNF- α , IL-6, IL-8 and CRP, were significantly higher in those patients, comparing to patients with D-dimer = 0.55 mg/L (P=0.0057, 0.001, 0.0093 and 0.0075, respectively). The presence of PTS or RVO were not associated with increased inflammatory or coagulation activity. Only ADAMTS13-CBA and plasma levels of IL-8 were higher in patients with PTS comparing to patients without PTS. All inflammatory markers and coagulation parameters studied were similar in patients regardless the presence of RVO. **Conclusions:** Our findings suggest that there is an inflammatory and pro-coagulant activity in patients even after the acute episode of DVT. However, these activities were not related to the persistence of clinical and radiological sequels of DVT. Moreover, the increasing levels of VWF support the hypothesis that the inflammation is chronically activated. In this context, the increasing levels of ADAMTS13, also observed in patients, could be explained as a compensatory mechanism and maybe act as a protection against pro-thrombotic activity seen in these patients.

A1010

ENOXAPARIN ENCAPSULATED IN MUCOADHESIVE NANOPARTICLES FOR ORAL ADMINISTRATION AND ITS IN VIVO EVALUATION IN DVT MODEL. Prado LB*, Marcato PD**, Barnabé A*, Bassora FDS*, Paixão DS*, Annichino-Bizzacchi JM*. *Hematology and Hemotherapy Center, Medical Sciences Faculty, State University of Campinas (UNICAMP), Campinas-SP, Brazil; **Chemistry Institute, Biological Chemistry Laboratory, State University of Campinas (UNICAMP), Campinas-SP, Brazil.

Introduction: Deep vein thrombosis (DVT) is defined as a vessel occlusion of the venous system. The pathophysiology of DVT is based on the altered blood flow, vascular wall and / or blood elements. Enoxaparin is the anticoagulant used for the primary treatment of DVT in acute and prevention in patients at high risk for thromboembolic phenomena. The use of nanoparticles (NP's) for the encapsulation of drugs is a very interesting field, because this strategy could allow the drug concentration in plasma in the therapeutic range for an extended time, using fewer doses compared conventional forms and can improve the oral bioavailability of drugs. **Objective:** The aim of this study was the production of PCL-chitosan nanoparticles loading enoxaparin for oral administration in rats to assess the size of the thrombus formed. **Material and Methods:** NP's with enoxaparin was prepared by modified double emulsion as described by Meng et al. (2003). The size (Z-average) and surface charge (Zeta potential) of the NP's were analyzed using dynamic light scattering technique. The encapsulation efficiency (E.E.) of enoxaparin encapsulated in nanoparticles was quantified with Azure II colorimetric method modified as described by Lam et al (1976). For the animal experiment were used male Wistar rats aged 5-7 weeks and DVT was induced by the modified method

of Reyers et al (1980) and Carvalho et al (1992), and the method of hypercoagulability modified by Herbert et al (1992). Was administered a concentration of 2 mg/kg body weight of PCL-chitosan nanoparticles with enoxaparin and after one hour the thrombus was induced. 20 minutes after the animal was sacrificed and the thrombus was measured. **Results:** The average size of nanoparticles was 512.8 ± 13.8 nm and the zeta potential 30.9 ± 1.3 mV. The size of nanoparticles is very relevant to the pharmacokinetics, since the smaller particle is better absorbed by the wall of the gastrointestinal mucosa. The E.E. was $99.04 \pm 0.001\%$. The high E.E. was possible due to presence of chitosan. In animals, there was statistical difference of the weights of thrombi (34.30 ± 0.81 mg Control vs. NP's 2.55 ± 0.14 mg). There was no difference between the animals body weight. **Conclusions:** The enoxaparin encapsulated in mucroadhesive PCL-chitosan nanoparticles showed a good reduction in the thrombus after oral administration in rats with DVT induced showing an interesting system for oral administration of drugs.

A1121

VON WILLEBRAND FACTOR PLASMA LEVELS AND MULTIMERIC PATTERN IN ACUTE CORONARY SYNDROME. López-Castañeda SE*, Godínez-Hernández D**, Gutiérrez-Castellanos S*, Areán-Martínez CA***, Viveros-Sandoval ME*. * Laboratorio de Hemostasia y Biología Vascular, Facultad de Medicina, UMSNH, Morelia, México. ** Instituto de investigaciones Químico Biológicas, UMSNH, Morelia, México. ***Hospital General Dr. Miguel Silva, Morelia, México.

Introduction: The central role of Von Willebrand Factor (vWF) in thrombogenesis has made it a promising target for research into new antiplatelet therapies that specifically inhibit vWF. vWF plays a crucial role in platelet adhesion and aggregation at sites of high shear rates. Several studies have investigated the relationship between vWF plasma levels and thromboembolic cardiovascular events. Plasma levels typically rise during the course of acute coronary syndrome, and the extent of this vWF release is an independent predictor of adverse clinical outcome in these patients. Various lines of evidence indicate that vWF is not only a marker but also an important effector in the pathogenesis of myocardial infarction. vWF thrombogenic activity is proportional to multimer size but little is known about multimeric pattern in cardiovascular disease. **Objective:** To study vWF multimeric composition and plasma levels in patients with Acute Coronary Syndrome (ACS). **Material and Method:** 50 individuals, 30 patients with ACS and 20 healthy controls. Blood samples were obtained from each participant, Platelet-poor citrated plasma was prepared and stored at $-70 \text{ }^{\circ}\text{C}$. vWF antigen plasma levels were quantified by ELISA (American Diagnostica, USA), Modified vWF Multimer analyses was carried out using agarose and agarose/polyacrilamide gels. Plasma samples of Thrombotic Thrombocytopenic Purpura patients (TTP) were used as a positive control for HMWM. Blots were then incubated with rabbit anti-human vWF polyclonal antibody (Dako, California) followed

by polyclonal anti-rabbit-HRP conjugate (Dako), X-ray images were obtained (X-Ray Fujifilm products), scanned and analyzed. Optical density values representing the concentrations of vWF multimers of different molecular weights were plotted as densitographs. **Results:** 30 patients, 26 men (86,6%) and 4 women (13,33%), age (mean±SD) 61±10.3 years. 14(40%) had history of Cardiovascular disease, 20(66,6%) smoking habit, 11(36,6%) DM, 119(63,3%) of arterial hypertension, and 22(63.3%) atherogenic dyslipidemia. Diagnostic after Percutaneous Coronary Intervention: 19(63.3%) Acute myocardial infarction (AMI), 11(36.6%) unstable angina (UA). Patients with ACS had higher vWF plasma levels than healthy subjects ($p<0.001$), patients with AMI had higher vWF plasma levels than patients with UA ($p<0.001$). Patients with Ischaemic Cardiopathy had presence of High molecular weight von Willebrand factor (vWF) multimers (HMWM) in a multimeric pattern similar to that found in TTP. **Conclusion:** Presence of HMWM was demonstrated in Ischaemic Cardiopathy patients. The multimeric structure of vWF could be part of the alterations that generate thrombosis in this kind of patients. These findings outline the relevance of therapeutic interventions targeting vWF for acute coronary syndrome patients.

A1124

PLATELET REACTIVITY INDEX BY FLOW CYTOMETRY VASP/P2Y12 ASSAY TO EVALUATE RESPONSE TO ANTIPLATELET TREATMENT. Taboada-Cortina A*, Areán-Martínez CA**, Gutiérrez-Castellanos S*, García-Larragoiti N*, Solorio-Ramos R**, Viveros-Sandoval ME*. *Laboratorio de Hemostasia y Biología Vascular, División de Posgrado, Facultad de Ciencias Médicas y Biológicas Dr. Ignacio Chávez, UMSNH; **Servicio de Hemodinamia, Hospital General Dr. Miguel Silva, Morelia, México.

Introduction: Platelets play an important role in the pathophysiology of Acute Coronary Syndrome (ACS), so antiplatelet therapy has become a cornerstone in the prevention and treatment of this disease. Inhibition of platelet P2Y12 ADP receptor has reduced the incidence of major adverse cardiovascular events (MACE). However, there is a group of patients with low response to Clopidogrel identified as patients with high on-treatment platelet reactivity (HPR). The analysis of the phosphorylation status of vasodilator-stimulated phosphoprotein (VASP) by flow cytometry expressed as platelet reactivity index (PRI) has shown a predictive value for MACE post percutaneous coronary intervention (PCI). The use of flow cytometry for monitoring platelet function has several advantages with respect to platelet aggregation, considered until now as the gold standard. This is the first study in Latin America of determination of PRI by VASP/P2Y12 flow cytometry. **Objectives:** To determine Platelet Reactivity Index by flow cytometric analysis of platelet activity in patients on treatment with Clopidogrel. **Material and methods:** The present study was performed in 100 patients with ACS undergo-

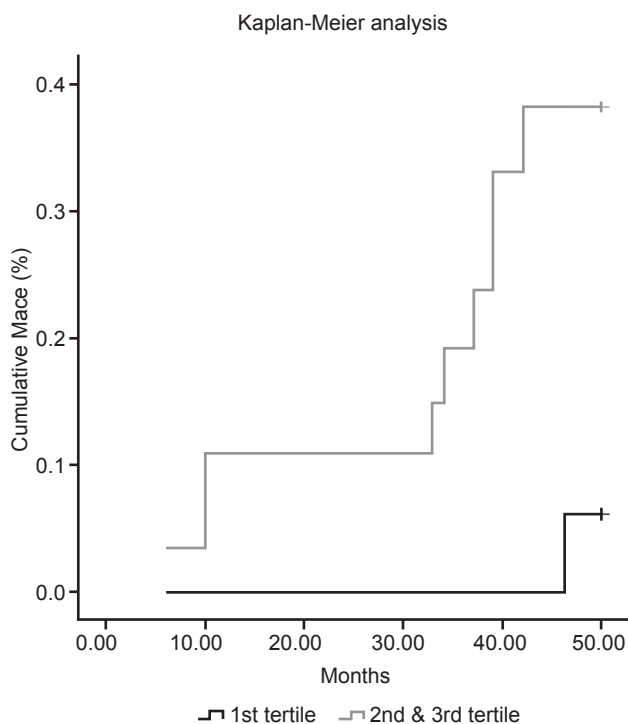
ing PCI. PRI was determined by VASP analysis (Platelet VASP/P2Y12 Biocytex, France) in flow cytometer (Beckman Coulter Epics Altra). Low response to Clopidogrel was defined as a PRI = 50% after 600 mg loading dose (Bonello L, 2010). Two blood samples for analysis: six to eight hours after administration of loading dose before PCI and 24 hours after PCI. **Results:** 100 patients: 77% men and 23% women, age (mean±SD) 62.66 ± 10 years in men and 65.58 ± 8.55 years in women. We found that 64.2% showed low response to Clopidogrel and 35.8% showed an adequate response to treatment. **Conclusions:** There is a high prevalence of High on-Treatment Platelet Reactivity (HPR) among patients treated with Clopidogrel. Response to Clopidogrel can be demonstrated by the persistence of VASP phosphorylation using flow cytometry. This method shows the degree of platelet P2Y12 receptor inhibition and allows the determination of specific response to Clopidogrel. The determination of PRI index allows identification of patients with low response to treatment that require adjustments in dose or switch to another antiplatelet treatment.

A1157

THROMBIN-ANTITHROMBIN COMPLEXES AND LONG-TERM PROGNOSIS FOR PATIENTS WITH STABLE CORONARY ARTERY DISEASE. Valente-Acosta B***, Baños-González MA*, Peña-Duque MA*, Martínez-Ríos MA*, Quintanar-Trejo L*, Cruz-Robles D*, Cardoso-Saldaña G*, Rodas-Arvizu R**, Fernández-Pellón R**, Aptilon-Duque G**, Reyes-Soto A**, de la Peña-Díaz A***. *Instituto Nacional de Cardiología Ignacio Chávez, Grupo Genética Intervencionista, Departamentos de Biología Molecular, Hemodinámica, Endocrinología. México. **Departamento de Farmacología, Facultad de Medicina, Universidad Nacional Autónoma de México, México.

Introduction: Thrombin has a role regulating the atherosclerotic progression, it is neutralized by its physiological inhibitor antithrombin and thus thrombin-antithrombin (TAT) complex is a reliable marker of thrombin generation. Several follow-up studies have shown an association between thrombin and the prognosis of ischaemic heart disease, although most of the studies were done in patients with an acute coronary syndromes, so the prognostic value of thrombin generation in patients with stable coronary artery disease (CAD) is still unrevealed. **Objectives:** Evaluate the role of TAT on the long-term cardiovascular prognosis in patients with stable CAD. **Material and methods:** Forty-seven patients with stable CAD were monitored during 50 months, the primary end point was the incidence of MACE (Major adverse cardiovascular event) that was compared across the first tertile versus the second and the third tertile of the TAT concentration. Comparison between groups was carried out using the log-rank test. **Results:** There was one death registered in a patient in the third tertile. In the multivariate analysis, the second and third tertile of TAT levels shown an increased risk of MACE compared with the first (OR= 9.26, CI=1.076-79.716, $p=0.043$).

The cumulative incidence of MACE at 50 months of follow-up is shown in figure 1. **Conclusion:** High TAT concentration is an independent risk factor for MACE in patients with stable coronary artery disease in follow-up.



A1232

ASSOCIATION OF PAI-1 LEVELS AND SEXUAL HORMONES WITH METABOLIC SYNDROME IN ELDERLY

MEN. Vázquez AL, Córdova N, Saucedo R, Vega S, Martínez-Murillo C, Gaminio E, Gilván R, Rocha A, Zárate A, Basurto L. Endocrine Research Unit, National Medical Center, Instituto Mexicano del Seguro Social and Thrombosis and Hemostatic Unit, General Hospital of Mexico SSA.

Introduction: The alterations of the hemostasis of Metabolic Syndrome (MS) that favor a prothrombotic state include modifications in the coagulation and fibrinolytic systems. Testosterone is also functionally connected with hemostasis; a negative correlation was found between testosterone and PAI-1 levels in males. In the other hand, the effects of estrogens on cardiovascular risk factors have been less well defined in men than in women. **Objectives:** The aim of this study was to analyze the relationship of PAI-1, estradiol and testosterone, with metabolic syndrome in elderly men. **Material and Methods:** Cross-sectional study including 97 healthy men aged 60-80 years. MS was defined according to the International Diabetes Federation. Main outcome measures were PAI-1 estimated by ELISA, estradiol and testosterone by radioimmunoassay. **Results:** Of the total of 97 participants, 33% had MS, and 77% were healthy men. PAI-1 levels of control group were 73.7 ± 36 pg/mL, in comparison with 92.8 ± 83.8 of men with MS. The estradiol levels in both groups were respectively 20.7 ± 8.6 and 28.0 ± 11.2 pg/mL, $p < 0.05$. By contrast, testosterone levels were similar in both groups. The negative correlation between PAI-1 and estradiol was $r = -0.249$, $p < 0.05$; this correlation remained significant after adjustment for BMI and age. It was also been observed that PAI-1 correlated with glucose ($r = 0.256$, $p < 0.05$), cholesterol ($r = 0.283$, $p = 0.01$) and weight ($r = 0.206$, $p < 0.05$). Testosterone was not correlated with PAI-1 levels. **Conclusions:** In men, low concentrations of E2 were associated with increased of PAI-1, independent of other traditional cardiovascular risk factors. Thus, these data suggest that thrombotic state in men could be determined by estradiol.

POSTERS

A1003

ACETYLSALICYLIC ACID THERAPY: INFLUENCE OF METFORMIN USE ON URINARY 11-DEHYDRO THROMBOXANE B2 LEVELS. Carvalho MG*, Gonçalves LH**, Dusse LMS*, Fernandes AP*, Gomes KB*, Silva MVF*, Lages GFG*, Komatsuzaki F*, Sousa MO*, Bosco AA***. *Clinical and Toxicological Department-Faculty of Pharmacy-Federal University of Minas Gerais, Belo Horizonte, Brazil; **Rede Sarah de Hospitais de Reabilitação, Belo Horizonte, Brazil; ***Santa Casa Hospital, Belo Horizonte, Brazil

Introduction: Acetylsalicylic acid (ASA) is the antiplatelet agent most commonly used for preventing thromboembolic events in patients with risk factors. The effect of ASA may be measured through the analysis of the urinary levels of 11-dehydro throm-

boxane B2 (11-dhTXB2), a metabolite of thromboxane A2, which is a potent platelet aggregant agent. It has been suggested that metformin could improve oxidative stress and control platelet activation in type 2 diabetic patients, potentially reducing cardiovascular risk. **Objective:** Our aim was to determine the levels of urinary 11-dhTXB2 in type 2 diabetic patients taking ASA and associate its levels with metformin use and other clinical variables, considering as a target a reduction of at least 75% in the levels of this marker compared to results before ASA intake. **Material and Methods:** The urinary levels of 11-dhTXB2 of 81 type 2 diabetic patients were measured before and after 15 days taking 100 mg of aspirin daily. The clinical variables evaluated were: C-reactive protein (CRP), glycosylated hemoglobin (HbA1c), hypertension, polymorphisms in the genes of COX-1 and GPIIb/IIIa and metformin (an oral hypoglycemic agent) use. All

statistical analysis were performed by using Pearson Chi-Square test. **Results:** It was observed that mostly patients who presented a reduction of 11-dhTXB2 above 75% was under metformin use. This reduction was achieved in 51.5% of patients taking this drug, against 20.0% in the patients who wasn't ($p=0.027$). The analysis of the other variables related to a reduction $\geq 75\%$ did not show a significant difference, once all p values were $>0,05$. According to previous reports, hyperglycemia control by itself seems to be a determinant factor for the success of ASA therapy, explaining the influence of metformin in the reduction of 11-dhTXB2 levels. **Conclusions:** Based on the results, the use of metformin seems to play a role in the reduction of 11-dhTXB2 levels in type 2 diabetic patients.

A1006

LOW DOSE RITUXIMAB WITH PLASMA EXCHANGE AS FIRST LINE TREATMENT IN THROMBOTIC THROMBOCYTOPENIC PURPURA IN A HUMAN IMMUNODEFICIENCY VIRUS INFECTED PATIENT. Pequeño-Luévano M*, Jaime-Pérez J*, Cantú-Rodríguez O*, Gutiérrez-Hernández H*, Tarín-Arzaga L*, Gómez-Almaguer D*. *Departamento de Hematología. Hospital Universitario "Dr. José Eleuterio González".

Introduction: Thrombotic thrombocytopenic purpura (TTP) is a rare disorder that was relatively common among human immunodeficiency virus infected (HIV) individuals before the introduction of highly active antiretroviral therapy (HAART). The current therapy goal is to eliminate ADAMTS13 antibodies with plasma exchange, but some cases are refractory to this first line therapy. Rituximab has been previously used as a second line treatment with promising results. Previously, rituximab has produced remission in a significant proportion of patients with TTP refractory to plasma exchange, lowered plasma exchange requirements and avoid the complications of other immunosuppressive therapy. **Objective:** We report on a newly diagnosed HIV patient with acute TTP, who received plasmapheresis and low-dose rituximab, in order to show that this treatment modality might have a role in similar cases. **Material and Methods:** A 33 year-old female patient with altered consciousness and fever was admitted to the "Dr. José Eleuterio González" University Hospital of the Medicine School of the Autonomous University of Nuevo Leon in Monterrey, México. She presented with normocytic normochromic anemia, thrombocytopenia, incipient renal failure, indirect hyperbilirubinemia, and an important elevation of the lactate dehydrogenase. The presence of multiple schistocytes and thrombocytopenia were documented. During initial assessment, a positive HIV ELISA test was documented, which was afterwards confirmed by Western- Blot. Plasma exchange and rituximab 100 mg/m² IV after the plasmapheresis session per week were started. **Results:** After the fourth plasma exchange, the patient showed improvement reflected by the degree of orientation and normothermic state, as well as improvement

of blood and biochemical parameters. During the ambulatory five month follow up the patient has shown a progressive improvement in hematologic and biochemical parameters, and currently is in complete remission. **Conclusions:** We suggest that, in combination with plasmapheresis, low-dose rituximab could have a role as first line therapy in TTP HIV-positive. Additional studies combining these two therapeutic approaches are needed to better define the optimal treatment of TTP in HIV infected patients.

A1007

IDENTIFICATION OF DIFFERENT EXPRESSION IN PLASMA SAMPLES FROM DEEP VENOUS THROMBOSIS PATIENTS. Flores-Nascimento MC*, Paes-Leme AF**, de Paula EV*, Zanella JL**, Annichino-Bizzacchi JM*. * Hematology-Hemotherapy Center, State University of Campinas (UNICAMP), Campinas, SP, Brazil; ** Biosciences National Laboratory (LNBio), Campinas, SP, Brazil.

Introduction: Deep venous thrombosis (DVT) is multi-causal disease associated to a high morbi-mortality due to complications as pulmonary embolism and post-flebotic syndrome, and around 30 % of the patients will present recurrence in 5 years. The identification of new risk factors is important on the clinical practice to prevent new thrombotic events. The plasma has all the anticoagulant and procoagulant factors, it is the media for all the blood cells and many proteins associated to immune and inflammatory functions, and potentially provides a window into the individual's state of health and diseases. **Objective:** Based on this we analyzed the plasma protein profile of samples from 3 DVT patients and compared to results obtained from 1 sibling and 1 neighbor for each patient in order to minimize the genetic and environmental interferences. These patients presented spontaneous and recurrent episodes of proximal DVT and mentioned a familiar history of coagulation disorders. **Material and Methods:** The plasmas samples were separated and the albumin was removed by Affi-gel Blue gel. The proteins were alkylated with DTT, reduced with iodoacetamide, precipitated with acetone and hydrolyzed by trypsin. The peptides were desalted in C18 columns and then separated by liquid chromatography cation exchange. The 7 fractions obtained were directed to the ESI Q-TOF Premier mass spectrometer. The proteins search was performed by Mascot. **Results:** The individual analysis of proteins showed that a significantly higher number of peptides from complement C4-A plasma protease, C1 inhibitor Inter-alpha-trypsin, inhibitor heavy chain H1, serum amyloid A and isoform gamma-B of fibrinogen gamma chain protein were hyper expressed. Peptides from alpha-2-HS-glycoprotein and apolipoprotein A-IV were present in reduced amounts were on the patient samples. **Conclusions:** In this study it was possible to identify some proteins up to date non-related to the physiopathology of DVT, which could be involved with immune, metabolic and inflammatory processes.

A1016

HOMOCYSTEINE PLASMA LEVELS IN ELDERLY PEOPLE WITH AND WITHOUT MILD COGNITIVE IMPAIRMENT AND WITH ALZHEIMER DISEASE. Gonçalves G S*, Silva MVF*, Silveira JN*, Carvalho MG*. * Faculty of Pharmacy, Federal University of Minas Gerais, Belo Horizonte, Brazil.

Introduction: Alzheimer's disease (AD) is the most common dementia among older people in the world, which involves many costs to public health. A key event in the pathogenesis of AD is the formation of insoluble aggregates of amyloid beta peptides. Microvascular changes can be observed in the brains of patients with AD. These changes have been associated with the disease process and may precede neurodegeneration. Thus, the assessment of microvascular pathology provides a promising approach to developing biomarkers useful for early detection and characterization of AD pathology. There is evidence that hyperhomocysteinemia may be an important risk factor for the development of AD since it may potentiate the neuronal and endothelial damage, leading to vascular disease. **Objective:** The aim of this study was to determine plasma levels of total homocysteine (Hcy) in a set of patients with AD or with mild cognitive impairment (MCI), in Minas Gerais State, Brazil, and compare them to a control group with no history of disease. **Material and Methods:** Sixty 60 patients with AD and 43 with MCI treated at University Hospital of the Federal University of Minas Gerais (UFMG), Brazil, were selected besides to 60 healthy individuals (controls). Plasma levels of (Hcy) were measured using (HPLC) with fluorimetric detector, at the Laboratory of Toxicology, Faculty of Pharmacy / UFMG. Firstly, thiol groups were reduced and followed by protein precipitation and subsequent derivatization of Hcy in the 7-fluorobenzo-2, 1,3 - oxadiazole-4-sulfonic acid (SBD-F). **Results:** The results obtained for Hcy were 13.85 ± 7.69 ; 15.45 ± 5.62 and 11.99 ± 14.12 mmol / L. for controls, and group with AD or with MCI, respectively. There was no significant difference in plasma levels of Hcy among the three groups ($p = 0.523$, Kruskal Wallis test). However, the results obtained for the control and CCL are within the reference values from 5.0 to 15 mmol / L while the group with AD showed a tend to higher values. **Conclusions:** Hcy plasma levels were not different among elderly people with and without MCI and in those with AD. Replacement of vitamins is very common among elderly people, which may contribute to reduce plasma levels of Hcy. Supported by CNPq and FAPEMIG, BRAZIL.

A1018

ANTIHEMOSTATIC EFFECT OF A PROTEOLYTIC FRACTION FROM CARICA CANDAMARCENSIS LATEX ON WISTAR RATS PLASMA, IN VITRO. Bilheiro RP*, Carvalho MG**, Bravo CES*, Lopes MTP*. *Department of Physiology and Pharmacology, Institute of Biological Sciences, Federal University of Minas Gerais, Brazil; **Department of Clinical and Toxicological Analysis, Faculty of Pharmacy, Federal University of Minas Gerais, Brazil.

Introduction: Carica candamarcensis, a plant native of South America, produces a fruit rich in latex. Our research group has been involved in the biochemical, pharmacological and toxicological characterization of such latex, from which a fraction rich in cysteine proteases, named P1G10, has been purified. During the toxicological evaluation of P1G10, signs of hemorrhage were observed in Wistar rats. Also, the literature describes various effects of cistein proteases on hemostatic patterns. **Objective:** We evaluated the effect of the fraction P1G10 on both plasma coagulation and platelet aggregation. **Material and Methods:** In the coagulation assays, plasma samples of 50 microliters of Wistar rats ($n=5$) were incubated at 37°C with either P1G10 or saline for 5 minutes (final concentrations: 0.25; 0.50 and 1.0 micrograms/microliter). Activated Partial Thromboplastin Time (APTT) (Actin®, Dade-Behring, USA), Prothrombin Time (PT) (Thromborel®, Dade-Behring, USA) and Thrombin Time (TT) (Triniclot Fibrinogen®, Trinity Biotech, Ireland) assays were then performed in a coagulometer (BFT II®, Dade-Behring, USA). Platelet aggregation assay was performed at 37°C using an aggregometer (Packs-4®, Helena Labs., USA) after a 10 minutes incubation of either P1G10 or saline with 450 microliters of a pool ($n=4$) of platelet-rich plasma (PRP) (final concentrations: 0.25; 0.50 and 1.0 micrograms/microliter) from Wistar rats. ADP (600 microM) was used as aggregating agent. **Results:** P1G10 increased PT (1.4 fold) at 1.0 micrograms/microliter, whereas a dose-dependent increase in APTT was found at 0.5 micrograms/microliter (2.5 fold) and 1.0 micrograms/microliter (4.8 fold) and by TT at 0.5 micrograms/microliter (2.5 fold) and 1.0 micrograms/microliter (4.4 fold). P1G10 also inhibited significantly platelet aggregation by 23% at 0.5 micrograms/microliter and by 44% at 1.0 micrograms/microliter. **Conclusions:** Taken together, these results strongly suggest that P1G10 has a significant antihemostatic activity, which is most likely due to fibrinogen cleavage. Supported by FAPEMIG, CNPq, CAPES.

A1019

HEMOSTATIC AND LIPID PROFILE OF PATIENTS WITH HYPERTHYROIDISM BEFORE AND AFTER TREATMENT. HEMOSTATIC AND LIPID PROFILE OF PATIENTS WITH HYPERTHYROIDISM BEFORE AND AFTER TREATMENT. Carvalho MG*, Vieira LM*, Feranandes AP*, Borges MA**, Rosário PW**, Dusse LMS*, Rodrigues KF*, Sousa MO*. *Department of Clinical and Toxicological Analysis, Faculty of Pharmacy, Federal University of Minas Gerais, Brazil; **Santa Casa Hospital, Belo Horizonte, Brazil.

Introduction: Various abnormalities in the coagulation and fibrinolytic systems as well in lipid profile have been reported in patients with thyroid dysfunction. Several studies indicate that hyperthyroidism state is associated with hypercoagulability, hypofibrinolysis, endothelial dysfunction and hypocholesterolemia. **Objective:** The aim of this study was to evaluate the hemostatic and lipid profiles in a set of Brazilian subjects with hyperthyroid-

ism and correlate these data with free T4 (fT4) levels before and after treatment. **Material and Methods:** Twenty-four patients with hyperthyroidism were selected in Santa Casa Hospital, Belo Horizonte, Brazil. D-Dimer, PAI-1, F1+2 and thrombomodulin was measured in citrate plasma samples; lipid profile (ApoA, ApoB, Lpa) and fT4 levels were measured in serum samples. All tests were performed using diagnostic kits. Statistical analyses were done by Wilcoxon and Spearman tests. A value $p < 0.05$ was considered statistically significant. **Results:** Hemostatic profile was not significantly different when compared before and after treatment. However, similar to other studies, PAI-1 levels were elevated in patients with hyperthyroidism even after treatment. By other hand, although within normal range, it was observed a slight elevation in the levels of ApoA, ApoB and Lpa after treatment which is consistent with increased turnover of cholesterol observed in states of hyperthyroidism. ApoB and Lpa levels were significantly different when compared before and after treatment (ApoB $p = 0.001$; Lpa $p = 0.039$), while ApoB and fT4 levels, after treatment, showed an inverse and significant correlation ($r = -0.595$, $p < 0.01$). **Conclusions:** Results suggest that both ApoB and Lpa levels suffer significant influence upon thyroid status. In addition, increased PAI-1 in these patients represent a potential hypofibrinolytic state. Further studies are needed to better assess the influence of thyroid hormones on hemostatic and lipid profiles. Supported by CNPq and FAPEMIG.

A1021

HEMOSTATIC AND LIPIDIC VARIABLES IN NORMOLIPIDEMIC AND DYSLIPIDEMIC SUBJECTS. Sousa MO*, Ferreira CN**, Lima LM***, Gomes KB*, Pinheiro P*, Lages GFG*, Fernandes AP*, Carvalho MG*. *Department of Clinical and Toxicological Analysis, Federal University of Minas Gerais, Belo Horizonte, Brazil; **COLTEC, Federal University of Minas Gerais, Belo Horizonte, Brazil; ***Department of Medicine and Nursing, Federal University of Viçosa, Brazil.

Introduction: Dyslipidemia is an important risk factor for atherosclerosis, which is characterized by alterations in endothelium and coagulation. **Objective:** The study evaluated the association between hemostatic and lipidic variables in normolipidemic and dyslipidemic subjects. **Material and Methods:** Dyslipidemic ($n=109$) and normolipidemic ($n=107$) subjects were selected from Brazilian population in Minas Gerais state. Venous blood samples were drawn after an overnight fast. Plasma Thrombomodulin (TM), Fragment 1+2 (F1+2), Plasminogen activator inhibitor-1 (PAI-1), Thrombin-activatable fibrinolysis inhibitor (TAFI) and D-dimer (D-Di) were assessed by ELISA methods. Total cholesterol (TC), triglycerides (TG) and HDL cholesterol levels were measured with Cobas Analyzer. LDL-c levels were estimated by Friedewald equation. For statistical analysis were performed the tests: One-way ANOVA, Student-Newman-Keuls and Pearson correlation. p -value < 0.05 was considered as significant. **Results:** TM, F1+2, D-DI and PAI-1 levels were similar between groups.

However, dyslipidemic subjects presented higher levels of TAFI compared to normolipidemic ones ($p < 0.001$). Dyslipidemic women also showed higher levels of TAFI compared to normolipidemic ones, as well, compared to men from both the groups ($p < 0.05$). Considering all participants women presented higher F1+2 ($p < 0.01$) and D-Di ($p < 0.01$) levels compared to men. A significant correlation between F1+2 and D-Di was observed ($r = 0.22$; $p < 0.01$). TAFI levels presented a positive correlation with TG ($r = 0.346$; $p < 0.001$) and LDLc ($r = 0.199$; $p < 0.01$) and negative with HDLc ($r = -0.278$; $p < 0.001$). **Conclusions:** Results suggest that dyslipidemia is associated with TAFI levels, while women particularly those dyslipidemic ones appear to show an unfavorable hemostatic status. Also, in these patients TAFI levels have been associated to an undesirable lipid profile potentially increasing the risk for atherosclerotic events. Acknowledgements: FAPEMIG and CNPq, Brazil

A1025

MICROPARTICLES DERIVED FROM PLATELETS AND SEVERE PRE-ECLAMPSIA. Marques FK*, Carvalho AT**, Nunes FFC**, Carvalho MG***, Dusse LMS***, Gomes KB1***. *Department of General Biology, Institute of Biological Sciences, Federal University of Minas Gerais, Belo Horizonte, MG, Brazil; **Centro de Pesquisas Rene Rachou, FIOCRUZ, Belo Horizonte, MG, Brazil; ***Department of Clinical and Toxicological Analysis, Faculty of Pharmacy, Federal University of Minas Gerais, Belo Horizonte, MG, Brazil.

Introduction: Pre-eclampsia (PE) is a multi-system disorder, characterized by hypertension and proteinuria, occurring after 20th week of pregnancy. Microparticles (MPs) are small vesicles released after cell activation or apoptosis and have been proposed to play a role in thrombosis, inflammation and angiogenesis. Studies have shown that MPs are usually increased in pregnancy; however, this increase is especially important in pregnant women with PE, who show an extensive activation of endothelial cells, leukocytes, and the coagulation system. **Objective:** To measure MPs derived from platelets (PMP) in severe pre-eclampsia (sPE), in normotensive pregnant and in non-pregnant women. **Material and Methods:** Twenty-eight pregnant women with sPE (group 1), 28 normotensive pregnant (group 2) and 28 non-pregnant women (group 3) were enrolled in this study. Severe PE was defined by systolic blood pressure ≥ 160 mmHg or diastolic blood pressure ≥ 110 mmHg; and proteinuria > 2 gL⁻¹ or at least 2+ protein by dipstick. Normal pregnant women had systolic/diastolic blood pressure below 120/80 mmHg and no history of hypertension or proteinuria. All pregnant women had gestational age ≥ 29 weeks. Non-pregnant women had no clinical and laboratory alterations. The MP phenotype was analyzed by flow cytometry using the classical MP marker, annexin, and fluorochrome-labeled monoclonal antibodies against CD41 marker. **Results:** Median levels of PMP observed in each group were: 3.76 for sPE group; 1.74 for normotensive pregnant group; and

3.44 for non-pregnant women. When the groups were compared by Mann-Whitney test, no significant difference was observed ($p=0.14$ for groups 1x2; $p=0.93$ for groups 1x3; $p=0.09$ for groups 2x3), although it was observed a trend towards decreased levels in group 2. When the three groups was compared by Kruskal-Wallis test, it was not observed difference between the groups ($p=0.18$). **Conclusions:** Although a reduction in platelet count is often observed in sPE, there was a trend to higher PMP levels in this group when compared to the normotensive group, suggesting greater cell activation or apoptosis in sPE pregnant women, associated to extensive activation of coagulation system. Acknowledgments – CNPq, Brazil

A1026

BIOCHEMICAL AND HEMOSTATIC BIOMARKERS FOR PREDICTING CORONARY ARTERY DISEASE. Lima LM*, Carvalho MG**, Vieira LM**, Junqueira DRG**, Sousa MO**. *Department of Medicine and Nursing, Federal University of Viçosa, Minas Gerais, Brazil; **Department of Clinical and Toxicological Analysis, Federal University of Minas Gerais, Belo Horizonte, Brazil.

Introduction: Biomarkers can be used in the early detection of subclinical coronary artery disease (CAD), selection of therapy, diagnosis of acute coronary syndromes and risk stratification. This requires a clear understanding of the validity of biomarkers available to predict the disease. **Objective:** This study evaluated five biomarkers related to different pathophysiological pathways and their associations with CAD, trying to identify high-risk patients for CAD with non-invasive diagnostic techniques. **Material and Methods:** The study has involved 31 patients with mild/moderate atheromatosis and 57 patients with severe atheromatosis, assessing by coronary angiography. The discriminant power was determined for anti-oxidized LDL, high-sensitivity C-reactive protein (hs-CRP), phospholipase A2 (PLA2), plasminogen activator inhibitor type 1 (PAI-1) using the area under the receiver-operating-characteristic (ROC) curve. We also studied cut-off points which would best optimize the trade-off between sensitivity and specificity. **Results:** The discriminant power of anti-oxidized LDL, hs-CRP and PLA2 did not show difference among them. However, a higher plasma PAI-1 concentration ($>53.7\text{ng/mL}$) showed the greatest specificity (94%) and positive predictive value (71%) for coronary atheromatosis, but low sensitivity (60%). The area under the curve (AUC) for PAI-1 was 0.705 (0.598 – 0.798, $p=0.0003$). **Conclusions:** The results obtained by using AUC allow to conclude that higher levels of PAI-1 showed to be the best predictor of coronary atheromatosis compared to anti-oxidized LDL, hs-CRP and PLA2 in the studied patients. Sponsored by CNPq and FAPEMIG, Brazil.

A1027

CROTALUS DURISSUS ACCIDENT: CHANGES IN NATURAL ANTICOAGULATION AND FIBRINOLYTIC SYSTEMS.

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Introduction: Snakebite envenoming is an important public health problem in many tropical and subtropical developing countries. *Crotalus durissus* rattlesnakes are responsible for the most lethal cases of snakebites in Brazil. Disturbances in hemostasis including spontaneous bleeding and blood uncoagulability are important symptoms of these accidents and are correlated with the action of snake venom metalloproteinases. **Objective:** This study has as aim to investigate the changes in the natural anticoagulant and fibrinolytic systems in patients who suffered *C. durissus* accident. **Material and Methods:** Thrombomodulin (TM), activated protein C (aPC), activated factors V (FVa) and VIII (FVIIIa), plasminogen, fibrin degradation products (FDP) and plasminogen activator inhibitor type 1 (PAI-1) were assessed in blood sample from eight subjects who were bitten by *C. durissus*, before starting antivenom therapy. Plasma samples from twelve healthy subjects were used as controls. **Results:** Plasma levels of TM, PDF and PAI-1 in patients samples were significantly higher than the values measured in healthy subjects ($p<0.05$). Plasma level of aPC, FVa and plasminogen were significantly lower in patients when compared with the control group ($p<0.05$). There was no significant difference in the FVIIIa plasma levels between patients and controls. It was a significant negative correlation between TM e aPC ($r=-0.41$, $p=0.02$). The finding of elevated plasma TM levels in patients compared to controls suggests the existence of endothelial injury in this accident, viewed as key in triggering the mechanism of platelet activation and coagulation. The natural anticoagulation system, mediated by TM, was probably affected by endothelial injury judging by the inverse correlation between levels of TM and aPC. The finding of reduced levels of FVa in the patients compared to controls suggests consumption of this factor in the *C. durissus* accident. **Conclusions:** These data support the hypothesis of exacerbation of the fibrinolytic system activation in the *C. durissus* accident, besides to endothelial injury. Supported by CNPq and FAPEMIG-Brazil.

A1039

ENDOTHELIAL PROGENITOR CELLS FROM THROMBOSIS PATIENTS HAD NUMERICAL AND FUNCTIONAL ABNORMALITIES. Hernández-López R*, Chávez-González A**, Hernández-Contreras A*, Cébulo-Vázquez A***, Majluf-Cruz A*, Alvarado-Moreno A*. *UIMTHA Hospital HGR No. 1 Dr. Carlos Macgregor. IMSS, México DF, México; **UIMEO Hospital de Oncología. CMN Siglo XXI. IMSS, México DF, México; ***Instituto Nacional de Perinatología. SSA, México DF, México.

Introduction: Endothelial colony forming cells (ECFC) represent a rare population in the mononuclear cells of peripheral blood (PB-MNC) from clinically healthy subjects. However, in the literature has shown that the number of cells (EPC) endothelial progenitor increases under vascular damage, generating a dysfunctional state of endothelial cells. In the case of patients with thrombosis have not been studies focused on the generation of the ECFC. **Objective:** In the present work we analyzed numerical and functional properties from ECFC in subjects with a history of thrombosis. **Material and Methods:** Mononuclear cells were isolated from the peripheral blood of 10 patients with a history of thrombosis and aged-matched-healthy volunteers 10 and growth in appropriate conditions to generating endothelial cells. Between 7 and 21 days of culture, endothelial colonies numbers were counted and phenotypically characterized. Endothelial progenitor cells function and morphology was also evaluated by the formation of tubular structures in geltrex and electronic microscopy respectively. **Results:** ECFC presents in culture and number were significantly higher in patients with thrombosis compared with healthy control subjects. Moreover this numbers were much lower in men than women. The vascular structural formation abilities in matrigel assay were significantly reduced in thrombosis patients than normal subjects. The flow cytometry analysis showed phenotype differences between normal and thrombosis cells and electronic microscopy analysis indicate lower Weibel Palade bodies in thrombosis patient cells compared with normal cells. **Conclusions:** ECFC number and the function may thus represent an accumulated endothelial cells dysfunctional status in the patient with thrombosis, without an efficient recovery of the cells of the vascular network.

A1041

ERADICATION OF INHIBITORS IN MILD HAEMOPHILIA A – CASE PRESENTATIONS. Nemes L*, Szelesy Zs*, Rona-Tas A*, Szabo T**. *National Haemophilia Center and Haemostasis Department; **Central Laboratory, State Health Center

Introduction: The development of neutralizing antibodies against factor VIII (FVIII) is a serious complication of treatment in haemophilia A (HA). The inhibitor formation in moderate-mild FVIII deficiency is a recently emphasized phenomenon with an annual incidence of 0.084 % versus 0.35 % in the severe form according to the data of the UKHCDO. 28 % of all inhibitors occurred in moderate-mild haemophilia. In mild HA, the reaction kinetics of the inhibitor and the presenting bleeding symptoms are different from what is seen in the severe form with alloantibody. Frequent occurrence of the inhibitor was noted after major bleedings, operations or trauma, following intense FVIII replacement therapy. The inhibitor is usually recognized by the altered bleeding pattern, reduced recovery, half-life and basal FVIII:C. A Dutch retrospective cohort study demonstrated the higher risk of inhibitor formation in patients with a cluster of missense mutations on the A2 domain and at the junction region

between C1 and C2 (e.g. Arg2150His, Trp2229Cys) domains. Immune Tolerance Induction (ITI) is currently the most effective therapeutic modality for the long-term treatment of patients with congenital HA complicated by inhibitor formation against FVIII but less data exist on ITI treatment in mild HA. **Objective:** To study the feasibility of ITI in mild HA. **Material and Methods:** The age of our first mild HA patient was 50 at the development of the inhibitor, with a basal FVIII:C of 10 %. His inhibitor has been developed after a heavy FVIII replacement period because of intracranial bleeding. The peak- and pre-treatment inhibitor levels were 35,0 BU/ml and 25,0 BU/ml respectively. The ITI regimen we used consisted of once a day administration of 150 IU/ bwkg of FVIII/vWF. The second mild HA patient was born in 1986. He developed the inhibitor following a series of dental surgical procedures under the cover of HP-PD FVIII. 3 month later extensive left femoral haematoma occurred and VIII:C decreased to 0.9 %, the inhibitor titer was 3.2 BU. Acute treatment with rFVIIa (NovoSeven) has been successful and the same ITI protocol resulted in complete remission after 6 months. **Results:** The ITI therapy with the administration of a FVIII/vWF product was successful in the presented cases after an ITI duration of 6,0 and 8,0 months respectively. **Conclusions:** According to the current data of the literature and our own experience, ITI can be safely and effectively used in the mild/moderate HA + inhibitor population.

A1042

GENETIC VARIATIONS IN FIBRINOGEN IN THE CZECH REPUBLIC. Kotlin R, Zichova K, Suttner J, Dyr JE. Institute of Hematology and Blood Transfusion, Praha, Czech Republic.

Introduction: Fibrinogen, a 340 kDa glycoprotein, consists of three different pairs of polypeptide chains (A α , B β , and γ) each encoded by a distinct gene (FGA, FGB, and FGG), clustered on chromosome 4q32.1. The molecule is post-translationally modified – phosphorylated, glycosylated and sulphated. Fibrinogen is synthesized by hepatocytes in the rough endoplasmic reticulum and is secreted to the circulation, where it plays a crucial role in the hemostasis, angiogenesis, platelet aggregation, cell migration and inflammation. Inherited defects in fibrinogen are very rare and may cause life threatening complications like thromboses or serious bleeding. Dysfibrinogenemia is a disease characterized by inherited abnormality in the fibrinogen molecule, resulting in functional defects; hypofibrinogenemia is characterized by low plasma fibrinogen level. **Objective:** We have investigated 62 unrelated families from the Czech Republic with abnormal coagulation test results suspected with congenital (hypo)dysfibrinogenemia. **Material and Methods:** Purified genomic DNA was amplified by PCR using specific primers and dideoxysequencing was performed. Functional examinations were carried out using fibrin polymerization, fibrinolysis, and measurement of kinetics of fibrinopeptide release. **Results:** Congenital defects in one of three genes coding fibrinogen

chains have been found in 20 families in the Czech Republic up to January 2012. A mutation in FGA gene (coding fibrinogen Aalpha chain) has been found in 11 unrelated families. The most widespread mutation in Aalpha chain was a single amino acid substitution of Aalpha 16 Arg to Cys or His. The clinical symptoms varied from asymptomatic to serious bleeding. A mutation in FGB gene (coding fibrinogen Bbeta chain) has been found in 4 unrelated families. The mutations mostly manifested as hypofibrinogenemia with a low fibrinogen level. A mutation in FGG gene (coding fibrinogen gamma chain) has been found in 5 unrelated families. Four mutations were in the C-terminal gamma nodule and one mutation in N-terminal central region of the molecule. Two cases (gamma 363Tyr/Asn and Aalpha 106Asn/Asp) were associated with thromboses. **Conclusions:** Molecular defects in fibrinogen may cause serious life-threatening conditions like bleeding or thrombosis. We found 20 unrelated families with inherited defect in fibrinogen molecule responsible for either dysfibrinogenemia or hypofibrinogenemia. This work was supported by a grant of the Grant Agency of The Czech Republic nr. P205/12/G118.

A1054

OUTCOMES ON A 5-YEAR HOME-DELIVERY PROGRAM FOR HEMOPHILIA PATIENTS IN MEXICO. Berges A*, Garcia-Chavez J**, Epstein J***, Aguilar L***, Arvizu J⁵. *Hospital General CMNR IMSS, México City, México; **Hospital de Especialidades CMNR IMSS, México City, México; ***Baxter Healthcare, Westlake Village, CA; ****Baxter Mexico, Mexico City, Mexico; ⁵Baxter Healthcare, Ft Lauderdale, FL, USA.

Introduction: The majority of hemophilia patients in Mexico receive Factor VIII (FVIII) or Factor IX (FIX) treatment on-demand when a bleed occurs. Factor home delivery programs allow patients to receive and infuse FVIII/FIX in optimal conditions at home rather than travel to the hospital to receive treatment. Home treatment also allows for prophylaxis treatment. **Objective:** The objective of this study was to evaluate the benefits of a home delivery program compared to a system where patients obtained their Factor treatment at the hospital. **Material and Methods:** Clinical and economic data was prospectively collected between 2004 and 2008 for severe hemophilia A and B patients who participated in the IMSS home-delivery program in Mexico. Data for patients no longer in the IMSS home-delivery program was retrospectively collected for 2008 and 2009 from one hemophilia treatment center to act as a control. Treatment data and outcomes were captured and descriptive analysis was performed on key variables. It must be noted that the control sample was limited in that these patients were able to bring Factor treatment obtained at the hospital to their home for future use. **Results:** Data was collected for 327 patients in the home-delivery program and 60 in the control group. These patients experienced 20,686 and 3,097 bleeding events, respectively. The mean patient age (range) was 20 (2 to 68) and 20 (4 to 64) years old in the home-delivery and

control group, respectively. 55.2% and 53.3% of all bleeds were treated within 1 hour after the patient recognized a bleed had occurred, respectively. 98.7% of bleeds were able to be adequately treated at home in the home delivery program compared to 70.3% in the control group ($p < 0.0001$). The median cost of Factor utilized per bleeding episode treated at home and at the hospital/ER for children was \$1670 and \$2505, respectively (\$2505 and \$3340 for adults). **Conclusions:** This data demonstrates the value of a home delivery program in Mexico. Patients enrolled in the program were able to treat almost all of their bleeds at home. Less Factor product was used for bleeds treated at home compared to the hospital. Future research should compare the outcomes and costs of home delivery to the alternative of requiring patients to receive treatment at the hospital.

A1055

ACCESS TO HEMOPHILIA TREATMENT AND COMPREHENSIVE CARE IN MEXICO: A COMPARISON OF THE MINISTRY OF HEALTH INSTITUTIONS AND INSTITUTO MEXICANO DEL SEGURO SOCIAL INSURANCE SCHEMES. Berges A*, Epstein J**, Xiong Y**, Ito D**, Aguilar L***, Arvizu J****. *Hospital General CMNR IMSS, México City, México; **Baxter Healthcare, Westlake Village CA, USA; ***Baxter México, México; ****Baxter Healthcare, Ft. Lauderdale, Fla, USA.

Introduction: It has been well documented that a prophylaxis treatment regimen of Factor VIII (FVIII) and access to the appropriate level of care will result in improved outcomes for patients with severe hemophilia A. In Mexico, hemophilia patients who may be unemployed or are economically disadvantaged may be covered by the Ministry of Health (MoH), a government institution that provides treatment for those who otherwise have very limited access to care. It remains uncertain however, whether the level of care under MoH is comparable to the Instituto Mexicano del Seguro Social insurance scheme (IMSS), which provides coverage to approximately 50 million employed individuals in Mexico. **Objective:** The purpose of this study was to assess whether hemophilia A patients covered under MoH received comparable levels of treatment to those with IMSS. **Material and Methods:** This study was a cross-sectional, self-administered survey of patients aged 18 or older, or the parent/caregiver of patients aged 2-17, who were diagnosed with severe hemophilia A. Participants were recruited by local hemophilia patient associations in partnership with the Mexican Hemophilia Federation in the following cities: Veracruz, Estado de Mexico, Ciudad de Mexico, Michoacán, Jalisco and Yucatan. Patients meeting inclusion criteria were consented to participate and completed the questionnaire during May and June 2011. Patients were asked a range of questions about their hemophilia treatment and outcomes, including treatment regimen, bleeds rates and access to comprehensive care. **Results:** 200 patients (100 adults, 100 pediatric) participated in the study, with 125

(63%) receiving IMSS, 37 (19%) with MoH, and the remaining 18% covered by other Government institutions. IMSS patients were more likely to be on prophylaxis treatment (30%) compared to MoH (11%). More IMSS patients received convenient home delivery of FVIII compared to MoH patients (33% vs 0%). Furthermore, IMSS patients were more likely to have FVIII at home (70% vs 16% MoH), and were able efficiently treat their bleeds during the past year (78% vs 30%) compared to MoH patients. Over half of MoH patients reported having difficulty accessing their hematologist in the past year (57%) compared to IMSS patients (24%). **Conclusions:** The results suggest that hemophilia A patients affiliated with IMSS have better access to hemophilia treatment and care compared to MoH patients. Ensuring that patients have access to comprehensive hemophilia care and increasing the proportion of patients on a prophylaxis treatment regimen will facilitate improvement in outcomes for those patients with MOH coverage.

A1056

COMPARISON OF HEMOPHILIA A TREATMENT PATTERNS AND OUTCOMES BETWEEN ARGENTINA, CHILE, COLOMBIA, AND MEXICO. Berges A*, Perez-Bianco R**, Krishnan S***, Ito D***, Aguilar L****, Arvizu J⁵. *Hospital General CMNR IMSS, México City, México; **Instituto de Investigaciones Hematológicas-Academia Nacional de Medicina, Buenos Aires, Argentina; ***Baxter Healthcare, Westlake Village CA, USA; ****Baxter México, México; ⁵Baxter Healthcare, Ft. Lauderdale, Fla, USA.

Introduction: There are large gaps in global hemophilia treatment standards within Latin America. The 2009 World Federation of Hemophilia (WFH) Survey reported the FVIII IU per capita for Argentina, Chile, Colombia and Mexico as 2.4, 2.2, 1.4, and < 1.0, respectively. **Objective:** The objective of this analysis was to understand differences in treatment patterns and outcomes among hemophilia A patients within these 4 countries to identify potential disparities in care. **Material and Methods:** We used a multinational, cross-sectional design to administer an IRB-approved paper-based survey to consenting hemophilia A patients = 18 or caregivers for patients age 2-17. Patients were recruited through local hemophilia associations in Argentina, Chile, Colombia, and Mexico. Data collection occurred in two phases from October to November 2009 (Argentina) to June-August 2011 (Chile, Colombia, Mexico). Inhibitor patients were excluded. Patients were asked questions about their hemophilia treatment and outcomes, including treatment regimen, bleed rates, target joints, and access to care. **Results:** 460 patients were surveyed overall with 268 adults and 192 children across the four countries. The mean age for adults and children was 32.5 years (Range 18-68) and 9.3 years (Range 2-17), respectively. The majority of patients were on recombinant FVIII (66%), 31% on plasma-derived FVIII, 2% on cryoprecipitate and 0% on fresh frozen plasma. In the past, patients reported receiving cryoprecipitate

(59%) and fresh frozen plasma (35%). Overall, 45% of patients in the study were on either primary or secondary prophylaxis. Patients in Colombia reported the greatest percentage (93.2%) on primary or secondary prophylaxis versus Chile (48%), Argentina (30%), and Mexico (23%) ($p < 0.0001$). Of the patients receiving prophylaxis therapy, 33% were dosing at the recommended dose and frequency according to MASAC guidelines (25-40 IU/kg, 3 times weekly). On a country-level, 53% of Argentina and 57% Colombia patients received the recommended FVIII therapy dose versus Mexico and Chile where only 7% and 4% received their recommended dose ($p < 0.0001$). The annual median bleed rates (total bleeds) differed significantly between primary prophylaxis, secondary prophylaxis, and on-demand (5, 12, and 25 bleeds respectively ($p < .0001$)) demonstrating the benefits of prophylaxis overall for these patients. Similar trends for joint bleed rates and target joints were also observed. **Conclusions:** The results suggest there is a large disparity among patients in different Latin America countries receiving prophylaxis versus on-demand treatment. Primary prophylaxis in these respective patients has translated to superior outcomes such as reduced total and joint bleed rates, and reduced target joints.

A1062

PRIMARY THROMBOPHILIA IN MÉXICO IX: THE GLYCOPROTEIN IIIA PLA1/A2 POLYMORPHISM MAY RESULT IN THE STICKY PLATELET SYNDROME. Ruiz-Arguelles GJ*****, Garcés-Eisele J*****, Camacho-Alarcón C**, Moncada-González B*****, Valdes-Tapia P**, Leon-Montes N**, Ruiz-Delgado GJ*****. *Centro de Hematología y Medicina Interna de Puebla; **Laboratorios Clínicos de Puebla; ***Universidad Popular Autónoma del Estado de Puebla; ****Universidad Autónoma de San Luis Potosí.

Introduction: The sticky platelet syndrome phenotype (SPS) is a rather common cause of arterial and venous thrombosis defined by platelet hyperaggregability with adenosin-diphosphate and/or epinephrine. The platelet abnormality seems to be congenital and the precise nature of the defect is at present not known; it is supposed that glycoprotein receptors on the platelet surface membrane may be involved, its abnormality leading into platelet hyperfunction. No molecular substrate to explain the platelet hyperaggregability has been found in the SPS phenotype, this being the reason why only few research groups have accepted this entity as a true thrombophilic condition. **Objective:** To analyze a possible association between the SPS phenotype and the platelet GP IIIa PLA1/A2 (HPA-1a/b) gene polymorphism in Mexican mestizo patients with a clinical marker of thrombophilia. **Material and Methods:** Along an 18-month period, all consecutive Mexican Mestizo patients referred to the Centro de Hematología y Medicina Interna de Puebla by physicians from different parts of the country were prospectively accrued in the study if they had clinical markers associated with a primary hypercoagulable state. The SPS phenotype was assessed by the aggregometry method

of Mammen, whereas a tetra-primer ARMS polymerase chain reaction based analysis was used for the detection of the PLA1 and PLA2 alleles. **Results:** One hundred individuals with SPS and 127 normal donors were studied; in 11 of the donors and 18 of the SPS patients the A2 allele of the gp IIb/IIIa was found yielding a weak association (odds ratio 2.31, 95% CI 1.03 – 5.16). **Conclusions:** In Mexican mestizo patients, the platelet GP IIIa PLA1/A2 gene polymorphism may lead into the SPS phenotype. Other variables may also account for the phenotype.

A1071

GENETIC FACTORS FOR THROMBOPHILIA IN PATIENTS WITH A HISTORY OF THROMBOSIS IN THE UMAE PUEBLA. Pérez-Lozano U, Ruiz-Ovalle JL, Solis-Poblano JC, Limón-Flores JA, Lobato-Tolama R. UMAE IMSS Puebla.

Introduction: The most frequent genetic prothrombotic defect in the world population is resistance to activated C protein (RCP). The V Leiden mutation in western countries it represents up to 10 times more cases than other genetic factors of thrombophilia such as deficiency of protein C, S and AT-III, among the most frequent ones. Hyperthrombinemia associated to the G20210A mutation of prothrombin is the first cause of thrombophilia in Spain. The incidence of MTHFR C 677-T in Mexico is one of the highest reported worldwide. **Objective:** Describe the epidemiology of primary thrombophilia in Mexico. **Material and Methods:** 41 patients were studied with a history of thrombosis documented through tomography, Doppler ultrasound or angiography, at least 4 months before; patients who were pregnant or in puerperium, with collagen disease, who underwent surgery and known cancers were excluded. The mean age was 34 years (range was 10 – 55 years); 17 women (41.5%); 24 men (58.5%); 41.5% (17) had relatives with thrombosis. **Results:** 61% (25) of patients had a normal BMI, 24.4% (10) were obese and 14.6% (86) were overweight. 23 patients (56.1%) presented MTHFR C677-T, 31.7% (13) of these were heterozygotes and 24.4% (10) homozygotes, and in all cases homocysteine was normal when evaluated. All patients evaluated for the G20210A mutation were negative; 22% (9) presented RCP, only 7.3% (3) had the V Leiden mutation; 19.5% (8) had C protein deficiency and 2.4% (1) had deficiency of protein S; 7.3% (3) had antithrombin III deficiency. 2 cases were factor XII deficient, and in 3 patients there was no disorder found and 4 patients had multiple congenital disorders evaluated. 29.3% (12) of the series presented cerebral thrombosis; 17.1% (7) had pulmonary thromboembolism (PTE); 36.6% (15) deep venous thrombosis; 7.3% (3) arterial thromboembolism, 2.4% (1) acute myocardial infarction. 80% of patients with stroke presented the MTHFR C667-T ($p=0.004$), 25% of patients DVT ($p=.232$) and 33% of patients with PTE presented the mutation. **Conclusions:** The MTHFR C677-T in this small case series has a very high frequency in general and in patients with cerebral thrombosis the cause is not explained. Hyperthrombinemia did not explain any case of hypercoagulability and the RCP was explained in

a very low rate from a genetic cause, therefore we observed a significant difference with Caucasian reports

A1090

THE BEHAVIOUR OF LABORATORY CLINICAL FINDINGS IN PATIENTS WITH SEVERE SEPSIS OR SEPTIC SHOCK MANAGED WITH ACTIVATED C PROTEIN (ACPrh). DATA WITH PROGNOSTIC VALUE. Mariscal II, Lim-Baga GJ. Internal Medicine, Hospital Gómez Farias ISSSTE, Guadalajara, México

Introduction: ACPrh has been used for the management of Severe sepsis (SSp) and septic shock (SSh) with contradictory results; in November 2011 the FDA recommended its withdrawal from the market. Before that, this hospital carried out a prospective study with 28 patients with SSp or SSh treated according to international guidelines, 10 received ACPrh as well. **Objectives:** Clinical and laboratory variables were determined upon admission, at 96 h and at discharge to know the usefulness of ACPrh, its behavior and prognostic value. **Results:** 18 men were included; mortality was 70% in ACP and 33% in the control group (PNS) and 43% in SSp and 57% in SSh. In patients with ACPrh, TP* and INR* were prolonged and Fg* descended. In controls the following improved: temperature*, FC*, white blood cells*, Hb**, Fg***, Chlorine* and bilirubins**. At 96 h, Hb was lower*** in ACPrh and at discharge, the TTP* was prolonged. Of the variables upon admission that predict mortality, the following were significant: ** sodium*, chlorine***, -Fg***, TTP** and SOFA* (* $P<0.05$, ** $P<0.01$, *** $P<0.001$). We observed in patients an inflammatory response expressed by white blood cells, bandemia and hyperfibrinogenemia present upon admission, variables that improved throughout the disease in the controls. Patients with ACPrh had a progressive and significant decrease of Hb that could have contributed to organ damage and because of unnoticeable bleeding, hemolysis or other; they also had a continuous decrease of Fg and platelets with increase of INR, which was absent in the control group. Among mortality predictors there were hemostasis and renal function disorders that were present from admission, there was a decrease of TAM and an increase of bilirubins at 96 hours and a decrease of Hb and hematocrit at discharge. ACPrh did not improve prognosis, nevertheless, there was an increase (PNS) in mortality that matches the PROWESS SHOCK trial. We expect prognosis to improve when the immune and inflammatory responses, the vascular and organ damage, as well as hemostasis and the triggering pathology are corrected. Thrombomodulin and heparanase inhibitors, PAR and TLR 1, among others, appear to be promissory. **Conclusions:** There were anomalies shown in hemostasis, renal and liver function and inflammatory response data at different times, where hemostasis and renal anomalies were the first to appear, then cardiovascular and liver, and at a later stage Hb and hematocrit. The inflammation variables improved in survivors. In patients with ACP, the prognosis did not improve; they even had a higher mortality than the control group.

A1100

DETERMINATION OF HOMOCYSTEINE AND ANTI-THROMBOTIC PROTEINS AND THEIR ASSOCIATION WITH THE COMMON DISEASES IN OLDER MEXICAN MESTIZOS.

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Introduction: Homocysteine (Hcy) is an amino acid originated from the metabolism of methionine, which in high concentrations: hyperhomocysteinemia (HHC)($> 15 \text{ mol / L}$) is considered atherogenic factor in several diseases. The HHC causes damage and dysfunction endothelial by generating reactive oxygen species, which promote thrombosis by increasing thrombin generation, decreases nitric oxide production and inhibit the activation of antithrombotic protein: protein C (PC), protein S (PS) and antithrombin (AT). **Objectives:** Determinate the levels of Hcy, PC, PS and AT in older adults, Hcy levels correlate with these proteins and associated diseases. **Material and Methods:** We studied 115 participants aged 50 to 98 years old who attended the Geriatric Rehabilitation Service of the Instituto Nacional de Rehabilitacion and Laboratorio de Investigacion de Hematopatologia del departamento de Morfologia de la Escuela Nacional de Ciencias Biologicas del Instituto Politecnico Nacional. They answered a questionnaire and those who had diseases were considered as patients and who did not present diseases, were considered as controls. Both groups (95 patients and 20 controls) were determined: the concentration of Hcy in blood by automated immunoassay and activity of PC, PS and AT by chromogenic and coagulometric methods by automated equipment and ILACL Elite Pro version Diagnostics Participants divided into age groups 50-59, 60-69, 70-79 and > 80 years. **Results:** We obtained the following results: 24% increased Hcy which was correlated with age. There was variation between gender and age groups of the activity of PC and only age group to the PS. There was correlation between the activity of PC-PS, PS-AT and PS-Hcy; association between hypertension with the concentration of Hcy and AT, chronic renal failure with the concentration of Hcy and PC, as well as between cerebrovascular disease and dyslipidemia with the activity of PC. **Conclusions:** The high concentration of Hcy in peripheral blood affects the PS in adults 50 years or more, not to the PC and AT. He obtained significant association between hypertension with Hcy concentration and activity of AT. The chronic renal failure was associated with Hcy concentration and activity of the PC. Also the cerebrovascular disease and dyslipidemia with the activity of the PC.

A1101

DETERMINATION OF THE COAGULATION FACTORS AND PROTEIN C, S, AT AND PLASMINOGEN IN OLDER MEXI-

CAN MESTIZOS. Gonzalez-Espinosa L*, Rodríguez-Villa A*, Hernández-Zamora E**, Zavala-Hernández C**, Rosales-Cruz E*, Martínez-Murillo C***, Reyes-Maldonado E*. *Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, México DF, México; **Instituto Nacional de Rehabilitación, México DF, México; ***Hospital General de México OD, México DF, México.

Introduction: The haemostasis consist in a set of reactions which going to be activate when a vascular injury starts, which includes, endothelial surface, platelet, coagulation factors (CF), coagulations inhibitors (CI) and fibrinolysis system. As the first time in history, in few years the population over 65 years will be superior than population under 5 years. In México, the elderly population is expected to increase 232% in 2040. This population does not have any normal range (NR) of the coagulation factors and natural anticoagulant proteins. **Objective:** Determinate the levels of CF, Protein C (PC), Protein S (PS), antithrombin (AT) and plasminogen (Plg) in an over 55 years population. **Material and Methods:** The investigation includes 200 patients of geriatric rehabilitation service. We performed this transversal study with analysis of variance procedures to analyze changes between different ages and sex groups. Blood samples was anticoagulated with trisodium citrate, we separate plasma and determinate CF by coagulometer assays. **Results:** We have 120 patients, which were classified by gender and age groups. The age range varied from 50 to 98 years with a mean of 70. General characteristics of these participants, 17% have diabetes, 46% hypertension, 16% osteoporosis, 7% cardiovascular disease (CVD) and 60% venous insufficiency, which were correlated to determine if these diseases modify the values of the CF. The plasma concentration of fibrinogen increases with age ($p = 0.0014$). FVIII and FXII activity increases with age without significant difference ($p > 0.05$) and PS activity increases too with age and decreases in the over 85 years group ($p = 0.0024$). FII, FV, FX, PC and Plg activity decreases with increasing age ($p < 0.05$). While the FVII, FIX, FXI and AT percent activity remain at a stable level. with age. With respect to sex, those which are statistically significant differences FVII, FXI and PC ($p < 0.05$). CF were also associated with various conditions such as hypertension, osteoporosis, CVD, and venous insufficiency. **Conclusions:** It is necessary investigate our population because the levels of CF and CI can be modifying by: illness, race, environmental factors and others.

A1102

DETERMINATION OF MULTIMERS VON WILLEBRAN FACTOR AS A CONFIRMATORY TEST DIAGNOSTIC VON WILLEBRAND DISEASE. Ochoa-Rico A*, Hernández-Zamora E**, Zavala-Hernández C**, González-Espinosa L*, Rosales-Cruz E*, Martínez-Murillo C***, Quintana-González S****, Reyes-Maldonado E*. *Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, México DF, México; **Instituto

Nacional de Rehabilitación, México DF, México; ***Hospital General de México OD. México DF, México, ****Instituto Mexicano del Seguro Social, Mexico DF, Mexico.

Introduction: Von Willebrand disease (VWD) is the mucocutaneous haemorrhagic disease, its prevalence is estimated at ~1%. It is an underdiagnosed disease because it is poorly understood, its diagnosis is complicated by individual variability and laboratory tests required specialized hemostasis. **Objectives:** Determinate the pattern of multimers of von Willebrand factor (vWF) and other special tests in patients with VWD and classify possible. **Material and Methods:** We studied 45 patients with suspected VWD and 45 healthy donors were carried out basic tests: bleeding time (Ivy method), platelet count (Celdyn automated method), analysis of blood smear (Wright stain). The activated partial thromboplastin time (aPTT), prothrombin time (PT) and the quantification of PT derived fibrinogen by using a team approach coagulometric ACL ELITE PRO (IL diagnostic). Special tests: clotting factor VIII activity coagulometrico (FVIII: C), quantification of vWF antigen (vWF: Ag), ristocetin cofactor activity of vWF (vWF: RCo) and blood group (ABO). Classification tests: ristocetin-induced platelet agglutination (RIPA) and analysis by immunoelectrophoresis of vWF multimers. **Results:** Of the 45 patients analyzed, twenty were classified as type 1 VWD, a type 2B patient (absence of large multimers),

five patients and type 3 (complete absence of vWF), a patient with decreased platelet aggregation with epinephrine, three patients likely type 2N and probable type 2M fifteen patients. However it would be necessary to perform other tests such as vWF bound to collagen (VWF:CB). **Conclusions:** They consider the analysis of multimers as an important test for the classification of VWD types 1, 3 and subtypes 2A and 2B, but not for VWD types 2N and 2M, so further tests are needed and molecular biology.

A1104

A CASE WITH PSEUDOTHROMBOCYTOPENIA WHO SHOWS DIFFERENT FEATURES: TEMPERATURE-INDEPENDENT PSEUDOTHROMBOCYTOPENIA COMPLETE WITH CITRATE AND INCOMPLETE WITH ETHYLENE-DIAMINETETRAACETIC ACID AND HEPARIN. Beyan C*, Kaptan K*, Eriki A**. *Gulhane Military Medical Academy, Department of Hematology, Etlik, Ankara, Turkey; **GATA Haydarpasa Training Hospital, Department of Hematology, Uskudar, Istanbul, Turkey.

Introduction: Pseudothrombocytopenia (PTP) is a phenomenon due to agglutination of platelets in the complete blood count tube anticoagulated with ethylenediaminetetraacetic acid (EDTA) leads to falsely low platelet counts. **Objective:** Herein, a case with PTP who shows different features is presented. **Material and methods:** A case with 62 year-old woman admitted to our Hematology department referred from Medical Oncology department because evaluation of thrombocytopenia. She had colon cancer and colectomy operation was performed two and a half years ago with no need for chemotherapy or radiotherapy. At the time of operation, platelet counts were normal. She had variations in platelet counts during the last year. Platelet counts were 19 x10⁹/l with EDTA and 4 x10⁹/l with citrate from fresh blood samples. Peripheral blood smear revealed 61% neutrophils, 27% lymphocytes, 9% monocytes, 3% eosinophils, and erythrocytes were normochromic and normocytic. Platelet clumps were present with mean 9.3 platelets in every x1000 magnification. Because of false manipulation of laboratory personnel, the same samples were recounted after three hours platelet values were 50 x10⁹/l in EDTA and 10 x10⁹/l in citrate tubes, respectively. **Results:** After these contradictory results, a profile was made by using different anticoagulants. According to 24-hour platelet count profile with different anticoagulants, our case had different features from classical EDTA-dependent PTP: PTP was observed

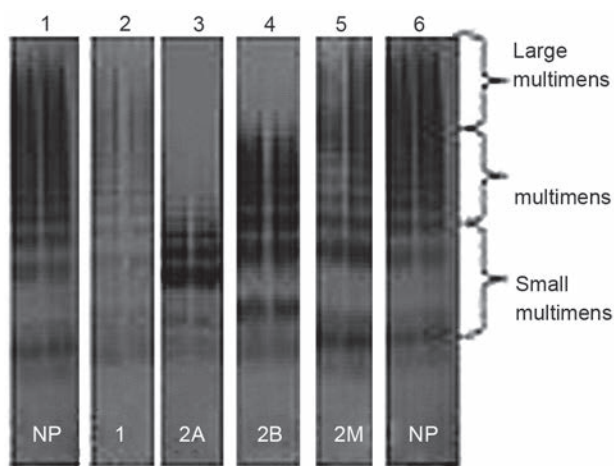


Figure 1. Screenshot from the analysis of vWF multimers. In normal plasma line 1 and 6 (NP), line 2: type 1 VWD, line 3: type 2A VWD, line 4: type 2B VWD and line 5: type 2M.

Group	Number of patients	Diagnosis	Characteristics
1	15	Differentiated from other coagulopathies	Normal electrophoretic pattern, with bleeding symptoms.
2	20	EvW Type 1	Qualitative decline of all multimers (high, intermediate and low MW)
3	1	EvW Type 2B	Decrease of high MW multimers.
4	5	EvW Type 3	Absence of multimers
5	3	EvW Probably type 2N	Normal electrophoretic pattern, with bleeding symptoms.
6	1	Probable alteration of platelet receptor level	Normal electrophoretic pattern, with bleeding symptoms

in the fresh blood with EDTA, citrate and heparin independently from temperature changes (warm antibodies, not cold agglutinin). In the presence of citrate, antibodies bind platelets more effectively (complete antibody); this effect was incomplete in the presence of EDTA and heparin (incomplete antibody). Therefore, the variations of platelet counts in different hours were observed. Sodium fluoride which is a glycolysis inhibitor explains the false results in tubes with only EDTA and true results in tubes with EDTA combined with sodium fluoride. Sodium fluoride causes inhibition of glucose utilization by platelets in test tube. This leads inhibition of morphological changes of platelets and prevents the formation of PTP. **Conclusions:** As a conclusion, our case which has a temperature-independent PTP complete with citrate and incomplete with EDTA and heparin shows different features from classical EDTA-dependent PTP. True platelet count could be made with addition of sodium fluoride into EDTA.

A1123

ANALYSIS OF VON WILLEBRAND FACTOR BY VERTICAL SDS-AGAROSE GEL ELECTROPHORESIS AND SEMI-DRY ELECTROPHORETIC TRANSFER CONDITIONS. López-Castañeda SE*, Godínez-Hernández D**, Gutiérrez-Castellanos S*, Areán-Martínez CA***, Viveros-Sandoval ME*. *Laboratorio de hemostasia y Biología Vascular, Facultad de Medicina, UMSNH; **Instituto de investigaciones Químico Biológicas, UMSNH; ***Hospital General Dr. Miguel Silva, Morelia Michoacán México.

Introduction: vWF has been proposed as a biomarker, a prognostic indicator, and a mediator of atherosclerotic cardiovascular disease. Larger vWF multimers are more effective in hemostasis. Electrophoretic analysis of vWF multimers allows the study of multimeric structure of vWF, and identification of High Molecular Weight Multimers. Existing methodologies are complicated reason why a reproducible and easy method that allows routine analysis of vWF is needed. SDS-PAGE by vertical agarose gel electrophoresis could be the election method to study the multimeric structure of von Willebrand Factor, focusing on high plasma vWF levels and presence of High Molecular Weight Multimers (HMWM) present in thrombotic diseases. **Objectives:** To standardize a novel method for multimer analysis of vWF by vertical SDS-agarose gel electrophoresis, followed by Western blotting using semi-dry electrophoretic transfer conditions. **Material and methods:** Multimeric analysis of vWF by vertical (sodium dodecyl sulfate) agarose gel electrophoresis, followed by Western blotting and semi-dry electrophoretic transfer conditions and detection with Rabbit Immunoglobulin to Human vWF. We used a discontinuous gel system with agarose 1% for the resolving gel and agarose 1.3% for the stacking gel. The present study was performed in individuals with Atrial Fibrillation (AF), Acute Coronary Syndrome (ACS) and Thrombotic Thrombocytopenic Purpura (TTP) focusing on high plasma levels of VWF and presence of HMWM. **Results:** The results of multimeric studies showed differences between the multimeric pattern of normal

controls and patients with ACS, AF and TTP. These patients had a greater concentration of high molecular weight multimers than normal controls. **Conclusions:** Vertical agarose gel electrophoresis and semi-dry transfer conditions offer excellent results that allow identification of vWF HMWM. The study of vWF multimeric structure in pro-thrombotic pathologies could be useful to identify patients in risk

A1126

HIGH ON-TREATMENT PLATELET REACTIVITY BY TWO DIFFERENT PLATELET FUNCTION TESTS: FLOW CITOMETRY AND VERIFY-NOW. García-Larragoiti NI*, Taboada-Cortina A*, Areán-Martínez CA**, Gutiérrez-Castellanos S*, Viveros ME*. *Laboratorio de Hemostasia y Biología Vascular, División de Posgrado, Facultad de Ciencias Médicas y Biológicas Dr. Ignacio Chávez, UMSNH; **Unidad de Hemodinamia, Hospital General Dr. Miguel Silva, Morelia, México.

Introduction: Several studies have shown inter-individual variability in platelet response to antiplatelet treatment and might be associated with increased risk of recurrent atherothrombotic events. High on-treatment platelet reactivity (HPR) to ADP measured by multiple methods has been linked to adverse post-PCI clinical event occurrence, but the correlation between these methods is still a matter of debate. In this study we compared two different methods to assess platelet reactivity: flow cytometry VASP phosphorylation assay and VerifyNow System, a “point of care” instrument, to measure platelet function and evaluate response to Clopidogrel therapy. **Objectives:** To evaluate platelet reactivity in response to antiplatelet treatment by two different methods. **Material and Methods:** P2Y12 reaction units (PRU) were determined by the VerifyNow System (Accumetrics), Platelet reactivity Index (PRI) was determined by the analysis of VASP (Platelet VASP/P2Y12 Biocytex, France) in flow cytometer (Beckman Coulter Epics Altra) in stable CAD patients on dual antiplatelet therapy. Correlations between assays were determined by Pearson statistics, and ROC curve analysis was used to compare HPR defined by different assays. Low response to Clopidogrel was defined as a PRI > 50% after 600 mg loading dose (Bonello L, 2010) and a PRU > 208 (Price et al, 2011). Two samples for analysis: six to eight hours after administration of loading dose before PCI and 24 hours after PCI. **Results:** PRU showed a significant correlation with PRI. Risk stratification (HPR vs. non-HPR) based on Consensus PRU criteria (>208) correlated well with PRI criteria (> 50%). **Conclusions:** PRU showed good correlation with PRI and could be useful in identifying patients with inadequate response to antiplatelet treatment. Studies involving larger number of patients are needed.

A1127

THROMBOCYTOPATHY DUE TO DEFICIENCY OF GRANULES STORES: TWO CASE REPORT. Moreno-González AM*, Hurtado-Obispo B**, Castillo-Martínez ID*. *Hospital Infantil

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Introduction: The thrombocytopathies are a heterogeneous group of inherited disorders characterized by variable reduction in the number and content of granules in megakaryocytes and platelets. The gray platelet syndrome (GPS) is characterized by thrombocytopenia and abnormal giant platelets with absent platelet alfa-granules. Patients with GPS have a bleeding tendency of variable bleeding severity. Characteristically the platelets of these patients are gray in the peripheral blood smear due the absence of alfa-granules and their constituents. Delta-storage pool disease (SDP) patients lack delta-granules resulting in a deficiency of ADP, ATP and serotonin, which when released, enhance platelet aggregation. The blood count provides information about platelet number and volume distribution. SPD and GPS platelets require the use of electron microscopy for reaching a definitive diagnosis.

Objective: Describe two patients with macrothrombocytopathy. **Material and methods:** Case report. **Results:** Case 1. A seventeen year old female, with no familiar bleeding history, 3 years of recurrent epistaxis requiring cauterization one time and trasvaginal bleeding 4 days a week. Due to defective aggregation with ADP is suspected a storage pool disease and electronic microscopy reports ultrastructural alteration in platelet granules shape and number. The result of mutation in 3p is pending. Case 2. A ten year old female, with cleft palate correction surgery without complications, father's mean platelet volume (MPV) elevated, with easy bruising and thrombocytopenia. The platelet count decreases to 3000, was treated with prednisone 2mg/kgd, due to lymphopenia, systemic eritematous lupus is suspected and discarded. All the immunological labs and familiar study were normal. The bone marrow reported normal. Macrothrombocytopathy is suspected because MPV 10.3. Electronic microscopy reports platelets with decreased submembranous cysterna and granules. The result of mutation in 3p is pending. **Conclusions:** Macrothrombocytopathy is a rare diagnosis that should be considered in any child with bleeding disorder. The diagnosis is suspected with thrombocytopenia and elevated mean platelet volume, and is confirmed with electronic microscopy and mutation in 3p, but this is still not available in many centers.

A1128

ALTERNATIVE ANTICOAGULANTS IN EVALUATION OF EDTA-INDUCED PSEUDOTHROMBOCYTOPENIA AT THE INCMNSZ. Saldaña-Lopez RV*, Vargas-Ruiz AG*, Rojas-Maya S*, Torres-Jimenez MD*, Leon-Moreno E*, Galindo L*, Barrales-Benitez O*, Valencia-Martínez AA*, Marquina ML**. *Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubiran, México D.F., México. **Instituto Mexicano del Seguro Social, México D.F., México.

Introduction: Pseudothrombocytopenia occurs because EDTA (etilendiamintetraacetic acid) is the anticoagulant employed in the tubes used for routine complete blood counts. Is a false

decrease in platelet count and is an in vitro phenomenon caused by EDTA-dependent agglutinins which causes time-dependent platelet clumping in presence of EDTA. Traditionally, in these cases, Heparin has been recommended as the alternative anticoagulant. **Objectives:** To compare the performance of three anticoagulants (sodium citrate, heparin and acid citrate dextrose or ACD) in the assessment of platelet counts in patients with EDTA-induced pseudothrombocytopenia. **Material and Methods:** From November 2009 to October 2011 we identified patients with EDTA-induced thrombocytopenia (low platelets in the EDTA -blood count and platelet clusters in the blood smear). We take simultaneous blood samples using tubes with EDTA, sodium citrate (3.2%), heparin and ACD. In each sample was performed an automatic blood count (using the LH750 Coulter), Wright-stained smear, leukocyte and platelet count in Neubauer chamber, at baseline, 30, 60, 120 and 180 minutes. We reviewed the clinical record of each patient. **Results:** We included 20 patients, 13 women and 7 men, aged from 22 to 72 years, none had hemorrhagic manifestations and in four there was an underlying disease (malignancy, viral infection, liver failure and nephrotic syndrome). EDTA and heparin had poor correlation between the manual platelet count and the automatic blood count, however, with sodium citrate and ACD the correlation was higher. **Conclusions:** The best anticoagulants to assess EDTA-induced pseudothrombocytopenia are sodium citrate and ACD which have the most real platelet count in automated equipment. Heparin has a poor correlation with manual counting. For blood smears, EDTA anticoagulant should remain.

A1137

GENETIC VARIABILITY OF THE GLYCOPROTEIN VI GENE IN PATIENTS WITH STROKE AND PLATELET HYPERAGGREGABILITY. Kubisz P, Staško J, Ivanková J, Škerenová M, Holý P. Jessenius Faculty of Medicine, Comenius University, University Hospital.

Objectives: The aim of the investigation was to study the genetic polymorphisms of the GP6 gene in patients with sticky platelet syndrome (SPS). **Material and Methods:** 71 patients with SPS and a history of ischemic stroke, and 77 controls without SPS were involved. SPS was diagnosed by platelet aggregometry (PACKS-4 aggregometer, Helena Laboratories) according to the method of Mammen. Seven single nucleotide polymorphisms (SNP) of the GP6 gene (rs1654410, rs1671153, rs1654419, rs11669150, rs1613662, rs12610286, rs1654431) were investigated with the use of restriction fragment length polymorphism (RFLP) analysis. **Results:** All allele and genotype frequencies were comparable between both SPS patients and control group with no statistically significant differences. The haplotype analysis showed a higher occurrence of the one major haplotype (TTGTGA, 0,228 vs. 0,174; OR 1,421; CI 0,799-2,526) and two minor haplotypes (CGATAA, 0,026 vs. 0,006; OR 4,117; CI 0,443-38,25; TTGTGG, 0,018 vs. 0,009; OR 2,107; CI 0,259-17,12) in patients with SPS. None of

haplotype differences was statistically significant. However, both the allele G of SNP rs12610286 ($p = 0,029$; OR 2,411; CI 1,134-5,123) and one major haplotype (TTGTGA; $p = 0,012$; OR 2,749; CI 1,223-6,174) were found significantly more frequent in patients with SPS type I in comparison with controls. **Conclusions:** Our results, especially higher occurrence of selected haplotypes in SPS patients, can support an idea that variability of the GP6 gene may be associated with the platelet hyperaggregability in SPS. Supported by the Project CEVYPET (Center of Excellence for Research in Personalized Therapy) and grant Vega 1/0029/11.

A1144

EVALUATION OF PLATELET FUNCTION IN PATIENTS WITH MYELODYSPLASTIC SYNDROMES AND ACUTE MYELOID LEUKEMIA. CORRELATION WITH BIOPHYSICAL PROPERTIES OF PLATELET MEMBRANE. Popov V***, Vladareanu AM****, Kovacs E**, Savopol T**, Iordache M**, Begu M****, Bumbea H****, Onisai M****, Nicolescu A****, Popa C*. *County Emergency Hospital Arges, Hematology Department; **University of Medicine and Pharmacy Carol Davilla, Bucharest; ***University Emergency Hospital Bucharest.

Introduction: Patients with myelodysplastic syndromes (MDS) and acute myeloid leukemia (AML) present severe alterations of platelet function. Platelets from these patients are dysplastic and have alterations in membrane and granular content. **Objectives:** The purpose of this study was to identify abnormalities in platelet function and a possible correlation with changes in platelet membrane fluidity and the reactive species level. **Material and Methods:** We present a prospective study on 73 cases with MDS compared with 33 cases with AML admitted in University Emergency Hospital Bucharest. Three patients with AML were investigated in early stages and in the phase of complete remission of the disease. Platelet function was investigated by platelet aggregation using as stimuli ADP, collagen, epinephrine and ristocetin. Membrane fluidity was assessed by fluorescence anisotropy measurements using TMA-DPH, the ROS level determination was performed using DCFDA method. **Results:** Platelet aggregation was altered in both groups of patients, more pronounced for AML patients. (AML vs. MDS: ADP 28.10 vs. 37.88, $p = 0.009$; collagen 40.69 vs. 58.37, $p = 0.003$; epinephrine vs. 16.48 24.16, $p = 0.14$; ristocetin 43.73 vs. 50, $p = 0.37$). No significant differences were obtained in the lag phase in the two groups of patients. The platelet aggregation response was improved in remission phase of AML for all reagents. The membrane anisotropy was increased in AML patients compared to MDS patients ($r = 0.1711$ vs. 0.1372, $p = 0.002$), this result corresponding to low fluidity of membrane. There was not obtained a statistically significant correlation between the degree of membrane anisotropy and severity platelet function. Reactive species level is slightly higher in AML patients (0.0005280 vs.

0.0004051, $p = 0.52$). This level is significantly increased in advanced phase of the disease. (LAM patients 0.0006078 vs. 0.0001413, $p = 0.003$; MDS patients 0.0005452 vs. 0.0001804, $p = 0.005$). We could not establish a correlation of the fluidity changes depending on the level of ROS. **Conclusions:** AML patients have an advanced degree of alteration of platelet function and a low fluidity of platelet membranes compared to MDS patients. Achieving a clinical remission may improve platelet function, possibly due to a more appropriate expression of platelet receptors and / or improving cellular signaling, possibly correlated with low levels of reactive species.

A1145

SPLENOMEGALY CONSEQUENCE OF EXTRAMEDULLARY HEMATOPOIESIS IN A. Popov MV***, Vladareanu AM****, Kovacs E**, Savopol T**, Iordache M**, Begu M****, Bumbea H****, Onisai M****, Nicolescu A****, Popa C*. *County Emergency Hospital Arges, Hematology Department; **University of Medicine and Pharmacy Carol Davilla, Bucharest; ***University Emergency Hospital Bucharest

Introduction: Medullary hematopoiesis is commonly found in hereditary hemolytic anemia in chronic myeloproliferative syndromes, the most common place where this appears being spleen. There is a very rare combination of these two hematologic diseases. **Objectives:** In the literature only two cases have been described and in both, chronic myeloproliferative syndrome was detected many years after therapeutic splenectomy, as treatment of hemolytic crisis in hereditary spherocytosis. **Material and Methods:** We present the case of a patient with known hereditary spherocytosis who underwent the splenectomy complicated with the portal vein thrombosis after one month, at which time he was diagnosed with chronic myeloproliferative syndrome Unclassified JAK positive. **Results:** This patient showed abnormal platelet function (platelet aggregation with ADP, collagen, ristocetin and epinephrine), high level of oxidative reactive species, which was performed by fluorimetric method with DCFDA. **Conclusions:** In literature there were described only a few cases of the coexistence of a hereditary hemolytic anemia and chronic myeloproliferative syndrome. It was indicated an increase of oxide radical production in patients with chronic myeloproliferative disorders that may explain abnormal platelet function. The increase of oxide radicals worsen the symptoms of hereditary hemolytic anemia, causing hemolysis by altering membrane fluidity, due to oxidation of lipids

A1149

TISSUE FACTOR AND HIGH RESIDUAL PLATELET REACTIVITY IN PATIENTS TREATED WITH CLOPIDOGREL. Quintanar-Trejo L***, Flores-García M**, Valente-Acosta B**, Calderón-Cruz B**, Peña-Duque MA**, Martínez-Ríos MA**, De la Peña-Díaz A****. *Departamento de Farmacología, Facultad de Medicina, UNAM; **Grupo de Genética Interven-

cionista. Departamentos de Biología Molecular y Hemodinámica, Instituto Nacional de Cardiología Ignacio Chávez.

Introduction: Tissue factor (TF) is a key molecule in the hemostatic system, present in different cells of the vascular wall and the bloodstream. It is known that only a small amount of FT exerts a pro-coagulant activity, as inactive forms are encrypted inside the cell. However, after cellular activation, FT exposure and activity increases. A heterogeneous and variable response to the thienopyridine clopidogrel exists and has been attributed to genetic, pharmacologic, or clinical factors. **Objectives:** Identify concentration and procoagulant activity of the tissue factor in patients under clopidogrel therapy. **Material and Methods:** We performed a case-control study. 262 patients were enrolled who had received a 600 mg dose of clopidogrel after a percutaneous coronarography. At 24 hours, we took a blood sample to assess platelet function. TF was measured with a commercial ELISA kit, and its procoagulant activity analyzed by a two-stage chromogenic commercial assay. The protocol was approved by the Institutional Ethics Committee, and informed consent was obtained from each participant. **Results:** 43 (16.4%) poor responders to clopidogrel, were aged, and gender matched with 43 responders. Poor responders had higher TF concentrations and procoagulant activity compared with responders (470.17 ± 280.44 pg/mL Vs. 352.03 ± 160.01 pg/mL, $p=0.006$), and (8.90 ± 15.19 pM Vs. 4.77 ± 11.91 pM) respectively. In the multivariate logistic regression model, high TF (defined as higher than the 90 percentile of the responders group) was an independent risk factor for clopidogrel resistance (OR=7.174, CI=1.562-32.946 $p=0.011$). **Conclusion:** TF showed to be an independent risk factor for clopidogrel poor response.

A1172

THE EPIDEMIOLOGY DATA OF THE PATIENTS REGISTRY IN THE " FEDERACIÓN DE HEMOFILIA DE LA REPÚBLICA MEXICANA, AC (FHRM). Gaitán C, Monteros M, Benitez H, Berges A, Esparza A, Jaloma A, Reyes U, Vélez M, Gómez M, Gil P, Andrade R, Izquierdo-Franco M, Berrones G, Romo P, Guzmán M, Quintero A, Espinoza C, Mireles A, Bastar M, Ovando A, Ovando S, Davy G, Preza E, Hernández T, Curiel M, Bello S, Barrón H, Castro S, Rodríguez E, Santos A, Rizo M, Chavira G, Martínez M, de la Llata D, Izquierdo-Zarco M, Díaz F, Jiménez J, Fuentes J, Beltrán J, May F, Domínguez E, Nuñez M, Palencia A, Montes Y, Ojeda M, Olivares L, Arredondo G, Losada M, González M, Bermudez, L, Cancino F, Cervantes I, Vázquez A, García G, Guzmán R. Federación de Hemofilia de la República Mexicana, AC.

Introduction: The FHRM was founded in 1985 and is part of the World Federation of Hemophilia (WFH). Incidence of hemophilia in the world is 1: 10,000 inhabitant male. According to the last census in Mexico we are 114,000,000 inhabitants, so there are 5, 700 persons with hemophilia (PWH) but we don't

have a real registry of them. This is the first registry of PWH and others inherited coagulopathy (OIC) performed by the FHRM. **Objective:** To know the number and characteristic of PWH and OIC. **Material and Methods:** FHRM registers the information of PWH and OIC from all of the 32 states of the country. All the pieces of information are registered in a data base which is carefully secured and highly confidential. The FHRM is the owner of this data base and responsible for its actualization. The register began since 2000. **Results:** 4, 527 PWH are found in Mexico. Inhibitors in hemophilia A are present in 169 patients and 10 in hemophilia B. Infected with HIV are 24 PCW and contaminated with hepatitis C virus 118.

Distribution by type and severity of hemophilia and other inherited coagulopathy

	Severe	Moderate	Mild	Unknown
Hemophilia A (3 582)	901(25%)	904(25.2%)	742(20.7%)	1 032(28.8%)
Hemophilia B (576)	188(32.6%)	141(24.4%)	113(19.6%)	134(23%)
Undetermined Hemophilia vWD (191)	369(8.8%)			
Factor V (2)				
Factor VII (5)				
Factor X (1)				
Glanzmann T (1)				

Age groups and types of hemophilia and von Willebrand disease

Age (years)	Hemophilia A	Hemophilia B	vWD
0-4	99	16	5
5-13	578	99	23
14-18	426	78	25
19-44	1382	225	55
>45	276	54	8
Unknown	821	104	75

Conclusion: At this moment there are 4, 527 PWH are registered of the 5, 700 expected. There is a high number of PWH (1,166), more than 28% which are not classified for its severity yet, this is associated to the lack of resources available in some of the States to perform laboratory tests. In a smaller number of PWH (8.8%) the type of hemophilia is unknown. Also the number of patients with VWD and OIC is much lower than the numbers showed in literature. The FHRM is still working with medical doctors and associations to improve the record of these patients.

A1173

ANTIAGGREGANT EFFECT OF NEW ESTROGEN ENANTIOMERS. Flores-García M^{*,5}, Fernandez-G JM^{***}, Marín-Ocampo R^{****}, Ortiz-Cárdenas AL^{****}, Gutiérrez-Macias NG^{**}, Guerrero-Silva TD^{**}, León-Martínez M^{***}, Uribe-Lesama EC^{****}, Tovar-Vargas MA^{**}, Acevedo-Cuevas A^{*}, Pinzón E^{**}, de la Peña A^{***}. *Instituto Nacional de Cardiología Ignacio Chávez; **Facultad de Medicina, Universidad Nacional Autónoma de México; ***Instituto de Química, Universidad Nacional Autónoma de México; ****Facultad de Química de la Universidad Nacional Autónoma de México; ⁵Escuela Superior de Medicina, Instituto Politécnico Nacional.

Introduction: Most of the drugs that we use today for the treatment of different diseases are racemic mixtures. There is little information regarding the activity of enantiomers of estrogenic compounds. Moreover, the administration of the more active enantiomer could allow lower dosing. Continuing with our program of design and synthesis of new estrogen compounds with antithrombotic activity, we had designed and synthesized N-[(3-Hydroxy-1,3,5(10)-estratriene-17b-il)]-(R)-(+)-b-methylphenethyl-amine (R-fenetamE) and N-[(3-Hydroxy-1,3,5(10)-estratriene-17b-il)]-(S)-(+)-b-methylphenethyl-amine (S-fenetamE). **Objectives:** Determine the effect of new estrogenic enantiomers, R- and S-fenetamE on platelet aggregation in platelet-rich plasma (PRP). **Material and Methods:** This study was conducted according to the principles outlined in the Helsinki Declaration. The blood samples were obtained from healthy donors in the blood bank of the Instituto Nacional de Cardiología. For each assay a blood from 4 subjects, was collected by venipuncture into plastic tubes containing anticoagulant. The samples were centrifuged at 140g for 5 min to obtain the PRP, which were carefully withdrawn and pooled. The platelet count was adjusted to 250x10³/uL with platelet-poor plasma (PPP), which was obtained by centrifugation of pellet at 1200g for 20 min. The estrogen derivatives was designed and synthesized at Instituto de Química, UNAM. Which were dissolved in DMSO 5%, and this was used as control and considered as 100% of response. The aggregometry was performed using a lumi-aggregometer. We performed a curve concentration-response (0.5 to 500 uM). Platelet aggregation was induced with 5 uM ADP, 10 uM epinephrine and 1 ug/mL collagen. Aggregation was measured as a percentage of light transmission relative to PPP and was recorded for 6 min after the addition of the agonist. **Results:** ADP- and epinephrine-induced platelet aggregation was inhibited by both enantiomers, in a concentration dependent manner, contrasting the result obtained with collagen-induced platelet aggregation. IC₅₀=350 and 412 uM, and 370 and 450 uM, for R- and S-fenetamE, respectively, with epinephrine and ADP as agonist. **Conclusions:** The platelet aggregation was inhibited by both enantiomers; however, the S-enantiomer had higher inhibitory potency than the corresponding R-enantiomer. The S-enantiomer has significant potential and should be further examined in murine studies.

A1195

FARMACOGENOMIC STUDY OF PRASUGREL POOR RESPONSE. Calderón-Cruz B^{*}, Rodríguez-Galván K^{*}, Manzo-Francisco LA^{*}, Román-Pérez A^{*}, Herrera-González A^{*}, Peña-Duque MA^{**}, Pérez-González A^{**}, Vargas-Alarcón G^{**}, Fragoso-Lona JM^{**}, Reyes-Gómez CA^{**}, Martínez-Ríos MA^{**}, De la Peña-Díaz A^{*}. *Faculty of Medicine, Pharmacology Department, National University Autonomous of Mexico (UNAM), Mexico City, Mexico; ** Hemodynamic Department, National Institute of Cardiology, Interventionist Genetic Study Group, Mexico City, Mexico.

Introduction: Prasugrel is a novel prodrug that is hidrolized by intestinal esterases into an intermediary metabolite R-95913, which by the action of CYP3A5, CYP2B6 and CYP2C19 enzymes, is transformed to its activated form R-138727. This activated form antagonize ADP-receptor P2Y₁₂, inhibiting irreversibly ADP pathway during platelet aggregation. **Objectives:** Evaluate prasugrel poor response frequency and its possible association with the polymorphisms of ABCB1, CYP2C19 and P2RY12 genes. **Material and method:** We included a total of 52 patients who underwent percutaneous coronary intervention at Instituto Nacional de Cardiología, and had received a prasugrel loading dose of 60 mg. Platelet aggregation was performed at 37°C in an aggregometer (Model 560 CA and accompanying software Model 810 AGGRO/LINK Chrono-log, Havertown, PA, USA) induced with 10 uM ADP. Single nucleotide polymorphisms (ABCB1, CYP2C19*2, CYP2C19*3, CYP3A5*3 and two polymorphic sites of the P2RY12 gene G52T and C34T) in genomic DNA were performed using 5' exonuclease TaqMan assays by a 7900HT Fast Real-Time PCR System (Applied Biosystem, Foster City, CA, USA). **Results:** Prasugrel poor response was found in 9 (17.3%) patients, only one of them had the ABCB1 polymorphism (TT), in the rest of the studied genes of the poor response patients no polymorphisms were found, most of them were wild type. The whole study population resulted in wild type for the CYP2C19*3 allelic variant. **Conclusions:** Prasugrel poor response was no associated to any of the allelic variants studied. The obtained results suggest the possibility of other unknown mechanisms that may explain the resistance. Supported by DGAPA UNAM IN221112

A1197

CLINICAL DIAGNOSIS AND LABORATORY SCREENING IN VWD PEDIATRIC PATIENTS OF THE NORTHWESTERN OF MEXICO. Jaloma-Cruz AR^{*}, Muñoz-Robles DC^{**}, Sotopadilla J^{**}. *Centro de Investigación Biomedica de Occidente, IMSS, Guadalajara, Jalisco, Mexico; **UMAE Hospital de Pediatría, Centro Medico Nacional de Occidente, IMSS, Guadalajara, Jalisco, Mexico.

Introduction: von Willebrand disease (vWD) is the most common inherited bleeding disorder affecting around 1% of the

world population. The diagnosis approach is complex and must include bleeding history, clinical examination and several biological laboratory tests. In Mexico there are few studies in vWD population, but the real incidence of vWD patients is unknown.

Objectives: To determine the correlation between bleeding symptoms and laboratory screening to detect different types of vWD in pediatric population from the Northwestern of Mexico.

Material and Methods: After obtaining informed consent, a pediatrician applied a clinical questionnaire of familial bleeding history and bleeding symptoms in children, who were subject to general physical examination and detection of bleeding symptoms. Plasma and DNA samples were obtained for laboratory testing: general clotting screening (BH, PT, aPTT, fibrinogen, Ivy BT); initial confirmatory testing (vWF:Ag, vWF:RCo, FVIII:C); final confirmatory testing (vWF multimers and molecular vWF gene analysis). **Results:** We studied 25 pediatric patients from 22 independent families native of Jalisco, Colima and Michoacán (11 girls, 14 boys), with ages between three and 16 years old. 21/25 (84%) have familial bleeding history; 19/25 (76%) were referred by bleeding symptoms and 6/25 (24%) after prolonged clotting times in pre-surgical testing. All patients but one have showed previously bleeding symptoms of variable importance. The main bleeding symptoms are petechiae and ecchymoses, epistaxis, bleeding tooth extractions and hematomas. By the ratio values ($<0,6$ or $>0,6$) of vWF:Ag/WF:RCo and FVIII:C/vWF:Ag we suggest a preliminary diagnosis of 10 patients with type 1 vWD, 6 type 2 vWD (2A, 2B or 2M), one type 2N vWD and 8 patients with normal values. All patients will be further confirmed by multimer vWF analysis. **Conclusions:** We found a good correlation between abundance and severity of bleeding symptoms in the patients and their initial confirmatory laboratory testing results (vWF:Ag; vWF:RCo; FVIII:C). Project supported by the World Federation of Hemophilia through the Federación de Hemofilia de la República Mexicana and CSL-Behring of México.

A1204

ESSENTIAL THROMBOCYTOSIS, EPIDEMIOLOGICAL DESCRIPTION AT THE INSTITUTO NACIONAL DE CIENCIAS MÉDICAS Y NUTRICIÓN SALVADOR SUBIRAN. González-Zenteno S*, Scherling-Ocampo A*, Agreda Vásquez G*. *Departamento de Hematología, Instituto Nacional de Ciencias Médicas y Nutrición Salvador Subiran, México D.F.

Introduction: Essential thrombocythemia (ET) is a myeloproliferative neoplasm characterized by increased risk of thrombohemorrhagic complications, and a natural propensity toward leukemic or fibrotic transformation. **Objectives:** Description of cases of essential thrombocytosis (ET) during the period between 2000 and 2011, total cases, treatment, course and prognosis. **Material and Methods:** We reviewed the medical records with clinical admittance or withdrawal diagnosis of TE of January 1, 2000 to December 31, 2011, we included all patients with confirmed diagnosis of TE and were entered into

the database and exported the Excel-based cases and performed the statistical analysis. **Results:** We analyze a total of 57 cases of ET with respect to their risk level, risk factors, cytogenetics, type of treatment, thrombotic events, progression to myelofibrosis and prognosis with respect to transformation to leukemia. Of these 4 cases (7%) progressed to myelofibrosis and 1 of these cases to acute leukemia and one case was transformed to leukemia cutis, a total of two acute leukemia (3.5%). **Conclusions:** The ET is a myeloproliferative neoplasm with the presence of thrombotic events, and the presence of these has been associated with Jak-2 mutation positive and leukocytosis, but these factors have not been standardized, but is known from several series the relationship thrombotic events between these factors. Now with respect to the transformation to leukemia is described also the presence of myelofibrosis and cytogenetic risk factors for the transformation to leukemia. In our experience of 10 years seem to reproduce these findings, although few cases of secondary leukemia in order to establish this association.

A1206

EXPERIENCE OF THE INSTITUTE OF MEDICAL SCIENCES AND NUTRITION SALVADOR ZUBIRÁN IN PATIENTS WITH THROMBOTIC THROMBOCYTOPENIC PURPURA. Dell-Sanchez JL, Amador-Medina F, Vargas Ruiz AG. Institute of Medical Sciences and Nutrition Salvador Zubirán

Introduction: Acquired thrombotic thrombocytopenic purpura (TTP) is an acute, life-threatening illness characterized by thrombocytopenia, microangiopathic hemolytic anemia (MAHA), and often signs of organ dysfunction—typically neurologic, cardiac, renal, or abdominal symptoms. In the majority of cases, there is no obvious underlying precipitant, although a genetic link has been identified. The mainstay of treatment remains plasma exchange (PEX) and steroids. **Objectives:** Describe the clinical features. Laboratory parameters. Treatments used. Mortality. Overall survival. Disease-free survival. **Material and Methods:** This was a retrospective descriptive study. Adults over the age of 18 years presenting with de novo or relapsed acute TTP were eligible for entry into the trial. The diagnosis was based on thrombocytopenia, microangiopathic hemolytic anemia, normal clotting screen, and increased lactate dehydrogenase (LDH) to one and a half times the normal upper limit. A total of 25 patients with a diagnosis of acute TTP were entered between 2000 and 2010 and follow-up was for at least 22 months after admission. **Results:** The female sex predominated with 89.5% of cases. The median age was 39 years. 94.7% of patients had neurological symptoms (headache, stroke, coma and seizures). Only a small percentage of patients presented with fever and abdominal symptoms, 21.1% and 15.8 respectively. The median hemoglobin, platelets, DHL, creatinine and indirect bilirubin at diagnosis was 7.6 g/dl, 12,000, 1151 IU, 0.9 mg/dl, 1.9 mg/dl respectively. All patients received steroids and plasmapheresis as a treatment. The median plasmapheresis sessions to achieve complete remission was 16. Other treatments were also required in 36.8% of cases. 3 patients

were splenectomized. 24% of patients relapsed. 24% of patients died. The median follow up was 22 months. The median relapse was 6 months. **Conclusions:** I. Sustained remission rates and relapse rates are similar to those mentioned in the literature and correspond to 75-80%, 25-50%, respectively. II. Unlike previous series expressing, our work showed that the onset of symptoms were mainly neurological disease.

A1208

PANEL OF GENETIC MARKERS ASSOCIATED TO PRE-DISPOSITION OF THROMBOPHILIA IN PATIENTS OF DIFFERENT ETIOLOGY. Fuentes-Chavez CA*, Lopez-Jimenez JJ**, Jaloma-Cruz AR**. *Maestria en Biomedicina Clinica, Ciencias Químico-Biologicas, Universidad de las Americas Puebla, Cholula, Puebla, Mexico; **Centro de Investigacion Biomedica de Occidente, IMSS, Guadalajara, Jalisco, Mexico.

Introduction: Thrombosis like multifactorial entity, involves genetic predisposition and diverse environmental risk factors. In most of the cases the identification of the genetic cause is not achieved. Genetic factors related to coagulation could explain the development of thrombotic events. **Objectives:** To evaluate the diagnostic informativity for thrombophilia from a panel of coagulation genetic markers involving endothelium, hemostasis and fibrinolysis: FVG1691A, FIIG20210A, MTHFR [C677T y A1298C], ECA-1 I/D Int16, PAI-1 5G/4G y FT D/I 5'. **Material and Method:** DNA samples were obtained after informed consent from 168 patients who suffered thrombotic events by diverse etiology. Genetic polymorphisms were identified according original protocols by PCR and enzyme-restriction analysis and genotyping were realized by PAGE and silver staining. **Results:** The frequencies of polymorphisms FVG1691A ($p < 0.05$); MTHFR A1298C, PAI-15G/4G, ECA-1I/D ($p < 0.01$) and FIIG20210A ($p = 0.05$) in thrombophilic patients showed significant statistical differences respect to general population by Chi2 analysis and were similar for MTHFR C677T y FT D/I 5'. In 75% of thrombophilic cases the presence of at least one of the genetic polymorphisms was found ($p < 0.05$). **Conclusions:** The selected panel showed high informatively for molecular diagnosis of thrombophilia in the studied patients. The most frequent polymorphism in Mexican population is FIIG20210A and our results suggest an association of MTHFR A1298C homozygous CC mutant in affected population. The heterozygous MTHFR [677C/T-1298A/C] genotype can be considered as candidate for arterial thrombosis risk factor.

A1209

SEVERE HEMOPHILIAC WITH A HIGH RESPONSE INHIBITOR WHO DEVELOPED THROMBOSIS. Pérez-Lozano U. Ruiz Ovalle JL. Limon Flores JA. Lobato Tolama RD. IMSS UMAE Puebla.

Introduction: It has been suggested that hemophilia is a phenomenon that protects against thrombosis, there are 49 cases

reported in the literature of severe hemophiliacs who develop thrombosis. Hepatitis C (HCV) has been identified as a risk factor for thrombosis. **Objective:** To describe the evolution of a patient with severe hemophilia with inhibitors who develops thrombosis. **Material and Methods:** 67 year old male, the father died of acute myocardial infarction, the mother died of a stroke, four hemophiliac brothers died from hemorrhages during childhood, another brother died from an acute infarction at 30, two living sisters who are carriers of hemophilia A, two nephews with severe hemophilia A. At the age of seven he presented moderate bleedings treated with fresh plasma in the hospital, he had less than two bleedings per year in non vital sites. He attended our service in 1996 with a diagnosis of Hemophilia B with FIX 32%. A new determination of FIX 9.3%, FVIII 0.7%, FVW84% was requested. He started plasma derived factor VIII. **Results:** In January 2006 he presented inhibitor against factor VIII of 101 UB. Due to the lack of bypass therapy he was treated with high doses of factor VIII due to once a month bleedings in target joint. In 2010 the PCR for HCV was 2010000 UI/ML genotype 2B, he received Peg – Interferon a 2b and rivabirin. In September 2011 he presented with chest pain that is typical of angina pectoris with CPK MB and elevated troponin, the electrocardiogram showed ischemia and necrosis in lower aspect of left ventricle, treated with vasodilators and aspirin without thrombolysis. The FEVI was 40% without HAP. He developed compartmental syndrome due to a hematoma on the left arm after arterial blood gases receiving factor VIII and low molecular weight heparin. At discharge he was prescribed 100mg of aspirin per day. Two months after he was discharged he referred a decrease in visual acuity, the fundoscopy showed thrombosis of the central vein in the left retina. He attended the outpatient care due to a 20 cm hematoma on the neck's skin and reported that he suspended the aspirin. A factor VIII inhibitor study was performed finding 512 UB. **Conclusion:** Given the longer survival rate of individuals with hemophilia, we expect thrombotic complications even in patients who are considered at low thrombotic risk.

A1230

RALOXIFENE AND BEVACIZUMAB IN A PATIENT WITH HEREDITARY HEMORRHAGIC TELANGIECTASIA (HHT) EXPERIENCE OF A CASE. Ledesma C, Payns E, Guzman L. IMSS

Introduction: HHT known as Osler Weber Rendu is a relatively common disease characterized by an autosomal dominant disorder, with two types HHT1 and HHT2, haploinsufficiency resulting in ENG and ALK1 proteins, the manifests as dysplasia clinic vascular system characterized by telangiectasias and arteriovenous malformations of the skin, viscera and mucous membranes, affecting all racial groups is the frequency distribution of 1 person per 5000 or 10000. Raloxifene to be a selective modulator of estradiol, is used in postmenopausal patients with this condition because it increases the expression of these genes haploinsufficiency, the bevacizumad be a molecular target of

VEGF, has been used by alterations in angiogenesis demonstrated in the condition. **Objective:** Show the use of raloxifene and bevacizumab in patients with HHT. **Material and Methods:** Female 60 years old, no history of hereditary hemorrhagic importance. Paternal side, unknown. APP: tonsillectomy at age 5, thyroidectomy for thyroid CA in 1993, approximately 13 surgeries nasal cautery and once with perforation of the nasal septum, multitransfused with HCV secondary. 4 children without studies to determine the condition, however two of them with symptoms of epistaxis. is current condition starts at 37 years old, presenting severe epistaxis, 1 once every month for 3 years, now daily, has been treated with transfusions and 5 months ago intranasal application bevacizumab 3 times, last time December. initiating raloxifene management five months ago to improve bleeding disorders. now without bleeding disorders at any level. **Results:** Scale was used to assess the epistaxis Sadick. initial: Grade I less of a once a week. Grade II several times a week. Grade III more than once a day. Patient with grade III. after treatment. Grade I specks on paper. Grade II handkerchief soaked. Grade III need to use something local. Patient Grade I. **Conclusions:** Hemorrhagic telangiectasia ataxia is an autosomal dominant disorder difficult to treat, requiring a multidisciplinary approach, currently under pathophysiologic mechanisms found to have attempted to administer the treatment in patients, in the case shown the two drugs have been improve the patient's clinical course, we know that raloxifene increases the risk of thrombosis, because it raises the FV, FVIII and protein C, in the case of patients with this disorder might be beneficial in this situation to be a tendency to the bleeding. We believe that to be a rare and difficult to treat more studies should be done to improve future management of these patients.

A1233

ASSOCIATION OF VON WILLEBRAND FACTOR, NITRIC OXIDE AND CAROTID INTIMA-MEDIA THICKNESS IN POSTMENOPAUSAL WOMEN. Córdova N,* Basurto L*, Vázquez A.L*, Vargas A*, Díaz A*, Jiménez A*, Villareal A*, Zárate A*, Reyes E**, Rosales E**, Martínez –Murillo C***. *Endocrine Research Unit and Blood Central Bank, National Medical Center, Instituto Mexicano del Seguro Social; **National School Biological Sciences IPN; ***Thrombosis and

Hemostatic Unit, General Hospital of Mexico SSA. Endocrine Research Unit and Blood Central Bank, National Medical Center, Instituto Mexicano del Seguro Social and Thrombosis and Hemostatic Unit, General Hospital of Mexico SSA.

Introduction: The Metabolic Syndrome (MS) is associated with the development of diabetes and increased cardiovascular mortality. The prevalence of the MS increases with menopause and may partially explain the apparent acceleration in cardiovascular disease in postmenopausal women. It was proposed that endothelial damage may be a central component of metabolic syndrome. Plasma von Willebrand factor (vWf) and nitric oxide (NOx) are a recognized circulating markers of endothelial damage/dysfunction and intima-media thickness of the walls of the carotid artery predicts cardiovascular outcomes. **Objectives:** The aim of this study was to analyze the relationship of vWF, NOx and carotid intima-media thickness in pre- and postmenopausal women. **Material and methods:** Cross sectional study including 189 women aged 45 to 60 years. Participants were interviewed form medical history and underwent a complete physical examination. Metabolic Syndrome (MS) was defined according to the International Diabetes Federation. vWF was estimated by immunoassay. NOx was measuring by Griess reagent, the mean intima-media thickness was measured by ultrasonographic images. **Results:** Results: Of the total of 189 participants, 41 (32.1%) were premenopausal and 68 (62.4%) were postmenopausal women. 32.4 % had MS and 67.6% were healthy women. vWF levels were similar in both groups. NOx of premenopausal women were $247 + 37.2$ uM, in comparison with $227 + 15.5$ uM in postmenopausal women. It was observed a correlation between vWf and NOx ($r = -0.224$, $p < 0.05$); this correlation remained significant after adjustment for BMI. The mean intima-media thickness was significantly increased in postmenopausal women ($0.61 + 0.17$ vs $0.68 + 0.01$, $p < 0.05$). **Conclusions:** Our data suggest more severe endothelial dysfunction in postmenopausal women. The association observed between NOx and vWf can be explained by hypercholesterolemia and atherosclerosis which interferes with the NOx production. Consequently, an increase in vW is produced. Nevertheless, we state how the weight was influential in getting these results, therefore. It is relevant to avoid the body weight increase in postmenopausal women.

TRANSPLANTATION

ORAL PRESENTATION

A1038

GENERATION OF NEURAL PRECURSOR CELLS FROM HUMAN UMBILICAL CORD BLOOD OR ALTERNATIVE TISSUE SOURCES. Ramírez-Jiménez EA, Toledo-Hernández D, Torres-Rodríguez P, Arce-Castañeda G, Esquivel-Estudillo J, Jaimes-Jiménez VD, Mojica-Cardoso C, García-Esquivel FI, Tenorio-Mina A, Colli-Magaña D, Nolasco-Quintana NY, García-Leyva JA, Acosta-Velazco K, Stephenson-Gussie T, Ángeles-Chimal JS, Acevedo-Fernández JJ, Rodríguez-Molina VM, Petricevich-López VL, Santa-Olalla-Tapia J. Unidad de Diagnóstico y Medicina Molecular, Facultad de Medicina/ Universidad Autónoma del Estado de Morelos-Hospital del Niño Morelense.

Introduction: The in vitro expansion of human neural precursor cells (HNPCs) will establish appropriate procedures for cell therapy in patients with neurodegenerative diseases. The challenge now is to isolate or generate these cells, which can be obtained from healthy nervous tissue or be generated from alternative cellular sources such as adult tissue or blood cells. It has been reported the generation of HNPCs from several tissues, but low efficiencies have been reported. Actually, there are no studies that compare HNPCs generation from alternative human tissues sources using the same procedure. **Objective:** The aim of this study is to evaluate several variables that affect HNPCS generation from different sources. **Material and Methods:** The analyzed tissues were skin, adult peripheral blood (APB), bone marrow and human umbilical cord blood (HUCB). Cells were cultured in defined medium supplemented with EGF and bFGF for twelve days, they were plated in 96 well-plate with density curve from 1×10^4 to 1×10^6 cells/well. Evaluation of Skin, Hematopoietic and neural markers was performed using RT-PCR.

Results: We standardized the procedure for collection of these tissues. Appropriate conditions were identified by the detection of neural differentiation markers with RT-PCR. In HUCB and APB, we characterized the mononuclear fraction isolated. At twelve days of culture aggregates were obtained and the expressions of neural differentiation markers confirmed the HNPCs identity. We determined optimal density (2.5×10^5 - 7.5×10^5 cells/well) for HUCB and APB to generate neurospheres. In skin, there was a

better generation of aggregates between 2×10^4 - 8×10^4 cells/well. **Conclusions:** It was established as an important variable the cellular density. It is necessary to identify the conditions that allow us the generation of HNPCs efficiently, to improve the propagation of HNPCs from tissues from the same patient to suppress the graft rejection.

A1058

BODY MASS INDEX AS AN INDICATOR OF PROGNOSIS IN PATIENTS UNDERGOING ALLOGENEIC HEMATOPOIETIC STEM CELL TRANSPLANTATION. Ruiz-Delgado GJ*****, Lutz-Presno JA****, Alarcon-Urdaneta C*****, Calderón-García J*⁵, Ruiz-Argüelles GJ*****. *Centro de Hematología y Medicina Interna. Clínica Ruiz; **Laboratorios Clínicos de Puebla. Clínica Ruiz; ***Universidad Popular Autónoma del Estado de Puebla; ****Universidad Autónoma de Puebla; ⁵Facultad de Medicina. Universidad La Salle. México.

Introduction: Both obesity and malnutrition are considered risk factors for complications and increased relapse and nonrelapse mortality in hematopoietic stem cell transplantation (HSCT). An inferior outcome after allogeneic HSCT has been reported in obese adult patients in both allogeneic and autologous HSCT: Overweight individuals seem to develop more complications of graft versus host disease and more infections than its normal counterparts. **Objective:** To elucidate the impact of pretransplantation body mass index (BMI) on clinical outcome in patients who underwent allogeneic HSCT in the Centro de Hematología y Medicina Interna of the Clínica RUIZ in Puebla, MEXICO. **Material and Methods:** Between March 1996 and December 2010, a total of 138 patients received an allogeneic stem cell transplantation in the Centro de Hematología y Medicina Interna of the Clínica Ruiz. Patients were stratified according to pretransplantation body mass index (BMI) values. **Results:** Seventeen patients had low BMI, 62 had normal BMI and 59 patients had high BMI. Median overall survival (OS) for these three groups were respectively 9, 12 and 22 months. Patients with a low BMI had a lower OS than those with a normal BMI (58-month OS of 24% versus 32%), whereas patients with an increased BMI had a better outcome (median OS of 22 months and 43% OS at 130 months) than those with a normal BMI. **Conclusions:** Our findings demonstrate a correlation between pretransplantation BMI

and posttransplantation survival and should provide insight into how to better manage nutritional support for patients undergoing hematopoietic stem cell transplantation

A1085

INCIDENCE OF GVHD IN ABO-INCOMPATIBILITY RECEPTORS OF HEMATOPOIETIC STEM CELL TRANSPLANT USING A REDUCED INTENSITY CONDITIONING. Gutiérrez-Aguirre CH, Alatorre-Ricardo J, Cantú-Rodríguez OG, González-Llano O, Jaime-Pérez JC, Mancias-Guerra C, Flores-Jiménez JA, Gómez-Almaguer D. Servicio de Hematología del Hospital Universitario ÓDr. José Eleuterio González, UANL. Monterrey N.L., México.

Introduction: Graft versus host disease (GVHD) is one of the major causes of morbidity and mortality after allogeneic hematopoietic stem cell transplantation (allo-HSCT). Major ABO incompatibility is a serious barrier to solid organ transplantation but allo-HSCT can be performed successfully across ABO-mismatching between donor and recipient. There are several lines of evidence suggesting that minor ABO-incompatibility may be associated with early and late transplant complications including increased risk of GVHD. **Objectives:** We retrospectively analyzed the impact of ABO blood group matching on the outcome of patients who underwent reduced intensity HSCT and their relationship with acute and chronic GVHD incidence. **Material and Methods:** We included 142 patients from the University Hospital in Monterrey Mexico, 86 men and 56 women. The patients received allo-HSCT from HLA-identical relatives (129 patients), haploidentical-HSCT (12 patients) or syngeneic-HSCT (1 patient) using a reduced intensity conditioning regimen. The median age was 33 years (1-71). The median number of infused CD34+ cells was 5.5×10^6 ($1.2 \times 10^6 - 13.6 \times 10^6$). ABO and Rh typing was performed by standard blood banking techniques. **Results:** The frequency of ABO and Rh blood group in our patients was: A 36 (25%), AB 4 (3%), B 10 (7%), O 92 (65%), Rh positive 130 (91%) and Rh negative 12 (9%) patients. The frequency of ABO and Rh blood group in donors was A 45 (32%), AB 2 (1%), B 8 (6%), O 87 (61%), Rh positive 129 (91%) and Rh negative 13 (9%) donors. One hundred and three patients (72%) received ABO matched and 39 (27%) ABO mismatched transplants, 129 patients (91%) received Rh matched and 13 (9%) Rh mismatched. The overall incidence of GVHD was 37% (53 patients). Comparing both, matched and mismatched ABO groups, the incidence of GVHD was 36% (37 patients) and 41% (16 patients) respectively ($p=0.6$), acute GVHD was 18% (19 patients) and 20% (8 patients) respectively ($p=0.8$), and chronic GVHD was 24% (25 patients) and 23% (9 patients) respectively ($p=1.0$). After a median follow up of 8.5 months (0-79) 56 ABO matched patients (55%) were alive compared to 26 (67%) from the ABO mismatched blood group. There was no significant association between ABO matching status and survival ($P=0.46$). **Conclusions:** In this study we find

that ABO-mismatches between donor and recipient had no influence in the incidence of GVHD and had not a measurable effect on survival.

A1117

100-DAY MORTALITY AFTER ALLOGENEIC HEMATOPOIETIC STEM CELL TRANSPLANTATION (ALLOHSCT) USING REDUCED DOSE OF BUCY AND G-CSF-PRIMED BONE MARROW. Leon-Rodríguez E, Perez-Alvarez SI, Guzman-Uribe P, Tuna Aguilar E, Espinosa Bautista KA, Castro Sanchez A, Armengol Alonso A. Departamento de Hematología-Oncología. Instituto Nacional de Ciencias Médicas y Nutrición. México D.F.

Introduction: The high morbidity and mortality associated with alloHSCT remains a limiting factor in its effectiveness. Therefore, we decided to introduce a new conditioning regimen with ~20% reduction of the standard BUCY dose and G-CSF-primed bone marrow. **Objectives:** The aim of this study was to describe epidemiological data, 30 and 100-day mortality in patients with alloHSCT. **Material and methods:** A prospective analysis of patients with alloHSCT using G-CSF-primed bone marrow and reduced dose of busulfan and cyclophosphamide was conducted at the INCMNSZ. Epidemiological, clinical characteristics and survival were described. Survival analyses were assessed by the Kaplan-Meier method. **Results:** We analyzed 29 patients undergoing alloHSCT between 1999 and 2011, using reduced dose of BUCY (BU: 12mg/kg and CY: 80mg/kg) and G-CSF-primed bone marrow (10ug/Kg/ day for 3-5days). Median age was 29 years (range 16-59) and 19 patients were men (66%). Diagnosis was myelodysplastic syndrome (MDS):10 patients (35%); chronic myeloid leukemia (CML): 9 (31%); acute lymphoid leukemia (ALL): 6 (21%); acute myeloid leukemia (AML): 2 (7%) and paroxysmal nocturnal haemoglobinuria (PNH): 2 (7%). Median of infused CD34+ cell dose was 1.9×10^6 /kg (range 0.99-4.5 $\times 10^6$ /kg). Median time to neutrophil engraftment was 20 days (range 14-29) and to platelet recovery 15 days (range 7-36). Acute graft-versus-host disease (aGVHD) was observed in 2 (7%) and chronic GVHD in 9 (31%). There was no mortality at 30 days and the 100-day mortality was 7% (2 patients), attributed to infectious complications. With a median follow-up of 43 months (range 2-149), the 5 year overall survival was 70%. **Conclusions:** These results support that the regimen described is associated with minimal morbidity and mortality including low incidence of aGVHD with cytotoxic and immunosuppressive effects that can eradicate the malignant clone, representing a new alternative in the field of alloHSCT.

A1218

RISK FACTORS FOR DEVELOPMENT OF ACUTE GRAFT VERSUS HOST DISEASE IN PEDIATRIC PATIENTS WITH ALLOGENEIC HEMATOPOIETIC STEM CELL TRANSPLANTATION IN PEDIATRICS NATIONAL INSTITUTE FOR

THE PERIOD JANUARY 1998 - DECEMBER 2011. Olaya-Vargas A, Pérez-García M, Castorena-Villa I, López García G, Del Campo A. Instituto Nacional de Pediatría. México DF. México.

Introduction: Acute graft-versus-host disease (aGVHD) and chronic (cGVHD) represent the most frequent and serious complication that occurs after of allogeneic transplantation of hematopoietic progenitors, the frequency of occurrence of GVHD grade I according to reports in the literature is 30% and grade II to IV ranges from 30 to 80% of patients, such as wide ranges are determined by various risk factors such as differences in transplantation protocols, the origin of the donor stem cells and the various schemes of prophylaxis against the disease, GVHD is responsible for 15 to 40% of post-transplant mortality of patients. **Objectives:** Identify the major risk factors that influence the development of aGVHD in patients receiving allogeneic hematopoietic stem cell transplantation (allo-HSCT) assisted in the service of Hematopoietic Stem Cell Transplantation in the National Institute of Pediatrics in the period January 1998 to December 2011. **Material and Methods:** We performed univariate analysis to determine the frequency of occurrence of aGVHD for each risk factor, by knowing bivariate associations between variables in estimating relative risk in the associations studied. Chi square was employed and

Fisher test for determination of statistical significance. The variables in the bivariate analysis were significant were entered into multivariate analysis (logistic regression) to determine the contribution of each of the factors for the appearance of disease. We considered significant value $p = 0.05$. **Results:** During this study, in the Institute has conducted allo-HSCT to 115 patients, with various illnesses, which are grouped as follows: hematologic malignancies 69 cases (60%), primary immunodeficiencies 27 cases (23.4%), benign hematological diseases 12 cases (10.4%), hereditary metabolic diseases 6 cases (5.2%) Others 1 case (0.86%). Of the 115 patients, only 27 of them developed GVHD (23.4%). With the results obtained by univariate and multivariate analysis found that factors which increase the risk for the development of GVHD are the source of peripheral blood (OR 4.11 $p < 0.001$), infection 3 months prior to transplant (OR 4.5 $P < 0.03$) CMV infection in the recipient (OR 4.6 $p < 0.03$), peripheral blood, bone marrow greater than (OR 3.16 $p < 0.001$), and cell dose $> 8.3 \times 10^6/\text{kg}$ (OR 4.6 $p < 0.03$). **Conclusions:** 23.4% of patients developed GVHD, which is a low value that may be secondary to that 50% of transplants are from the umbilical cord which have a lower risk of developing GVHD, however it is important to note that in our series the presence of infections in the receiver are important for the development of the disease.

POSTERS

A1015

POOR PROGNOSTIC FACTORS FOR ANGIOGENESIS IN PATIENTS WITH LOWER LIMB ISCHEMIA WITH HEMATOPOIETIC STEM CELLS AUTOTRANSPLANTED. Gomez-Baute RA, Fernández-Aguila JD, Cabrera-Zamora MM, Rodríguez-Marrero I, Ramirez-Martinez N, Alvarez-García. Dr. Gustavo Aldereguía University Hospital. Cienfuegos. Cuba

Introduction: Introduction The therapeutic potential of adult stem cells in coronary and peripheral arteries has been proposed in recent years. However, some factors may be adverse to the angiogenic potential of these cells are not yet clearly identified. **Objective:** Objectives To identify the prognostic factors for angiogenesis in autologous hematopoietic stem cells applied to patients with chronic lower limb ischemia in the Hospital General Universitario Dr. Gustavo Aldereguía Lima Cienfuegos. **Material and Methods:** Methods A retrospective analytical study of series of cases to assess prognostic factors associated with failure of hematopoietic stem cell autograft because of low angiogenic potential. The universe was constituted by 47 patients (19 women, 28 men) with critical limb ischemia stage IIb, III and IV of the Fontaine classification, treated in our hospital from January 2007 to December 2010, which underwent autologous stem cell transplantation. The variables used were sex, age, personal medical history (hypertension, ischemic heart disease, diabetes

mellitus, chronic renal failure), stage of ischemia according to the Fontaine classification, intermittent claudication, pain on a scale of 1-10, rate pressures in the affected limb, ulcers, and smoking. Analytical variables were also used as blood concentrations of hemoglobin, glucose, creatinine, liver enzymes, cholesterol, triglycerides, LDH and leukocyte count, platelets, stem cell viability, prothrombin time and ESR. As primary endpoint of response was considered failure to perform the amputation. We developed a prognostic score of 1 to 4 for risk factors with significance levels (p) of 0.10, 0.05, 0.025, and 0.01 respectively. **Results:** Results The prognostic factors that had significant values were correlated with poor angiogenesis were late leukocyte count $< 20 \times 10^9 / \text{L}$ after completion of the mobilization with granulocyte colony-stimulating factor, age > 60 years, scoring 10 in the pain score, blood glucose $> 6 \text{ mmol} / \text{L}$ and triglycerides $> 1.8 \text{ mmol} / \text{L}$. In patients with low scores (0-4) of prognostic factors the percentage of non amputations was 90.9% (1 / 11) versus 16.7% (10/12) in the high score (9-12). **Conclusions:** Conclusions We identified five risk factors for low angiogenic potential in autologous stem cells and established a score to predict which patients would be successful or not autologous.

A1023

FLUDARABINE WITH CYTARABINE AND SEQUENTIAL REDUCED-INTENSITY CONDITIONING WITH ALLOGE-

NEIC STEM CELL TRANSPLANTATION IN POOR-RISK CHRONIC LYMPHOCYTIC LEUKEMIA PATIENTS. Doubek M*, Krejci M*, Brychtova Y*, Koristek Z*, Stehlikova O*, Tichy B*, Navratil M*, Tomiska M*, Horky O*, Mayer J*, Pospisilova S**. *Department of Internal Medicine, Hematology and Oncology, University Hospital Brno, Brno, Czech Republic; **Central European Institute of Technology, Masaryk University, Brno, Czech Republic.

Introduction: Allogeneic stem cell transplantation is a treatment option for younger patients (pts) with poor-risk chronic lymphocytic leukemia (CLL). **Objective:** Sequential use of chemotherapy and reduced-intensity conditioning (RIC) for allogeneic stem cell transplantation (SCT) can increase therapeutic effect. **Material and Methods:** Poor-risk CLL was defined by one of the following: refractoriness or early relapse (within 12 months) after treatment with a purine analogue-containing regimen, or progressive disease in the presence of an unfavorable prognostic constellation. Fludarabine (30 mg/m²) and cytarabine (2 g/m²) for 4 days (FC) were used for cytoreduction. After 3 days of rest, RIC consisting of 4 Gy TBI, anti-thymocyte globulin (ATG-Fresenius) 10-20 mg/kg/day for 3 days, and cyclophosphamide 40-60 mg/kg/day for 2 days followed. We analyzed 15 pts with CLL undergoing chemotherapy and RIC SCT in our centre from August 2007 to June 2011. Types of donors and used grafts were as follows: HLA identical sibling, n=6; unrelated donor, n=9, PBSCs, n=13, BM, n=2. Median age of pts was 50 years (range 38-61). Quantitative monitoring of MRD after SCT was performed by flow cytometric analysis and real time PCR. **Results:** The median time of neutrophil engraftment was 16 days, all pts engrafted. The most frequent toxicities were grade III/IV infections according to common toxicity criteria in 12 of 15 pts and gastrointestinal toxicities (grade III in 8 of 15 pts). Incidence of acute GVHD was evaluated in 14 pts: 50% (7/14) of pts had GVHD (grade I+II in 4 pts, grade III in 3 pts). Incidence of chronic GVHD was evaluated in 13 pts, 46% (6/13) of pts had GVHD (limited in 4 pts, extensive in 2 pts). Nonrelapse mortality (NRM) after 1 year and 2 years was 7% and 13%. Causes of death were refractory GVHD (n=1) and infection (n=1). Treatment response was evaluated in 14 pts: complete remission was achieved in 13 pts (93%), MRD negativity (evaluated by both molecular genetic approach and by four-color flow cytometry) after SCT was observed in 10 pts. With median follow-up from SCT 27 months (range 3-33), 80% of all pts (12/15) were alive (11 pts in remission, 1 pt with relapse), 3 pts died (2 death from NRM, 1 death from relapse), 3 relapses (21%; 3/14) occurred. **Conclusions:** FC-RIC protocol represents a promising approach to the treatment of poor-risk CLL with high response rate (93%); progression-free survival and overall survival at 2 years from SCT were 67% and 80%, respectively.

A1059

AN INCREASED BODY MASS INDEX IS NOT AN ADVERSE PROGNOSTIC FACTOR IN PERSONS UNDERGOING

AUTOLOGOUS HEMATOPOIETIC STEM CELL TRANSPLANTATION. Ruiz-Delgado GJ*., Calderón-García J*., Alarcón-Urdaneta C*., Ruiz-Argüelles G*., *Centro de Hematología y Medicina Interna. Clínica Ruiz; **Laboratorios Clínicos de Puebla. Clínica Ruiz; ***Universidad Popular Autónoma del Estado de Puebla; ****Facultad de Medicina. Universidad La Salle. México; ⁵Universidad Autónoma de Puebla.

Introduction: Both obesity and malnutrition have been considered risk factors for complications and increased relapse and non-relapse mortality in hematopoietic stem cell transplantation (HSCT). An inferior outcome after HSCT has been reported in obese adult patients undergoing autologous HSCT; on the other hand, recent data indicate that obesity does not preclude safe and effective HSCT. **Objective:** To elucidate the impact of pretransplantation body mass index (BMI) on clinical outcome in a group of patients who received and autologous HSCT in the Centro de Hematología y Medicina Interna of the Clínica RUIZ in Puebla, MEXICO. **Material and Methods:** Between May 1993 and December 2010, 106 persons underwent an autologous hematopoietic stem cell transplantation in the Centro de Hematología y Medicina Interna of the Clínica RUIZ in Puebla, México, using a simplified method of autografting avoiding cryopreservation of the stem cells and conducting the procedure fully on an outpatient basis. **Results:** There were 39 patients with multiple myeloma, 31 with acute leukemia, 13 with Hodgkin's lymphoma, 7 with non-Hodgkin's lymphoma and 16 with other diagnosis. The median age was 47 years (range 8 – 78 years). Patients were stratified according to pretransplantation body mass index (BMI) values: 4 patients had low BMI, 45 had normal BMI and 57 patients had high BMI. Patients with an increased BMI had an 80-month OS of 65%, whereas those with a BMI between 18.5 and 25 kg/m², had an OS of 47% at 190 months. The small subset of 4 patients with a low BMI had an OS of 100% at 152 months. **Conclusions:** Our findings demonstrate a high prevalence of both overweight and obesity in Mexican patients undergoing an autologous HSCT and a correlation between pretransplantation BMI and posttransplantation survival; however, an increased body mass index was not found to be an adverse prognostic factor in autografting.

A1070

HOMOZYGOUS SICKLE CELL ANEMIA TRANSPLANTED WITH NON RELATED UMBILICAL CORD. Pérez-Lozano U, Ruiz- Ovalle JL. UMAE IMSS PUEBLA

Introduction: Sickle Cell Anemia (SCA) is a severe hereditary disorder that elicits complications putting life at risk at a short age. Umbilical cord blood transplant (UCB) is curative but it entails risks: graft rejection, post rejection medullar aplasia, relapse, graft versus host disease, etc. There are conditions that predispose to graft failure such as HLA mismatch, low unit cellularity, use of methotrexate, iron overload, splenomegaly. **Objective:** To

describe the evolution of a patient with SCA transplanted with two units of UCB. **Material and Methods:** Female patient from Oaxaca, single child, SCA heterozygous parents, presented anemia since she started walking and received transfusion therapy at 3 years of age, being diagnosed as SCA homozygote. **Results:** She was referred at 6 with a bone pain crisis and high transfusion requirements. She received hydroxyurea, improving requirements to one unit of packed red blood cells every two months to maintain hemoglobin of 7gr/dl, until she was 12 and entered an umbilical cord transplant protocol, with serum ferritin of 262ng/dl receiving oral iron chelation at 30 mg/kg, until achieving 880 ng/dl. A splenectomy was performed because of a grade IV splenomegaly 21 days before transplant. Pre-transplant conditioning with fludarabine 200mg/m² and cyclophosphamide 200 mg/kg. he two units of non related UCB were transplanted: the first one with a compatible HLA 5/6, female with a 0Rh+ ECLONE 38.1% hemotype, the second one 4/6 male product B Rh+ (no ECLONE was reported), 4/6 compatible between both units. With a total of 150 000 CD34+/kg, 4.5 x 10⁷ total nucleated cells/kg, weighing 40kg. Prophylaxis for graft versus host disease is with cyclosporine and oral Tacrolimus. The patient presented grade IV neutropenia according to the WHO for 8 days, anemia that was solved with transfusions up to day +76 post transplant. The patient also received antiviral, antifungal and P. Carini prophylaxis. There were no infectious or thrombotic events during the procedure. All blood products were infused radiated and with a leukocyte reduction filter. The patient required chelation for 3 months after transplant to maintain serum ferritin below 1000ng/dl. **RESULTS:** at 34 months follow up Hemoglobin is maintained at 10gr/dl without transfusion. HbS 64%, HbA1 33.9%, Hb A2 1.5%. ECOG 0. The patient has normal school activities. **Conclusions:** Transplant with UCB in SCA eliminates transfusion requirements and decreases hemolytic events even though a sickle cell trait persists. Oral chelation improves results and decreases complications.

A1076

INFLUENCE OF HLA GENES ON THE FREQUENCY OF ACUTE AND CHRONIC GRAFT-VERSUS-HOST DISEASE IN PAEDIATRIC PATIENTS UNDERTAKING HAEMATOPOIETIC STEM CELLS TRANSPLANTATION IN THE MEXICAN NATIONAL INSTITUTE OF PAEDIATRICS. Escutia-González A, López-Hernández G, Pérez-García ., Juárez-Nicolás F, Del Campo-Martínez A, Olaya-Vargas A. Instituto Nacional de Pediatría, Mexico DF.

Introduction: The compatibility of the HLA antigens during haemopoietic stem cells transplantation determines differences in the clinical result of this process. HLA incompatibility between the graft and the recipient (host) represent a risk of rejection and may avoid the success of the transplant, and the development of a clinical entity called Graft-versus-host disease (GVHD). Immunocompetent T cells from the donor, included within the graft may cause such disturbance.

Objectives: To describe the HLA class I and II antigens involved in the development of graft-versus host disease in patients undergoing haematopoietic stem cells transplant in the INP. **Material and Methods:** We included all the patients that received an allogeneic transplant of stem haematopoietic cells in our institution, and the risk factors to develop either acute or chronic GVHD, were evaluated. Genes from the HLA loci of both donors and recipients were characterized by PCR-SSP typing (median resolution). **Results:** 74 patients were enrolled to this study, 22 developed GVHD (29.7.3%). Two patients showed GVHD in first degree (4.5%), 4 patients degree II (18.1%), 13 patients degree III (59%) and 3 patients degree IV (13.6%). Seventeen patients (22.9%) from the total, showed GVHD to some degree, 11 patients from them, have had an earlier GVHD episode, one patient, degree II (5.8%), 8 patients, degree III (47%) and one patient, degree IV (5.8%). The mean elapsed time to develop GVHD was 273 days (133 to 679 days). Four patients showed GVHD de novo and limited (23.5%); five patients showed de novo and widespread (29.4%), three patients quiescent limited (17.6%), one patient quiescent widespread (5.8%), one progressive limited (1.3%) and three progressive widespread (4.05%). Regarding the differences in the patterns of histocompatibility antigens, we found that 27 patients showed HLA-A differences; 32 patients showed them in the HLA-B locus and 13 in the HLA-DR locus. None of the loci alone was able to induce GVHD. **Conclusions:** The use of tissues containing large amounts of cells, as in the case of concentrated blood samples and umbilical cord samples, increase the risk of developing graft-versus-host disease. Single HLA locus differences between the donor and the recipient induced neither acute nor chronic GVHD. However, antigen differences in more than one locus induced significant degrees of GVHD.

A1091

USEFULNESS OF MICROSATELLITE TESTING IN BONE MARROW TRANSPLANT DAY +14 COMPARED WITH ERYTHROCYTE PHENOTYPE AS PREDICTORS OF ENGRAFTMENT AND GRAFT VERSUS HOST DISEASE. Alvarado Ibarra M, Solano Ricardi M, Trejo Gómora J. Servicio de Hematología Centro Médico Nacional "20 de Noviembre". ISSSTE.

Introduction: Chimerism testing, performed by various techniques, has proved very effective in establishing the success of the transplanted allograft. Recently, some authors have reported that the performance of test day +14 chimerism has similar characteristics to the test performed at +28 days, thus allowing for a successful transplant earlier. **Objectives:** To determine the effectiveness of microsatellite chimerism test day +14 compared with erythrocyte phenotype to establish the success of allogeneic transplantation and relationship with graft versus host disease. **Material and Methods:** We included all patients over 15 years based on the underlying pathology underwent allogeneic bone marrow January 2010 to June 2011. We took them blood sample

day +14 and +28 day of transplantation to determine the degree of chimerism measured by erythrocyte phenotype and microsatellite and correlates with the outcome, as well as graft versus host disease (GVHD). Mixed chimerism was defined as the presence of 5-95% of the donor DNA sequences, complete chimerism the presence of +96%. **Results:** 18 patients were included, the average age was 44 years (14-55), 11 men and 7 women, bone marrow graft was found microsatellite day +14 in 88% and 55% erythrocyte phenotype $p = 0.2$. 16 patients with microsatellite chimerism (6 complete and 10 mixed) and 10 for erythrocyte phenotype (7 complete and 3 mixed) $p = 0.01$; microsatellite day +28 in 94% and 83% erythrocyte phenotype $p = 0.8$, by day +28 microsatellites were: 17 patients (2 complete and 15 mixed) and erythrocyte phenotype 15 patients (6 complete and 9 mixed) $p = 0.09$. +14 Day GVHD occurred in 13 patients with some degree of chimerism and chimerism 1 without, however in the measurement of the day + 28 of 14 patients with GVHD had chimerism, which means that the patient with GVHD and no chimerism was false negative. The evaluation of these days with erythrocyte phenotype were two false negatives for GVHD +14 day. **Conclusions:** Microsatellite comparison test is more effective than the erythrocyte phenotype in the day + 14 to determine the presence of graft, however does not preclude the use of day +28 and the graft is directly related to GVHD.

A1092

NON-MYELOABLATIVE-IMMUNOSUPPRESSIVE OUTPATIENT AUTOLOGOUS STEM CELL TRANSPLANTATION IMPROVES PATIENTS WITH TYPE 1 DIABETES. A PRELIMINARY REPORT. Cantú-Rodríguez O, Lavalle-González F, Gutiérrez-Aguirre C, Flores-Jiménez J, Treviño-Montemayor O, Sánchez-Cárdenas M, González-Llano O, Mancias-Guerra C, Zapata A, Zacarias-Villarreal J, Gómez-Almaguer D. Servicio de Hematología del Hospital Universitario "Dr. José Eleuterio González", UANL, Monterrey, Nuevo León, México.

Introduction: In recent years a theory of a possible "reset" of the immune system using autologous hematopoietic stem cell transplantation (ASCT) has emerged. Therefore and because of the pathogenesis, patients with newly diagnosis of type 1 diabetes mellitus (DM-1) may benefit with this therapy. **Objectives:** Show the effectiveness of a non-myeloablative autologous stem-cell

immunosuppressive outpatient transplantation in patients with type 1 diabetes. **Material and Methods:** A Prospective clinical study, in which 8 patients with newly diagnosed DM-1, until now, have been included. The diagnosis of DM-1 was confirmed by measurement of anti-GAD antibodies and pancreatic reserve by means of C-peptide levels. The ASCT using RIC was performed on an outpatient basis. A two-day cyclophosphamide (Cy) 1.5g/m²/day + mesna 300mg/m²/day as well as G-CSF (10mcg/kg/day x 6 days) was used for mobilization. The conditioning regimen included Cy (500mg/m²/day x 4) and fludarabine (30mg/m²/day x 4) from day -4 to -1. **Results:** Of the 8 cases, all were transplanted but until now only six completed the full evaluation. There were 5 male and 3 female, median age was 12.5 years (4-18), and the median time from the diagnosis of DM-1 to the transplant is 3 months (0-9). During the 1-18 months follow-up (median 12), 3 are currently free from insulin use and the remaining three have decreased their use by 70 and 40%, and 0% on one case; the mean amount of CD34 + cells infused is in 8.47x10⁶ (2.57-19.3 x 10⁶). In 7 patients only one apheresis procedure was needed, the other patient required two. The ASCT was completed 100% as outpatient procedure. **Conclusions:** After 12 months follow-up, 5/6 patients had showed a decrease in the insulin requirements and none patient had developed any complication or needed hospitalization. This procedure seems to be effective, without relevant toxicity and also very low cost.

A1112

PREVALENCE OF METABOLIC SYNDROME AND ITS RELATIONSHIP WITH BMI IN PATIENTS WITH HEMATOLOGICAL DISEASES. Jaime-Pérez JC*, Martínez-De La Garza KS*, Herrera-Garza JL*, Cantú-Rodríguez OG*, Gutiérrez-Aguirre CH*, Gómez-Almaguer D*. *Hospital Universitario "Dr. José Eleuterio González", Universidad Autónoma de Nuevo León, Monterrey, Nuevo León, México.

Introduction: Due to new and better treatments the survival rate in patients with hematologic diseases has improved and long-term life-threatening complications are increasingly reported. Cancer survivors are more likely to be significantly overweight, have higher fasting plasma glucose and insulin levels, and a decreased serum high-density lipoprotein (HDL) cholesterol levels. There

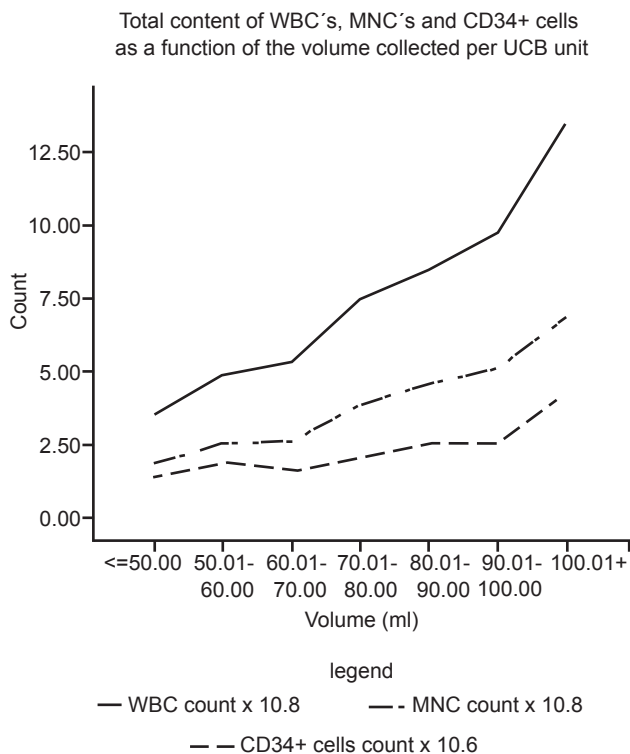
Age	Gender	Time from Diagnosis	CD34	Insulin Reduction 3m	Insulin Reduction 6m	Graft	Time Evol Days	Post ASCT months
4	M	9	8	0%	0%	14	534	18
18	M	2	8.3	65%	100%	14	529	18
9	F	8	8	15%	70%	14	511	17
12	M	0	15.1	80%	100%	14	322	11
13	F	3	3.78	20%	40%	14	266	9
9	M	3	19.3	70%	100% (4M)	13	133	4
15	M	3	2.57	15% (2M)	NA	14	61	2
13	F	4	2.76	25% (1M)	NA	13	42	1

is evidence to suggest that long-term cancer survivors may be at high risk for the premature development of characteristics associated with the metabolic syndrome (MS). **Objectives:** We determined the frequency of the MS and its relationship with the Body Mass Index (BMI). **Material and methods:** This prospective study evaluated the prevalence and risk factors for the MS in children and adults with hematologic diseases attending the Hematology Service of the “Dr. José E. Gonzalez” University Hospital in Monterrey, México from August to December 2011. Among 150 studied patients, 60 had a complete evaluation for MS and are the subjects of this report. Each eligible patient or his/her parents or legal guardian, if younger than 18 years of age at the time of interview, gave informed consent for the study. Metabolic Syndrome was defined according to the NCEP-ATP III revised in 2005. Patients were considered to have MS when they met at least 3 of 5 criteria (Table 1). Blood pressure was measured using a manual sphygmomanometer. Abdominal circumference was determined at midway between the iliac crest and the last rib employing a measuring tape. Blood samples for fasting glucose, triglycerides, and HDL cholesterol were obtained after overnight fasting. Body mass index was defined as the weight in kilograms divided by the square of the height in meters (kg/m²). WHO classifies the BMI in: underweight (<18.5), normal range (18.50-24.99), overweight (=25.00) and obese (=30.00). **Results:** Participants were more frequently males (56.7%); the median of age was 42 (range 17:78); Non-Hodgkin Lymphoma was the most common diagnosis n=20 (33.3%) (Fig. 1), the median weight and height were 74.6 kg and 1.66 m. respectively, with a median body mass index of 27.35. Metabolic syndrome was present in 31 of the 60 study subjects (51.7%) (Fig. 2), 51.6% had a body mass index =30 (Fig.3). **Conclusions:** Hematologists and other physicians who provide primary care to adult survivors

Table 1. Diagnostic Criteria for Metabolic Syndrome

Measure (any 3 of 5 criteria constitute diagnosis of metabolic syndrome)	Categorical Cut Points
Elevated waist circumference	≥102 cm in men ≥88 cm in women
Elevated Triglycerides (TG)	≥150 mg/dL or Drug treatment for elevated TG
Reduced High-Density Lipoprotein (HDL-C)	<40 mg/dL in men <50 mg/dL in women or Drug treatment for reduced HDL-C
Elevated Blood Pressure (BP)	≥130 mmHg systolic BP or ≥85 mmHg diastolic BP or Drug treatment for hypertension
Elevated fasting glucose	≥100 mg/dL or Drug treatment for elevated glucose

of cancer should be aware of the potential development of MS with its cardiovascular and diabetes risk as a consequence of treatment for hematologic diseases.



A1113

RESULTS OF A CORD BLOOD BANK PROGRAM AT A UNIVERSITY CENTER IN NORTHEAST MEXICO. Jaime-Pérez JC*, Colunga-Pedraza JE*, Monreal-Robles R*, Mancias-Guerra C*, Gómez-Almaguer D*. *Hematology Department, Internal Medicine Division, “Dr. José Eleuterio González” University Hospital of the School of Medicine of the Universidad Autónoma de Nuevo León, Monterrey, México.

Introduction: Umbilical cord blood (UCB) represents an alternative source of stem cells for autologous and allogeneic transplantation for the treatment of hematologic malignancies and genetic disorders. There is a lack information regarding Cord Blood Bank (CBB) collection and transplantation activities from developing countries. Our CBB was founded despite the many challenges to acquire the technology needed to process and cryopreserve UCB in order to offer our patients access to this important treatment option. **Objectives:** To document the CBB experience on collecting, processing and cryopreservation at a public university hospital in Northeast Mexico. **Material and Methods:** We carried out a retrospective, descriptive analysis of our CBB activity during the period from May 2002 to September 2010. Written informed consent was obtained from

healthy women with an uncomplicated pregnancy at term who volunteered to donate their UCB. Collection, processing and cryopreservation of CB were carried out following standard operating guidelines. The minimum volume and total nucleated cell content (TNC) for cryopreservation were 80 ml and 8.0×10^8 , respectively. **Results:** During the study period 1256 UCB units were collected; 428 (34%) were banked and 828 (66%) discarded. The main reasons for exclusion were a low volume and/or a low number of TNC, both accounted for 84% of the total discarded units. Cryopreserved cord blood units (CBUs) had a median volume of 113.0 mL and 13.0×10^8 TNC. Cell viability was 99.0%. During this period 15 units have been released for grafting. **Conclusions:** CBB demands significant human and financial resources, it is then essential for centers at developing countries to divulge their experience and results to increase the probability of finding matching units for their patients. UCB is an expensive resource acquired abroad from a certified CBB, and most of our patients cannot afford it. Establishing regional CBB like ours can make UCB allogeneic transplantation a viable therapeutic option for a greater number of patients living in non-industrialized nations.

Table 2. Reasons for rejecting UCB and their relative incidences

	n(%)
Volume <80 mL	498 (60.1)
Nucleated cells < 8.0×10^8	199 (24.1)
Positive donor serology	50(6.1)
Hold-up > 24 h for processing	28(3.4)
Clotted simple	7(0.8)
Problems during collection	6(0.7)
Other	40(4.8)

A1116

13 YEARS OF EXPERIENCE IN HEMATOPOIETIC STEM CELL TRANSPLANTATION IN THE INSTITUTO NACIONAL DE CIENCIAS MÉDICAS Y NUTRICION (INCMNSZ). Leon-Rodriguez E, Perez Alvarez SI, Guzman-Uribe P, Tuna Aguilar E, Espinosa Butista KA, Castro Sanchez A, Armengol Alonso A. Departamento de Hematología-Oncología. Instituto Nacional de Ciencias Médicas y Nutrición. México D.F.

Introduction: Hematopoietic stem cell transplantation (HSCT) is currently the treatment of choice and the only curative option for several malignant and nonmalignant hematologic diseases. In our country there are few centers specialized in this treatment, so it is important to know their experience. **Objectives:** The purpose of the present study was to describe demographic, clinical characteristics and outcome of patients undergoing HSCT in the INCMNSZ. **Material and Methods:** A prospective analysis of patients undergoing HSCT at the INCMNSZ was performed between 1999 and 2011. Epidemiological, clinical characteristics,

transplant related mortality (TRM) and survival were described. Survival analyses were assessed by the Kaplan-Meier method.

Results: We analyzed 160 patients undergoing HSCT. Median age was 30.5 years (range 15-65) and 108 (68%) were male. One-hundred patients (62.5%) underwent autologous HSCT: Germ cell tumor (GCT) 30 patients (30%), Acute Myeloid Leukemia (AML) 18 (18%), Non Hodgkin Lymphoma (NHL) 21 (21%), Hodgkin Disease (HD) 15 (15%), Multiple Myeloma (MM) 11 (7%), Acute Lymphoblastic Leukemia (ALL) 1 (1%) and others 4 (4%). Sixty (37.5%) underwent allo-HSCT (57 from sibling donor, 2 from umbilical cord blood, and 1 from a syngeneic origin): AML 6 patients (10%), Aplastic Anemia (AA) 18 (30%), Myelodysplastic Syndrome (MDS) 13 (22%), Chronic Myeloid Leukemia (CML) 10 (17%), ALL 8 (13%) and other 5 (8%). The overall 5 year survival for the entire group was 68%; for the autoHSCT group was 66% (GCT 67%, AML 59%, NHL 66%, HD 70% and MM 40%) and for the alloHSCT group was 71% (AML 55%, AA 72%, MDS 84%, CML 60% and ALL 70%). TRM for the entire group was 6.25%, for the autoHSCT group was 2% and for the alloHSCT 10%. Twenty patients (12.5%) underwent a second HSCT, being relapse the most frequent indication (40%). For these patients, the TRM was 20% and 5-year overall survival 55%. The average cost was US \$25,000 for autoHSCT and US\$ 44,500 for alloHSCT. **Conclusions:** These results show that HSCT is a feasible procedure in our country, with outcomes similar to those reported in international centers but significantly less expensive than in developed countries.

A1162

A NEW METHOD FOR THAWING HEMATOPOIETIC STEM CELLS AND ITS IMPACT IN THE RECOVERY OF THE HEMATOLOGICAL PATIENT. A PRELIMINARY REPORT. Mancías-Guerra C, Valdés-Burnes SL, Canales-Ortiz M, González-Llano O, Sosa-Cortez AC, Gómez-Almaguer D, Gutiérrez-Aguirre CH, Cantú-Rodríguez OG, Tarín-Arzaga LC. Servicio de Hematología del Hospital Universitario Dr. José E. González. Facultad de Medicina, U.A.N.L

Introduction: Clinical evidence indicates that cryopreserved hematopoietic stem cells (HSCs) can be a useful source for bone marrow reconstitution. There are several thawing methods for HSCs. All of them intend to avoid cell death and patient's side effects due to the dimethyl sulfoxide (DMSO) used as cryoprotector, at the time of infusion. We propose a new thawing method that can help diminish cell death and therefore a more rapid hematological recovery. **Objectives:** Modify the thawing method to obtain a better cell recovery and, therefore an improvement in the time of engraftment. **Material and Methods:** A standard thawing, diluting and removing the DMSO method was described by Rubinstein in 1995 (method 1). In our method we are increasing more than 10 fold the dilution (method 2) of cryopreserved HSCs in the standard washing solution (5% albumin + dextran 40) to try to decrease the amount of total nucleated cells (TNC)

that will die during the thawing procedure. Compare methods 1 and 2 to determine which is better as a thawing method for HSCs by means of the TNC count and viability by trypan blue and flow cytometry in each unit of HSCs at the time of collection, cryopreservation, thawing and, finally after removing the DMSO before its infusion, as well recording the patient's day of engraftment. **Results:** We evaluated 18 patients, nine with the traditional method (1), and nine with the new method (2). The mean of cell death was 26.44% (sd 11.304) in method 1, and 14.20% (sd 9.928) in method 2. (P= 0.027). The mean time of neutrophils' engraftment (>500 neutrophils/microliter) was 21 days (sd 10.150) in method 1, and 15 days (sd 8.492) in method 2 (P= 0.163). The mean time of platelets' engraftment (>20000 platelets/microliter) was 12 days (sd 5.745) in method 1, and 11 days (sd 3.833) in method 2 (P= 0.894). **Conclusions:** A greater dilution of the washing solution (5% albumin + dextran 40) as a new method of thawing cryopreserved HSCs may decrease the amount of cellular death, which will be reflected in a better viability, measured by flow cytometry, or a greater number of HSCs infused to the patient, decreasing as well the time of engraftment.

A1163

QUICKLY EVOLUTION OF MULTIPLE MYELOMA POST-AUTO TRANSPLANTATION TO MYELODISPLASTIC SYNDROME TO ERYTOLEUKEMIA , A CASE REPORT. Tuna-Aguilar E, Galván-López I, Couary-Aguilera P, Deffis-Court M, Morales-Juárez L, Amador-Medina F. Instituto Nacional de Ciencias Médicas y Nutrición Salvador Zubirán, México D.F, México.

Introduction: Secondary MDS describes the development of MDS or acute leukemia 10-15 years after known exposure to sources of chromosomal damage. Patients who survive a cancer treatment have a high risk. We describes a case of quickly evolution from auto-HSCT for multiple myeloma to MSD to erytroleukemia.

Objectives: 1. To report an unusual case of quickly clonal evolution from a normal karyotype after autologous-HSCT. 2. Analyze the case with the existing statistical data for secondary MDS. 3. Exhibit how fast the myelodysplastic syndrome can evolve after diagnosis. **Material and Methods:** We followed the patient prospectively and reviewed retrospectively their previous history and laboratory studies. **Results:** A 53 year old woman was diagnosed of multiple myeloma IgA lambda ISS-1,DS-IIA in August 2006. Karyotype:46XX. FISH negative . Chemotherapy started in September 2006, with the following schemes: Thalidomide/Dexamethasone/A.Zoledronic; Bortezomib (13) and DCEP (dexamethasone, cyclophosphamide, etoposide and cisplatin)(3); partial response was achieved, in June 2008, FISH:RB1,D13S319 and IGH+,D13q4,+13 q14.3,14q32+. In July 2008 autologous-HSCT was performed, in complete response since then. In September 2008 presented persistent pancytopenia, a bone marrow aspiration (BMA) reported: Myelodysplastic Syndrome Refractory Cytopenia with Multilineage Dysplasia(IPSS-1.5); karyotype: 18 = 46, XXX,t(1, 5, 9, 20),-5,+der(9), 2 = 46, XX. In January 2011 She

start a treatment with azacytidine (5) and transfusional therapy . The BMA 06/23/2011 unchanged ; karyotype:8 = 46, XX t(1, 5, 9, 20), -5, +der (9). 5 = 92 1 = 207 XXXX, XXXXXXXXXX. 6 = 46, XX; RIC allogeneic SCT URD was performed in August 24, complete haematological recovery without GvHD. The first post HSCT chimerism showed 70% donor. 11/08/11 in the presence of cytopenias , BMA again showed significant dysplastic erythroid , karyotype 1 = 46, XX, t (1, 5, 9, 20), -5, + der (9), a = 69, XXX, t (1, 5, 9, 20), -5, + der (9), 4 = 92, XXXX, t (1, 5, 9, 20), -5, + der (9), 14 = 46, XX, , so immunosuppression was discontinued in order to rescue the grafted cells. However she presented a unexpected MDS progression to pure erythroid-leukemia with complete loss of chimerism and a karyotype with a additional SUBCLONAL population. She was performed a second SCT-RIC URD, currently in post-transplant period. **Conclusions:** 1. IPSS is a prognostic scale for MDS, some cases average out. 2. The MDS secondary to HSCT may be after 10 years, our patient presented it unexpectedly fast. 3. It's possible a predisposing situation in the development of clonal evolution.

A1164

INTRATHECAL AUTOLOGOUS TOTAL NUCLEATED CELLS FOR CHILDREN WITH BRAIN INJURY. Mancías-Guerra C, Marroquin-Escamilla AR, González-Llano O, Sosa-Cortez AC, Valdés-Burnes SL, Barrera-Morales DC, Gómez-Almaguer D, Elizondo-Riojas G, Gutiérrez-Aguirre CH, Cantú-Rodríguez OG, Tarín- Arzaga LC. Servicio de Hematología del Hospital Universitario Dr. José E. González. Facultad de Medicina, U.A.N.L.

Introduction: It has been found that autologous bone marrow (ABM) derived total nucleated cells (TNC) may be transported through the cerebrospinal fluid and be delivered more efficiently to the injured area after their introduction in the subarachnoid space of the spinal cord when compared with the intravenous (IV) route in children with hypoxic/isquemic (H/I) brain injury. **Objectives:** Evaluate safety and effectiveness of ABM derived TNC for the treatment of children with brain injury. **Material and Method:** Patients with H/I brain injury between 1 month and 8 years old were evaluated with the Battelle Developmental Inventory (BDI) and magnetic resonance images (MRI). Subcutaneous granulocyte-colony stimulating factor was used for 4 days prior to the ABM harvesting. The buffy coat was obtained through BM centrifugation and infused through lumbar puncture. **Results:** Eight male and 5 female patients (13) with a median age of 42 (7 - 112) months and a mean weight of 15.9 (SD 7.8) kg were included. Eight patients presented quadriplegia, 3 diplegia, and 2 paraplegia, while 7 presented hypertonia, 3 dystonia, and 3 hypotonia. A median of 160.8 (9.3 - 513.0) /microliter CD45+ cells, with 6.2 (1 - 29.9) x 106 CD34+ cells, in a volume of 8 (4 - 10.5) mL were infused intrathecally. One patient developed lingual edema and another one stridor, 1 had fever and headache and vomit in the first 36 hours after the procedure. Patients showed an increase in BDI at the 30-day assessment

from 172.07 (22 – 576) to 197.76 (27 – 629) in the global score ($P = .001$) and from 25.46 (1 – 62) to 18.3 (1 – 71) months in age equivalent in months ($P = .005$). At the 6-month follow-up, global score was 217.53 (49 – 637) and age equivalent was 20.15 (3 – 73) months, both statistically significant when compared against initial and 1-month results ($P < .001$ and $P = .001$). No changes were found in the MRI. **ConclusionS:** This procedure proved to be safe, with minimal side effects, and it seems to be beneficial in patients' development according to BDI domains. Longer follow-ups and more specific image studies are needed to increase this evidence. It will be necessary a phase II comparative clinical trial to consider this approach as a therapeutic option.

A1166

MOBILIZATION OF CD34+ CELLS IN AN HEMATOPOIETIC STEM CELL TRANSPLANTATION UNIT AT NORTHEAST MEXICO. Martínez-Hernández RA, Ceballos-López AA, Cuervo-Sierra J, Flores-Jiménez JA, Salazar-Riojas MR, Méndez-Ramírez N, Cepeda-Cepeda MG, Gómez-Almaguer D. Servicio de Hematología. Hospital Universitario "Dr. José Eleuterio Gonzalez" Universidad Autónoma de Nuevo León.

Introduction: Peripheral blood progenitor cells (PBPC) are increasingly used as main source for allogeneic transplantation. At the present study we intend to find some variables pre-collection of PBPC that predict if the patient will hold the minimal required dose of CD34+ cells. **Objectives:** To describe factors influencing the collection of CD34 cells and the characteristics of the PBPC cell transplantation donors. **Material and Methods:** Retrospective study of CD34+ cells apheresis from healthy donors between 2006-2011. Stimulation with filgrastim 10 mg/kg/day was administered for 4 days and apheresis was performed on day 5 using a Cobe Spectra equipment. CD34 count was performed by flow cytometry by the ISHAGE method in FACS Canto II BD equipment. CD34 count previous to apheresis procedure was not performed systematically. **Results:** We review 173 apheresis performed to 155 donors, 102 (65%) by central venous catheter (CVC). In 137 (88.3%) donors CD34 $> 3 \times 10^6$ were obtained in the first procedure, and 3 (1.9%) donors had less than 3×10^6 CD 34 after 3 procedures. Median age was 31 (1-70), weight 70 kg, height 164cms, median minutes of apheresis 210, median processed total volume 17,972 ml, median peripheral blood (PB) leukocytes 47700/mcL, median PB monocytes 17.4%, median CD34 6.4×10^6 / kg / weight of the donor, median CD34x10L 3.8×10^6 per apheresis. CD34/kg difference was observed between gender, male 7.4 vs female 5.1 ($p < 0.05$). Patients > 45 years (percentile 75) had a median of 3.6 vs 4.2×10^6 CD34 / L of apheresis in < 45 years ($p < 0.01$). Patients who had an apheresis $> 3 \times 10^6$ CD34/10L had a median peripheral blood leukocytes 51700/ mcL, vs 42500/mcL ($p < 0.001$), no correlation was observed between PB leukocytes and CD34/10L leukocytes from apheresis ($r = 0.13$), there was no difference in PB monocytes. Patients who had $> 3 \times 10^6$ CD34/kg weight had a median donor leukocytes of 49800/mcL vs PB 39950/mcL ($p < 0.01$), there was

no correlation ($r = 0.31$). Patients who had a total leukocyte count greater than the median PB (47,700/mcL) were more effective collectors, but no correlation was observed with CD34 final total. There was no difference in the results by comparing apheresis performed using CVC vs peripheral catheter. **Conclusions:** The method used in our center for healthy donors to apheresis is effective with few side effects in the donor. Consistent with other studies observed better collections are obtained in men and under 45 years. We see a better trend with higher PB leukocytes ($p < 0.01$) but no correlation with the total CD34 ($r = 0.13$).

A1168

IMMUNOLOGICAL RECONSTITUTION MONITORING AND CHARACTERIZATION IN PEDIATRIC PATIENTS AFTER HAPLOIDENTICAL TRANSPLANT OF HEMATOPOIETIC PROGENITOR CELLS. Mújica-Guzmán F, Pérez-García M, Mendoza-González F, Romero-López V, Romero-Guzmán L, Medina-Torres EA, Ortíz-Dominguez CP, Olaya-Vargas A. National Institute of Pediatric, México DF, México.

Introduction: Presently, the transplant of hematopoietic progenitor cells (THPC) is an alternative for patients without compatible donors that require a transplant in an opportune form with relative urgency. This gives to such patients an alternative in the absence of histocompatible donors. Evaluation of immunological reconstitution pattern in patients subjected to this type of transplant would permit the establishment of the time in which such patients are at a risk of infection and the presentation of an inherent complication that could put his or her life in danger. **Objectives:** The objective of this work was to describe immunological reconstitution pattern of pediatric patients who underwent THPC using haploidentical transplant. **Material and methods:** 6 cases of patients who subjected to haploidentical transplant of hematopoietic progenitor cells (HPC) were studied. The levels of lymphocyte subpopulations were determined by flow cytometry using surface monoclonal antibodies: CD3, CD4, CD8, CD19 and CD56 at 1, 3, 6, and 12 months of post transplant follow-up. **Results:** The analysis of lymphocyte subpopulation showed that at one year post-transplant, 67% of the cases presented complete immunological reconstitution. We observed that immunological reconstitution follows a certain pattern with NK cells being the first to reconstitute after one month, followed by cytotoxic T lymphocytes at 3 months, B lymphocytes at 6 months, and T lymphocytes cooperators at a year. **Conclusions:** Post-transplant immunological reconstitution monitor has an important prognostic value, since there is a correlation between patient's clinical state and lymphocytic subpopulation levels in the different months of follow-up.

A1174

BUCY2 IN THE CONDITIONING THERAPY OF ABMT FOR LYMPHOID MALIGNANCIES. Tripp F, Ayala M, García E,

Rodríguez G, Ramírez A, Barón C, Avila E, Vela-Ojeda J. UMAE “La Raza” IMSS, México D.F.

Introduction: High-dose chemotherapy followed by autologous stem cell rescue is the standard of care for patients with non-Hodgkin lymphoma (NHL) after failure or relapse of primary therapy. In the autologous setting BEAM is the most widely used conditioning regimen to treat lymphoid proliferative disorders, but in recent years the availability of BCNU (carmustine), which needs an alcoholic solvent to be infused intravenously, seems hampered by technical problems in its production. Although Lomustine (CCNU) another nitrosurea, available for oral administration, is the most logical drug to overcome these difficulties in our country and our institution is not available. Now the bifunctional DNA alkylating agent Busulfan (Bu) is now widely used as an alternative to in conditioning therapy for hematopoietic stem cell transplantation. Bu based preparative regimens are commonly used in allogeneic HSCT but have been studied less frequently in autologous HSCT (autoHSCT). The use of busulfan based conditioning is extending rapidly also to hematopoietic stem cell transplantation (HSCT) for lymphoid malignancies. **Objective:** Analyze the BuCy2 as part of conditioning regimen for lymphoid malignancies. **Material and Methods:** We retrospectively analyzed the outcomes of 16 patients over age 18 years (7 females and 9 males) who underwent ASCT for NHL or HL using the BuCy2 regimen at the Centro Medico Nacional “La Raza” from January 2009 through 31 January 2012. **Result:** In our series the overall median age was 36.5 years (range 17-61 years), 15 patients was in complete remission at transplantation, 1 patient had a partial response to previous therapies. The median number of CD34+ cells infused was $3.02 \times 10^6/\text{Kg}$ (range 1.82-8.9). The median time to neutrophil ($>500 \times 10^6/\text{L}$) and platelet ($>20 \times 10^9/\text{L}$) engraftment was 11 days (range 9-16) and 17.5 days (range 10-27) respectively. Granulocyte-colony stimulating factor (G-CSF) was given to 16 patients (100%). Oral mucositis was observed in 15 patients (100%), 3 grade III (18.75%) and patients grade IV (81.25%). 12 patients (75%) were supported with parenteral nutrition. 8 (50%) patients had grade I-II diarrhea. No grade III-IV diarrhea was observed. Fever of unknown origin (FUO) was observed in 16 (100%), in 4 patients (25%) the infectious agent was identified. At a median follow-up of 540 days (range 150- 1320days) the overall survival (OS) is 93.7% and disease free survival (DFS) is 87.5%. **Conclusion:** Our data seem demonstrate that Bucy2 can be safely used as part of conditioning for lymphoid malignancies.

A1186

PRELIMINARY RESULTS AFTER NON-MYELOABLATIVE AUTOLOGOUS HAEMOPOIETIC STEM CELL TRANSPLANTATION FOR RELAPSING-REMITTING MULTIPLE SCLEROSIS AND SECONDARY PROGRESSIVE MULTIPLE SCLEROSIS. Tripp F, Molina E, Ayala M, García E, Rodríguez G, Ramírez A, Barón C, Avila E, Vela-Ojeda J. UMAE “La Raza” IMSS, México D.F.

Introduction: Autologous non-myeloablative haemopoietic stem cell transplantation is a method to deliver intense immune suppression. We evaluated the safety and clinical outcome of autologous non-myeloablative haemopoietic stem cell transplantation in patients with relapsing-remitting multiple sclerosis (MS) who had not responded to treatment with interferon beta, Mitoxantrone Glatiramer acetate, prednisone or methylprednisolone. **Objectives:** Progression-free survival will be the primary outcome. Secondary outcomes include interval changes in MS-specific imaging, time to progression, overall survival, causes of death, response to any MS-specific treatment required posttransplantation, and incidence of transplantation related late effects and secondary malignancies. **Material and Methods:** Eligible patients had relapsing-remitting MS, attended in UMAE La Raza, and despite treatment with interferon beta had had two corticosteroid-treated relapses within the previous 12 months, or one relapse and gadolinium-enhancing lesions seen on MRI and separate from the relapse. Peripheral blood haemopoietic stem cells were mobilized with 4 g per m² cyclophosphamide and 10 microg per kg per day filgrastim and Prednisone. The conditioning regimen for the haemopoietic stem cells was . The conditioning regimen was Rabbit Anti-Thymocyte globulin 3mg/kg for 4 day and cyclophosphamide 60mg/kg for two days. Primary outcomes were progression-free survival and reversal of neurological disability post-transplantation. We also sought to investigate the safety and tolerability of autologous non-myeloablative haemopoietic stem cell transplantation. **Results:** Between March 2011 and November 2011, 5 patients were treated. Engraftment of white blood cells and platelets was on median day 9 (range day 8-10) and day 14(range day 13-15) respectively None patient has complication. The median number of CD34+ cells infused was $19 \times 10^6/\text{Kg}$ (range 4.5-45). At a median follow-up of 150 days (range 90- 329days) No patient has relapsed, the median expanded disability status scale (EDSS) before transplantation was 6.0 and after transplantation the median of EDSS was 5.5. **Conclusions:** Non-myeloablative autologous haemopoietic stem cell transplantation in patients with relapsing-remitting and Secondary Progressive MS reverses neurological deficits, but these results need to be confirmed in a randomized trial

A1193

QUALITY CONTROL UNITS OF CORD BLOOD FOR AUTOLOGOUS TRANSPLANTATION. Hernández-Nevárez JA*, Sosa-Hernández NA*, Martínez-De la Cruz B*, Silva-Martínez JF*, Ramírez-Martínez JS*, Pérez-Codero CL*, Santiago-Osorio E**, Pier-D*, Vázquez-Meráz JE***. *Banco de Cordón Umbilical, Edo. de México, México; **Universidad Nacional Autónoma de México, México DF, México; ***Hospital General de Ecatepec Las Americas, Edo. de México, México.

Introduction: In Mexico, the cryopreservation of umbilical cord blood (UCB) for therapeutic purposes requires implementing the strictest quality controls as clonogenic assays to ensure the safety

and efficiency of the units to be transplanted. **Objectives:** Assess the ability of differentiation post-thawed of cryopreserved units of UBC as a method of validation and quality control pre-transplant for clonogenic assay. **Material and method:** Morphology studies were carried out of cells and CFU, quantification of CD34 + TNC, MNC, viability and microbiological studies from pilots thawed at room temperature of 12 UCB units cryopreserved in 2005 in BCU. These units were randomly selected and evaluated for the same parameters in 2010. For the CFU assay, cells were washed with IMDM with 2% FBS and cultured (1x10⁵ cells / ml) in methylcellulose semisolid medium with 12 units whit characteristic similar not cryopreserved (positive control). After 14 days of cultivation (37 ° C and 5% CO₂) they were quantified colonies (1colonia= 20 cells) and identified morphologically by Wright staining under an inverted microscope. All assays were performed in triplicate. Statistical analysis was performed using the Student-t test (p <0.05) with SPSS version 17.0. **Result:** Our results indicate post-thaw mean values of 7.22x10⁸ (TNC), 92.31% (viability), 65.02% morphology (blasts), 19.59% (monocytes), 15.39% (granulocytes), 2.81x10⁶ (CD34 +) and 6.9% (e-clone), which corresponds to a greater proportion of the erythroid lineage (60.3%), followed by the myeloid (39.2%) and finally the multilineage (9.5%). Interestingly, our results showed no significant differences with those reported prior to cryopreservation. **Conclusions:** This indicates that the process of cryopreservation, as well as, the thawing did not affect the integrity of the units, which, can be attributed to those units that met all inclusion criteria pre-and post-processing. It is also to highlight the implementation of UFC assays as an indicator of in vitro functionality of our units, which, together with CNT counting is a reliable quality control and reproducible. Thus, Umbilical Cord Bank shows the quality of cryopreserved units through the monitoring of their procedures comply with good manufacturing practices and national and international standards established.

A1199

AUTOLOGOUS BONE MARROW TNCs FOR THE TREATMENT OF KNEE OSTEOARTHRITIS - A PRELIMINARY REPORT. Mancias-Guerra C, González-Llano O, Villarreal-Martínez L, Marfil-Rivera J, Sosa-Cortez AC, Valdés-Burnes SL, Tarín-Arzaga LC, Cantú-Rodríguez OG, Gutiérrez-Aguirre CH, Garay-Mendoza D, Acosta-Olivo C, Díaz-Hutchinson C, Gómez-Almaguer D. Servicio de Hematología y Servicio de Traumatología y Ortopedia del Hospital Universitario Dr. José E. González. Facultad de Medicina, U.A.N.L.

Introduction: Hematopoietic stem cells (HSCs) have been studied to treat joint injuries with promising results. Osteoarthritis (OA) leads to severe disability, related to cartilage and subchondral bone degradation and osteophyte formation. Some patients will face progression even ending in joint replacement surgery. We describe a trial using bone marrow total nucleated cells (TNCs) as an alternative treatment for OA. **Objective:** Evaluate safety and effectiveness of autologous bone marrow

derived HSCs in OA treatment. **Material and methods:** Patients with X-Ray confirmed degenerative joint disease that met the inclusion criteria were recruited. After signing the informed consent, they were stimulated with subcutaneous granulocyte-colony stimulation factor for 3 consecutive days (10ug/kg/day). One hundred milliliters of autologous bone marrow was harvest on the 4th day (100ml) from posterior iliac crests. We obtained the buffy coat from centrifugation in our laboratory. The TNCs were infiltrated to the knee joint under local anesthesia on an ambulatory setting. Follow-ups are performed at 1 and 6 months with specialized scales and then compared to the initial one. **Results:** We included 13 patients, 9 females and 4 males. The median age was 61 (32-76) years. One patient presented grade 1 OA, four patients grade 2, seven patients grade 3, while one patient grade 4 OA. A volume of 10mL of stem cells were infiltrated to the patient's knee, with a mean of 19.12 CD34+ X 10⁶ cells (SD=7.68468), and 3.27 CD45+ X 10⁵ per microliter of cells (SD 0.847). Eleven patients showed significant pain improvement as well as disappearance of the claudication before the end of the first month of the follow up. One patient did not present any improvement, while one patient quit the trial, with any acute side effect. A longer follow up is needed before any radiologic or scale change. **Conclusions:** So far, this treatment has demonstrated that it is a safe, feasible option, with minor side effects, ameliorating in a large degree the symptoms, as referred by the patient. Additional experimental support is needed to prove the capacity of this treatment to relieve symptoms and possibly delay or avoid OA's complications.

A1205

HEMATOPOIETIC STEM CELL TRANSPLANTATION (HSCT) WITH REDUCED INTENSITY REGIMENS - UNIVERSITY OF CHIHUAHUA CENTRAL HOSPITAL EXPERIENCE Rivera-Olivas J, Acosta-Peña C, Martínez S. Hematology Department at University of Chihuahua Hospital

Introduction: The Hematopoietic Stem Cell Transplantation is today an important therapeutic approach not only for different malignancies that require myeloablative doses of radiation and/or chemotherapy, but also many other diseases where the production of any of the lineages is defective. Although it is a complex process, now has reduced morbidity with new preparative regimens for transplantation and other adjunctive treatments, as we see in this abstracts. **Objective:** Report the results of the HSCT program conducted at the Hospital of the Autonomous University of Chihuahua. **Material and Methods:** In a period of 2.7 years, from May 2009 to January 2012 there have been eight HSCT, six autologous and two allogeneic June. Both donors and patients recruitment was employed Filgrastim 5-10 mU / kg / day for 5 days, the collection is done on a Haemonetics MCS in 9000 machine. The conditioning regimen was according to diagnosis: The autologous transplants for patients with multiple myeloma received high-dose melphalan, in the Leukemia and Lymphoma patients was cyclophosphamide and etoposide, in

cases of allogeneic transplantation was used cyclophosphamide, fludarabine and busulfan. GVHD prophylaxis was with cyclosporine and methotrexate. **Results:** The age of patients was 26 to 51 years (average 40), 6 female and 2 male. In all cases we use the collection of peripheral blood hematopoietic progenitor cells. The diagnoses were in Autologous 3 with NHL, 2 with multiple myeloma and 1 with acute lymphoblastic leukemia; in relation to Allogeneic, 1 patient with AML and one with Glanzmann thrombasthenia. In both cases, the cell histocompatibility was with HLA-identical siblings went to 100%. Not in all cases, CD34 was able to count, only 5 with an average check of 5.4×10^6 (6), in the other 3 was mononuclear cell count average of 8.6×10^8 (8). The engraftment time for neutrophils was 11.3 days and 13.87 days for platelets. The mortality attributed to transplantation and 100 days was 0%. Major complications in autotransplantation were anal fissure in 2/6 cases, mucositis in 5 / 6, CHF 1 / 6, Sepsis 1 / 6. In patients with allogeneic transplant GVHD grade 1 was presented in 1 / 2 patients. The survival of patients is: 3 of 6 Autologous die, one of miliary TB and 2 activity, ALL and the other MM. The two Allogeneic patients are alive. **Conclusions:** There are few transplanted patients in our hospital, however we believe that the program is operated in accordance with national and international statistics.

A1212

ALLOGENEIC HEMATOPOIETIC STEM CELL, TREATMENT OF GLANZMANN THROMBASTHENIA - A CASE REPORT. Rivera-Olivas J, Acosta-Peña A, Martinez S. Hospital Central Universitario de la Universidad Autónoma de Chihuahua, México.

Introduction: Glanzmann thrombasthenia is a rare autosomal recessive bleeding disorder affecting the platelets function. It is characterized by normal platelet count and morphology, prolonged bleeding time, absent or diminished clot retraction and defective platelet aggregation. The clinical course of the disease varies from mild mucocutaneous bleeding episodes to severe life threatening bleeding. The disease severity may be unpredictable. **Objective:** Report of a case of Glanzmann thrombasthenia treated with Allogeneic Stem Cell. **Material and Methods:** This is a female aged 49 years old, diagnosed with Glanzmann thrombasthenia 3 years ago, with a history of an uncle killed by bleeding "uncontrollable" without specific diagnosis. The patient in the last 3 years had mucocutaneous bleeding in several places and with different degrees of intensity, especially large spontaneous bruising, difficult to control, and it was necessary transfusion of plateletpheresis, initially once every 2 months, but transfusion requirements to be made more frequently in the last months of 2010, and in the early months of 2011 came to require a transfusion of plateletpheresis each week. Suspected and confirmed the presence refractoriness to transfusions. For economic reasons could not administer recombinant Factor VII. Since her sister was 100% match HLA compatible and no clinical or laboratory Glanzmann thrombasthenia had

decide to do an Allogeneic Hematopoietic Progenitor Cells as the best treatment option. The conditioning regimen was with cyclophosphamide 350 mg/m², 30 mg/m² both fludarabine for 3 days and busulfan 4 mg / kg / day. For prophylaxis of graft versus host disease (GVHD) is used methotrexate (MTX) and cyclosporine (CSP). The CSP is started on day -1 continuously and adjusted to maintain plasma levels between 100 and 150 ng / L and the MTX is administered on days +1, +3, +5 and +10. Receive 2.69×10^6 (6) of CD34 + cells collected by apheresis 16280 ml in 10 hours. **Results:** Neutrophils grafting was observed at day +11 and day +13 platelets. The day +87 GVHD presents data from a gut level with Stage I ulcers in oral mucosa, mild abdominal pain, diarrhea of <500 ml per day and yield by adding prednisone. Chimerism at day 30 was 100% donor hematopoiesis. When 105 days passed after transplantation (3 months) she did not required any platelet transfusion due she did not bleed anymore. **Conclusions:** The Allogeneic Hematopoietic Stem Cells for patients with severe Glanzmann thrombasthenia is a good alternative treatment showing refractoriness to platelet transfusion and progressive clinical course.

A1227

CORD BLOOD BANKING AND TRANSPLANTATION IN THE FIRST PUBLIC BANK CREATED IN MEXICO. Mancias-Guerra C, Valdés-Burnes SL, González-Llano O, Jaime-Pérez JC, Cantú-Rodríguez OG, Gutiérrez-Aguirre CH, Tarín-Arzaga LC, Gómez-Almaguer D. Servicio de Hematología del Hospital Universitario Dr. José E. González. Facultad de Medicina, U.A.N.L.

Introduction: The Blood Bank (BB) at the "Servicio de Hematología" (Hematology Service) of the University Hospital of Monterrey initiated activities in May 2002. The Mexican Ministry of Health extended to our BB the first permit in Mexico to operate as a Cord Blood Bank (CBB), being therefore the first public CBB in our country. **Objectives:** We describe the experience generated during this period, in view of its 10th anniversary, next May. **Material and Methods:** International standard operating procedures have been used for donor selection, collection, processing and cryopreservation of umbilical cord blood (UCB) units. UCB units were selected, thawed, processed, and released for transplantation based on its HLA (from 4/6 to 6/6 matches), total nucleated cell and CD34+ cell content. Furthermore, if the unit search was unsuccessful, we conducted a search in the second public CBB "Centro Nacional de la Transfusión Sanguínea (CNTS)" and in some United States CBB, the National Cord Blood Program in the New York Blood Center (NYBC), StemCyte International Cord Blood Center, University of Colorado Cord Blood Bank (UCCBB) and in the National Marrow Donor Program (NMDP). **Results:** Nowadays 265 UCB units are stored in the "Servicio de Hematología" CBB. Ten of these units (3.77% of the stored units) have been released for transplantation. We have performed 277 searches in our CBB, 99 searches in the CNTS, located in Mexico City, 73 in the UCCBB, 77 in the NYBC, 53 in StemCyte, 22 in the NMDP, and 2 in the Caitlin Raymond International Registry. Thirty nine

percent of these patients corresponded to patients with acute lymphoblastic leukemia, 15% with acute myeloblastic leukemia, 8% with chronic myeloblastic leukemia, 7% with aplastic anemia, 7% with immunodeficiencies, 2% with myelodysplastic syndrome and 22% with other hematological and non hematological disorders. Due to financial restrictions less than half of the patients have the opportunity to be transplanted with a foreign unit(s) other from our CBB: 15 UCB transplants with units from the CNTS, 14 with units from the NYBC, 8 from the StemCyte, 8 from the UCCBB, and 5 from the NMDP. **Conclusions:** Although the experience presented is still limited and the period of analysis is still short, the results obtained during these years are encouraging, while we are the only CBB that offers the service of importing UCB units and even unrelated donors in all Mexico.

A1231

CD133+CD34+ AND CD133+CD38+ BLOOD PROGENITOR CELLS AS PREDICTORS OF PLATELET ENGRAFTMENT IN PATIENTS UNDERGOING AUTOLOGOUS PERIPHERAL BLOOD STEM CELL TRANSPLANTATION. Camacho-Villa AY*, Reyes-Maldonado E*, Montiel Cervantes LA***, Vela-Ojeda***. *Departamento de Morfología, Escuela Nacional de Ciencias Biológicas, Instituto Politécnico Nacional, México, DF; **Servicio de Hematología, Hospital de Especialidades "Antonio Fraga Mouret", Centro Médico Nacional "La Raza", IMSS, México, DF.

Introduction: Human hematopoietic stem cells (HSC) have been characterized by CD34+ antigen expression, and an adequate dose of CD34+ cells is associated with rapid, complete and sustained engraftment. Most cells expressing CD133+ also co-express the CD34+ antigen. CD133+ is a more specific marker of HSC than

CD34+. CD133+CD34-CD38-Lin- cells have high clonogenic potential. However, discrepancy is still frequent between the number of CD34+ cells reinfused and engraftment. **Objectives:** To better define which stem cell subset may actually predict a better engraftment, a prospective study was conducted on the relationship between graft content of CD34+ and CD133+ cells and their CD38+ subsets and trilineage engraftment in patients with different hematological diseases. **Material and Methods:** We prospectively studied the relationship between graft content of CD34+ and CD133+ cells and their CD38 subsets, and trilineage engraftment after autologous stem cells transplantation (auto-SCT). A prospective study was conducted on 24 patients with different hematological disorders – non-Hodgkin's lymphoma (NHL) 17, Hodgkin's lymphoma (HL) 5 and multiple myeloma (MM) 2. Samples were obtained before and after mobilization with recombinant granulocyte-colony stimulating factor (G-CSF, ug/kg), from apheresis collection bags and after transplantation. **Results:** Cell populations derived from CD34+ and CD133+ HSC were quantified by flow cytometry, and the dose of each population infused was correlated with speed and success of engraftment of the granulocytic, platelet and erythrocytic series. G-CSF induced mobilization of CD133+CD38+ cells (12.6-fold) and CD133+CD34+ cells (14.7-fold). The median dose of CD133+CD38+ cells infused was 0.50 x10⁶/kg and 0.51 x10⁶/kg for CD133+CD34+ cells. A correlation was observed between the infused dose of CD133+CD34+ and CD133+CD38+ cells and engraftment of the platelet series. **Conclusions:** CD133+CD34+ and CD133+CD38+ cells were mobilized with G-CSF and this was significantly correlated with success of engraftment of the platelet series, evidencing the clinical impact of these cells in hematopoietic recovery after autologous peripheral blood stem cell transplantation (auto-PBSCT).

TRANSFUSION MEDICINE

POSTERS

A1013

BLOOD DONATION AND PREVALENCE OF TRANSFUSION TRANSMITTED INFECTIONS IN AN ANGOLAN CLINIC IN LUANDA. Fernández Águila JD *, Baptista-Pocongo BL**, Sequeira-Pataca SA**, Fragoso M**, Rivero Jiménez RA***. *Dr. Gustavo Aldereguía Lima University Hospital, Cienfuegos, Cuba; **Multiperfil Clinic. Luanda, Angola. *** Hematology and Immunology Intitute, Havana, Cuba.

Introduction: Most of the sub-Saharan African countries do not have an adequate safe blood supply neither have reliable data about transfusion transmitted infections (TTI). **Objective:** To characterize the blood donation in an Angolan Clinic in Luanda, to determine the causes of donor rejection and to know the prevalence of the main screened TTI. **Material and Methods:** A descriptive and retrospective study was carried out at the Clínica Multiperfil. It was included that 8 043 consecutively recruited apparent healthy blood donor candidates from the year 2005 to 2010. The main analyzed variables were the blood donor status, laboratory test results, and causes of donor rejection. **Results:** Only the 9, 7 % of the blood donations were done by voluntary donor and the rest by family replacement donors. There was a statistics significant difference between the rates of rejection of the blood donor candidates (29.9% of the family replacement donor vs 6.8% of the voluntary blood donors). The main cause of rejection was the reactivity for HBsAg test, both in family replacement donors (13, 4%) and in occasional voluntary (4, 0%). For repetitive voluntary donors, malaria and non-infectious diseases were the main causes of rejection. Reactivity test for TTI was generally elevated (HBsAg: 12, 6 %; malaria: 2,9 %; VDRL: 2,4 %; Anti-HIV 1+2 : 2,3 % and Anti-CHV: 2,1 %), there was a statistically significant difference according to the status of donors. **Conclusions:** In spite of the lower risk for TTI in voluntary blood donor candidates its recruitment was lower than needed.

A1053

INTRODUCTION AND DEVELOPMENT OF REGENERATIVE CELL THERAPY IN CUBA. 2004-2011. Hernández-Ramírez P, Alfonso-Simón A, Ballester-Santovenia

JM, Cabrera-Zamora M, Cruz-Tamayo F, Díaz-Díaz AJ, Gámez-Pérez A, García-Ibarra R, Goicoechea-Díaz P, González-Suárez T, González-Iglesias AI, Lam-Díaz RM, León-Amado L, López-Medina O, Macías-Abraham C, Macías-González R, Obregón-Santos Á, Peix-González A, Pérez-Borrego, A, Pol-Marró N, Socarrás-Ferrer BB, Suárez-Monteagudo CV, Wilford-de León M. Instituto de Hematología e Inmunología, La Habana, Cuba. On Behalf of the Cuban Group for Regenerative Cell Therapy.

Introduction: The first cell therapy clinical trials in Cuba with hematopoietic stem cells began in February 2004 when bone marrow autologous stem cells were implanted into a critical ischemic lower limb of a patient with indication of a major amputation. Result was successful and amputation of the affected limb was avoided. **Objective:** Very encouraged by this result, thereafter regenerative cell therapy with adult autologous bone-marrow derived stem cells or obtained from peripheral blood after stimulation with granulocyte colony stimulating factor was extended to other diseases including bone and joint lesions, periodontitis, coronary heart disease and ischemic brain injury. **Material and Methods:** In the first months of 2011, 10 (66%) of the 15 Cuban provinces were already conducting clinical trials with this new procedure and at the end of the first half of this year a total of 2 799 cases had been performed in the whole country. Sixty seven percent of these cases (1 880) were angiology patients; 792 (28%) were in orthopedics and traumatology and 127 (5%) in other specialties. **Results:** It is noteworthy that in patients with ischemic lower limbs with an indication of major amputation this surgical intervention was avoided on average in 70% of them. Promissory results have also been obtained in the treatment of bone cysts, pseudoarthrosis, avascular necrosis of the femoral head and in some chronic spinal cord injuries. A case that must be highlighted is a 9 year-old boy with idiopathic pulmonary fibrosis and severe respiratory failure resistant to conventional treatments and with indication of lung transplantation. After endobronchial infusion of autologous hematopoietic stem cells the patient recovered, normalized pulmonary function tests and is incorporated into normal life activities. **Conclusions:** At present, we believe that while defining all scientific discussion and the safe therapeutic use of embryonic stem cells or of the embryonic-like induced stem cells is proved, there is sufficient evidence to support the therapeutic application of hematopoietic adult stem cells, because they have so far proved useful and without major adverse effects.

A1136

EFFICIENCY OF PLASMA EXCHANGE IN GUILLAIN-BARRE SYNDROME. Herrera-Aguilar A, Nahuatl-Ciau F, Pacheco-Güemez A, Gutiérrez-Juárez E, Ramírez-Valdez C, Méndez-Gerónimo ML, Pech-Caballero P, Zavala-Cervantes S. Department of Apheresis, Central Blood Transfusion State of Yucatan. Merida, Yucatan, Mexico.

Introduction: Plasma exchange (PE) is to separate the plasma from the cells using centrifugation or membrane filtration, cells reinfused into the patient at the same time that the plasma is discarded together with the toxic factors. Guillain-Barre syndrome (GBS) is a heterogeneous series of immune-mediated peripheral neuropathies, in which plasma exchange is classified as Category I of benefit from American Society for Apheresis. **Objective:** Report the experience of PE in patients with GBS in the Central Blood Transfusion State of Yucatan (CETS) for its acronym in Spanish. **Material and Methods:** We report 2 cases of GBS grade 5 according to the classification of severity of Hughes who required mechanical ventilation (MV) early. They use the apheresis machine CS-3000 which uses the technique of continuous flow centrifugation and in both cases the diagnosis was with support of the neurology service. The circulating plasma volume (CPV) was calculated using the following formula: circulating blood volume by 1 minus the hematocrit of the patient and the replacement was conducted with saline with 5% albumin. The first session was performed with replacement of 2 CPV and CPV following with 1, with a total of 5 sessions in each case. **Results:** Case 1: male 41 year old which evolved over 4 days starting symmetrically with paraesthesia and motor impairment with areflexia, which compromise the respiratory muscles and required MV, the first session was applied at day 7 after starting the condition and the following on 9, 12, 15 and 21; removal of MV was achieved in 27 day and muscle strength (MS) recovered to 4 of 5 on day 64 with the support of physiotherapy. Case 2: male 24 years with acute diarrhea event 4 days prior to the onset of the disease, which evolved with motor impairment and areflexia symmetrical and required MV at day 7. The sessions were applied on day 8, 11, 14, 17 and 23, ensuring the withdrawal of MV on day 8 and recovered MS to 4 of 5 on day 17, for the day 64 the MS was 5 of 5. **Conclusion:** In our short experience with the PE found that we get a very good response in patients with GBS. Currently plans to incursion with the PE in other diseases and the use of other equipment such as plasma separator Amicus.

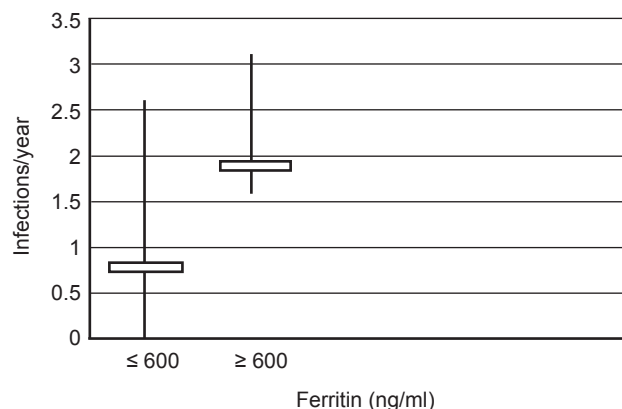
A1210

IMPACT OF IRON OVERLOAD AND INFECTIONS IN PATIENTS ON HEMODIALYSIS. Lopez-Arroyo JL. Hospital General de Zona No. 35, Instituto Mexicano del Seguro Social, Cd. Juárez, Chih, México Hospital General "B" del ISSSTE, Cd. Juárez, Chih, México.

Introduction: Systemic Iron overload (SIO) can be frequently encountered in hemodialysis patients and occurs a consequence

of recurrent blood transfusions. SIO can cause serious organ damage and death if not properly treated. Infections remain a substantial cause of morbidity in patients on hemodialysis being the vascular access the source of the majority of bacteremia.

Objective: The objective of this study is to investigate how the level of serum ferritin correlates with infections in a group of patients undergoing hemodialysis. **Material and Methods:** In a retrospective study of 24 patients undergoing on hemodialysis in two public institutions. calculated with a normal range between 20-380 ng/ml and separating the group, the first group with iron level above of 600 ng/ml serum ferritin and the second group with iron level under 600 ng/ml. **Results:** Of the 24 patients 18 had ferritin levels below 600 ng/ml (median 164.1 ng/ml) had 28 infections (0.8/year, range 0 - 2.6) (figure 1), the vascular access was the source in 78.5% (figure 2) after a follow-up of 15 months. Six patients had ferritin level above 600 ng/ml (median 830.3, range 605-1334 ng/ml) and had 24 infections (1.9/year, range 1.6-3.1) among these patients only a 25% had a vascular access infections between another source of infections after a follow-up of 24 months. **Conclusion:** In this sample of adult patients on hemodialysis serum ferritin above 600 ng/ml was mostly associated with infections complications and less associated to vascular access. This study shows that when the iron level above 600 ng/ml this type of patients are more prone to develop infections.

**A1225**

PREVALENCE OF NONINFECTIOUS SERIOUS HAZARDS OF TRANSFUSION IN THE CENTENARIO HOSPITAL MIGUEL HIDALGO AT AGUASCALIENTES, AGUASCALIENTES, MEXICO. Cardiel Silva M, Rosas Cabral A, Rosales Andrade J, Centenario Hospital Miguel Hidalgo, Aguascalientes, Aguascalientes

Introduction: As infectious complications from blood transfusions have decreased because improved and sophisticated infectious disease blood screening, non infectious serious hazards of transfusion have emerged as the most common complications of transfusion. The noninfectious serious hazards of transfusion included: transfusion reactions (hemo-

lytic, febrile, septic, allergic/anaphylactic) and to lesser known complications that mistransfusion, transfusion acute lung injury, transfusion-related overload, posttransfusion purpura, transfusion-associated graft versus host disease. **Objectives:** To determine the prevalence of transfusion reactions (TR) and increase the transfusion safety in our institution from 2008 to 2010, all adverse effects transfusion related were recorded and analyzed. **Material and methods:** A retrospective analysis of transfusion reactions from January 2008 to December 2010 was undertaken. Data were collected from transfusion reports on this time in our hospital. **Results:** A total of 37 transfusion reactions in 40,868 blood products transfused (0.09%) were

reported, of which 15 were classified as allergic reactions, followed by febrile non-hemolytic transfusions (FNHT) (14), one acute lung injury and seven intravascular hemolysis. 16 TR were on female and 21 in males. Mean age of presentation was 42 years. (Table 1). The blood component with more transfusion reactions was red blood cells (table 2), and male were more affected than female. There is not relation between time of blood extraction on days and TR occurrence. Table 3. **Conclusions:** The prevalence of transfusion reactions was 0.09%, these data conducted to adoption of corrective actions to increase safety transfusion in our institution, through introduction of universal leucoreduced blood components.

Table 1. Transfusion reactions in the Centenario Hospital Miguel Hidalgo at City of Aguascalientes from 2008 to 2010.

Year	Total of transfusions	Total of reactions	% Reactions	FNHT	Allergic	Reaction type	
						Acute lung injury	Intravascular
2008	14422	16	0.11	9	7	0	0
2009	13892	14	0.10	4	4	1	5
2010	12554	7	0.05	1	4	0	2
	40,868	37	0.09	14	15	1	7