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Impact of MYC, BCL2, and/or BCL6 expression on the survival of patients with diffuse large B-cell lymphoma.

Efecto de la expresión de MYC, BCL2 o BCL6 en la supervivencia de pacientes con linfoma difuso de células grandes B

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Abstract

OBJECTIVE: To evaluate the prognostic significance of MYC, BCL2, and BCL6 expression in a Mexican cohort of patients with diffuse large B-cell lymphoma.

MATERIALS AND METHODS: A retrospective study including patients with *de novo* diffuse large B-cell lymphoma diagnosed at the Hospital General de Mexico between 2014 and 2018. Immunohistochemistry was used to assess protein expression, with positivity defined as > 40% for MYC, > 70% for BCL2, and > 50% for BCL6.

RESULTS: There were included 220 patients. Median age was 58 years, and 56.4% were male. MYC, BCL2, and BCL6 were positive in 30%, 27.3%, and 10% of cases, respectively. With a median follow-up of 19 months, the 3-year overall survival was significantly lower in MYC-positive *vs* negative cases and in BCL2-positive *vs* negative cases. Double-expressor lymphomas (MYC+/BCL2+) had the worst prognosis. BCL6 expression did not significantly impact overall survival. In multivariate analysis, extranodal disease and MYC+/BCL2+ status were independent predictors of poor overall survival.

CONCLUSIONS: MYC and BCL2 expression, individually and combined, are associated with inferior survival in Mexican patients with diffuse large B-cell lymphoma, underscoring the importance of routine immunohistochemical evaluation for risk stratification.

KEYWORDS: Lymphoma, large B-cell, diffuse; Prognosis; Mexico.

Resumen

OBJETIVO: Evaluar la importancia pronóstica de la expresión de MYC, BCL2 y BCL6 en una cohorte mexicana de pacientes con linfoma difuso de células B grandes.

MATERIALES Y MÉTODOS: Estudio retrospectivo que incluyó pacientes con linfoma difuso de células B grandes *de novo*, diagnosticados en el Hospital General de México entre 2014 y 2018. Se utilizó inmunohistoquímica para evaluar la expresión proteica; la positividad se definió como más del 40% para MYC, más del 70% para BCL2 y más del 50% para BCL6.

RESULTADOS: Se incluyeron 220 pacientes. La mediana de edad fue de 58 años y el 56.4% eran varones. MYC, BCL2 y BCL6 fueron positivos en el 30, 27.3 y 10% de los casos, respectivamente. Con una mediana de seguimiento de 19 meses, la supervivencia global a tres años fue significativamente menor en los casos con MYC positivo frente a negativo y en los casos con BCL2 positivo frente a negativo. Los linfomas de

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doble expresión (MYC+/BCL2+) mostraron el peor pronóstico. La expresión de BCL6 no afectó significativamente la supervivencia global. En el análisis multivariable, la enfermedad extranodal y el estado MYC+/BCL2+ fueron predictores independientes de mala supervivencia global.

CONCLUSIONES: La expresión de MYC y BCL2, individual y combinada, se asocia con menor supervivencia en pacientes mexicanos con linfoma difuso de células B grandes, lo que subraya la importancia de la evaluación inmunohistoquímica rutinaria para la estratificación del riesgo.

PALABRAS CLAVE: Linfoma difuso de células B grandes; pronóstico; México.

INTRODUCTION

Diffuse large B-cell lymphoma (DLBCL) is the most common lymphoma in Mexico; it represents up to 64% of B-cell lymphomas, while in the United States up to 40% of all lymphomas.[1,2] DLBCL can be divided into germinal center B cell type (GCB type) and non-germinal center B cell type (non-GCB type) based on the presence or absence of three biomarkers, CD10, BCL-6, and MUM-1 in immunohistochemical staining using antibodies against CD10, IRF4/MUM1, and BCL6.[2,3] Treatment is based on immunochemotherapy with rituximab, cyclophosphamide, doxorubicin, vincristine, and prednisone (R-CHOP). Immunochemotherapy treatment with rituximab, cyclophosphamide, doxorubicin, vincristine and prednisone (R-CHOP) can achieve between 50-70% success, depending on factors such as age, extranodal involvement, DHL serum levels, ECOG-performance, abnormalities genetic, among others.[2,4]

MYC is rearranged in 5% to 15% of DLBCL, and is frequently associated with BCL2 or, to a lesser extent, BCL6 translocation, in the so-called "double-hit" or "triple-hit" lymphomas that are included in the updated WHO classification in the new category of high-grade B-cell lymphoma (HGBL), with rearrangements of MYC and BCL2 and/or BCL6.[5,6]

MYC protein expression is detected in a higher proportion of DLBCL (30%-50%) and is associated with concomitant expression of BCL2 in 20% to 35% of cases. In several but not all studies, the double-expressor lymphomas have a worse outcome than other DLBCL, NOS but they are not as aggressive as the HGBL, with rearrangements of MYC and BCL2 and/or BCL6.[5,7] Patients with either MYC/BCL6 rearrangements or MYC/BCL6 co-expression did not always have poorer prognosis.[8]

The study aimed to investigate the prognostic value of expression of MYC, BCL2, and/or BCL6 in a Mexican population of diffuse large B-cell lymphoma patients.

MATERIALS AND METHODS

A retrospective study conducted at the Hospital General de México Dr Eduardo Liceaga, Mexico City. The study protocol was approved by the institutional review board. The study comprised 354 patients with de novo DLBCL who were treated with immunotherapy, R-CHOP, or more

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intensive treatment between January 2014 and December 2018.

Patients' medical records were thoroughly revised, and relevant data were collected. These included baseline patient characteristics, pretreatment investigations, treatment response, the time of death, and the last time of follow up.

The histological diagnosis of DLBCL was performed by local pathologists using the initial diagnostic biopsies. Cell-of-origin (COO) was determined by the Hans algorithm. Subsequently, the expression of MYC, BCL2, and BCL6 was analyzed; specimens that expressed > 40% of MYC, > 70% of BCL2, and > 50% of BCL6 were classified as positive. The patients included in the present study were selected only by the availability of the evaluation of MYC, BCL2, and BCL6 expression. Continuous data were presented as median and range while categorical data were expressed as number and percentage.

The primary endpoint of the present study was overall survival (OS) calculated from the date of diagnosis until death due to any cause and was censored at the last follow-up. All survival curves were obtained by the Kaplan-Meier method and compared using the log-rank test. Values of $p < 0.05$ were considered statistically significant in all analyses.

To evaluate the presence of an interaction between expression of MYC, BCL2, BCL6 and other risk factors (age >60 years, ECOG >1, extranodal presentation, elevated LDH, COO and stage III/IV) a multivariate Cox regression model for OS was developed that included these variables and their product. Statistical analyses were carried out with SPSS 25.

RESULTS

Immunohistochemical data to evaluate the expression of MYC, BCL2 and BCL6 were available for 220 of the 354 patients. The median age was 58 years (18-90) predominantly affecting the male sex (56.4%). The general clinical characteristics of the patients are shown in **Table 1**.

Table 1. Patient general characteristics

	n (%)
Age	
< 60 years	110 (50)
≥ 60 years	110 (50)
Gender	
Male	124 (56.4)
Female	96 (43.6)
ECOG performance	
0, 1	86 (39.1)
2, 3, 4	134 (60.9)
Clinical stage	
I, II	71 (32.3)
III, IV	149 (67.7)
LDH level	
Normal	80 (36.4)
Elevated	140 (63.6)
Extranodal disease	
Absent	83 (37.7)
Present	137 (62.3)
Cell-of-origin	
GC	124 (56.4)
Non-GC	85 (38.6)
Unclassificable	11 (5)
IPI	
Low risk	49 (22.3)
Low-intermediate risk	57 (25.9)
High-intermediate risk	59 (26.8)
High risk	25 (25)
MYC expression	
Positive	154 (70)
Negative	66 (30)
BCL2 expression	
Positive	160 (72.7)
Negative	60 (27.3)
BCL6 expression	
Positive	198 (90)
Negative	22 (10)
First line treatment	
RCHOP or RCHOP-like regimen	195 (88.7)
None	25 (11.4)

Expression of MYC, BCL2 and BCL6 was 30%, 27.3% and 10% respectively.

Outcomes

Most patients (88.7%) received R-CHOP or CHOP-like chemotherapy of which 56.8% achieved a complete response. Patients whose lymphoma recurred despite first-line therapy were treated with a variety of secondary regimens. Hematopoietic transplantation was not performed in any patient for relapsed disease.

Overall survival

With a median follow-up of 19 months (range, 4 to 43 months) the 3-year OS in the entire patient cohort was 70% (66 of the 220 patients died), the median OS was not reached (**Figure 1**). There was a significant difference in 3-year OS between the MYC positive and negative cases (39% vs 78%; p < 0.001), BCL2 positive and negative cases (41% vs 76%; p < 0.001), but not for BCL6 positive or negative cases (53% vs 67%; p = 0.27; **Figure 2**). Median OS was not reached (95% CI not estimable) in the MYC negative vs 15 months (95% CI 11.6-18.3) in the MYC positive cases; not reached (95% CI not estimable) in the BCL2 negative vs 15 months (95% CI 11.8-18.2) in the BCL2 positive cases and not reached (95% CI not estimable) in the BCL6 negative and positive groups. In MYC and BCL2 positive cases, the 3-year OS was even lower (33% vs 75%; p < 0.001). No difference in OS was observed between CG and non-CG cases (69% vs 62%; p = 0.26).

By multivariate Cox's proportional regression analysis, the proportional individual risk for extranodal presentation (HR 1.98, 95% CI 1.15-3.41; p = 0.01), and MYC plus BCL2 expression (HR 4.03, 95% CI 2.40-6.75; p < 0.001) were significant for lower OS. **Table 2 and Figure 3**

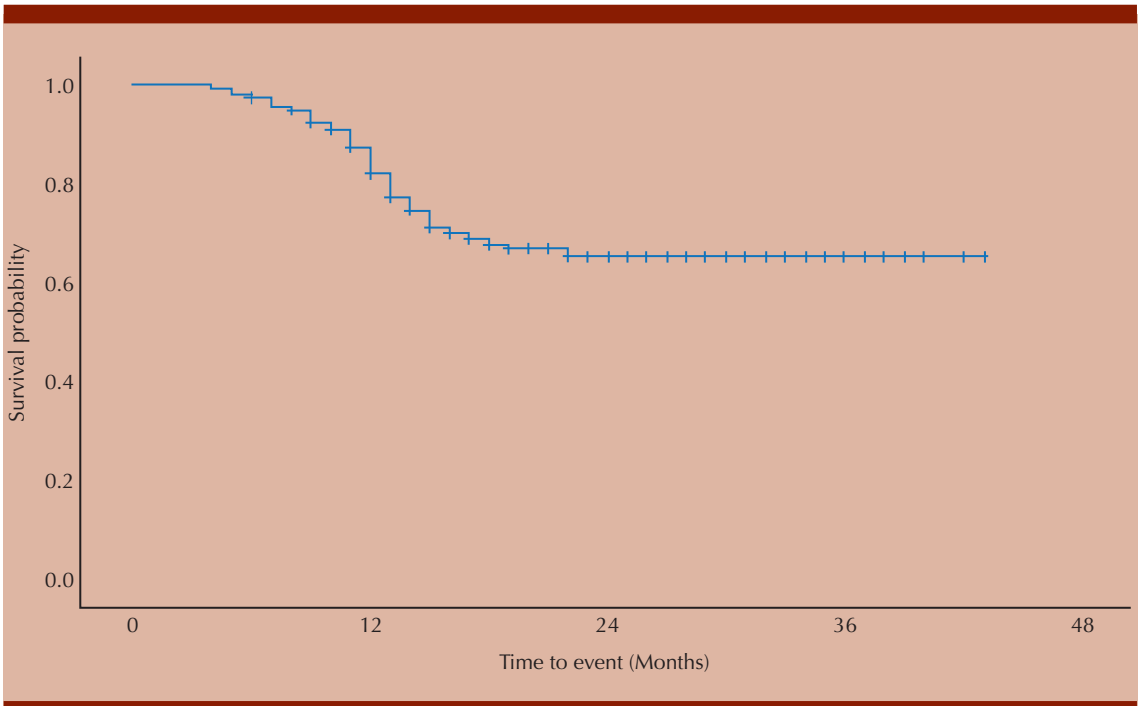

Figure 1. Overall survival in the entire patient cohort.

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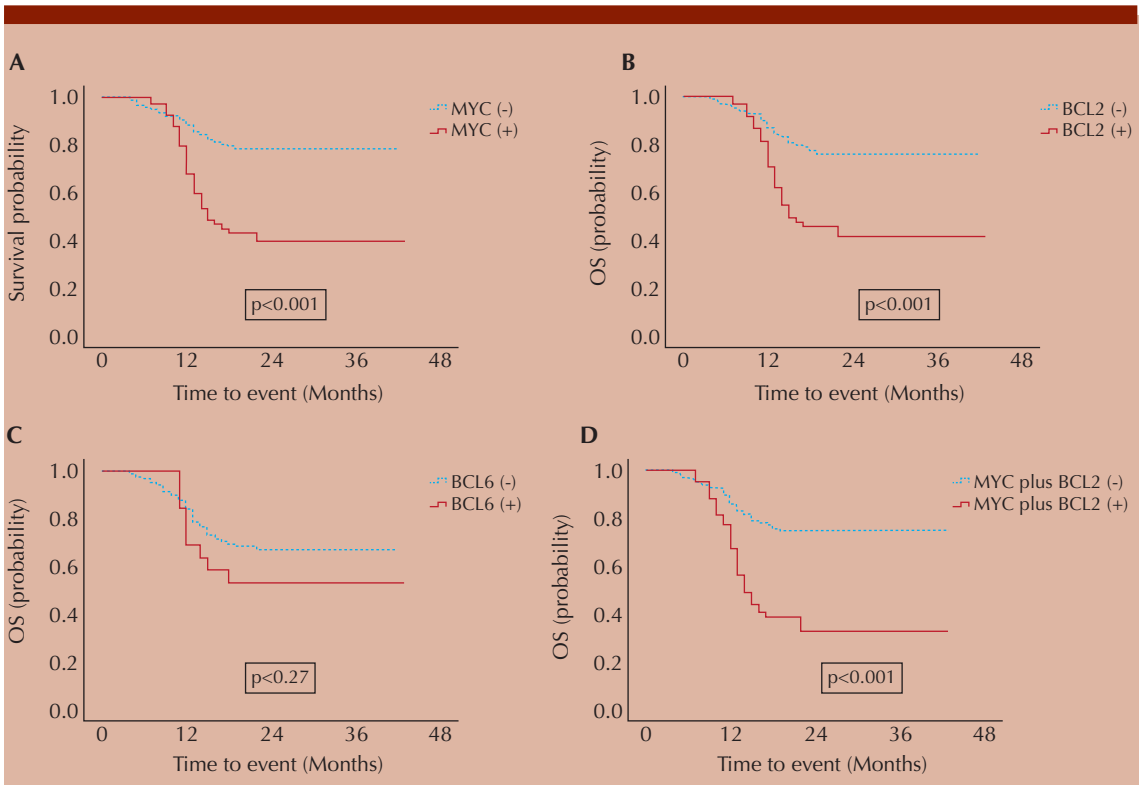

Figure 2. A. Overall survival between the MYC positive and negative cases. **B.** Overall survival between the BCL2 positive and negative cases. **C.** Overall survival between the BCL6 positive and negative cases. **D.** Overall survival between the MYC plus BCL2 positive and negative cases.

Table 2. Univariate and multivariate analysis of survival

	Unadjusted HR (CI 95%)	p	Adjusted HR (CI 95%)	p
Age > 60 years	1.38 (0.85-2.26)	0.18	1.38 (0.84-2.27)	0.19
ECOG performance > 1	0.82 (0.5-1.33)	0.42	0.69 (0.42-1.14)	0.15
Extranodal involvement	1.77 (1.03-3.01)	0.03	1.98 (1.15-3.41)	0.01
Cell-of-origin (Non-GC)	1.47 (0.98-2.20)	0.06	1.39 (0.93-2.08)	0.10
Elevated LDH	1.20 (0.83-1.73)	0.32	1.60 (0.94-2.72)	0.08
Clinical stage III-IV	1.36 (0.8-2.3)	0.24	1.32 (0.77-2.26)	0.30
MYC and BCL2 expression	3.33 (2.04-5.43)	0.00	4.03 (2.40-6.75)	0.00

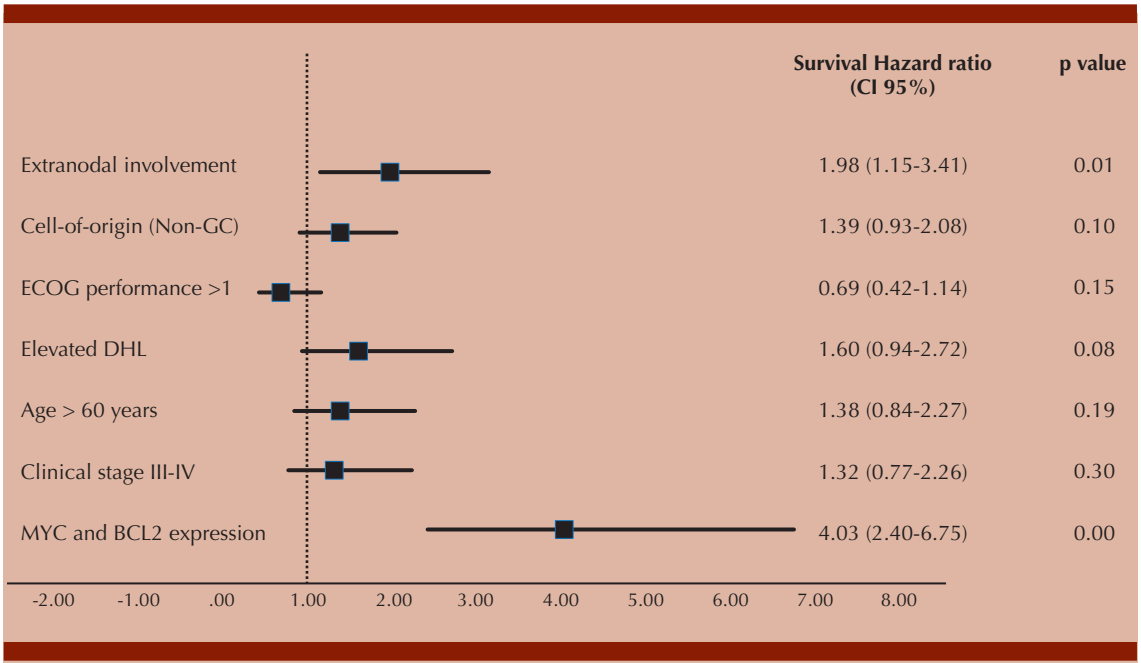

Figure 3. Forest plot showing hazard ratio estimates for overall survival.

DISCUSSION

Diffuse large B cell lymphoma is a heterogeneous disease, as its study progresses, different subtypes with particular biological characteristics have been described, which can influence the prognosis.

The epidemiological characteristics of our population and the response rates to first-line treatment are similar to those reported worldwide.

BCL2 is detectable in approximately 50% of DLBCL and 75% of high-grade B-cell lymphomas, whose effect is to inhibit cell apoptosis and promote cell proliferation, which interacts with the action of MYC. BCL6 is a nuclear transcriptional repressor. Abnormal expression of BCL6 can directly regulate cell differentiation, proliferation and apoptosis to promote tumor growth and differentiation.[9] MYC encodes a helix-loop-helix transcription factor that accentuates many cellular functions including proliferation, growth and apoptosis. MYC alterations also have been identified in other mature B-cell neoplasms and are associated with aggressive clinical behavior.[10] MYC is rearranged in 5% to 15% of DLBCL, and is frequently associated with BCL2 or, to a lesser extent, BCL6 translocation, in the so-called "double-hit" or "triple-hit" lymphomas that are included in the updated WHO classification in the new category of high-grade B-cell lymphoma (HGBL), with rearrangements of MYC and BCL2 and/or BCL6.[5]

Immunohistochemical detection of MYC, BCL2 or BCL6 expression is not equivalent to translocation, however studies have shown that the expression of MYC and BCL2 is related to more aggressive behavior and worse survival. The influence of BCL6 expression on survival is controversial.[8,11-15]

In our group of patients analyzed, MYC expression was the most frequent (30%), followed by BCL2 expression (27%) and finally BCL6 (10%),

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when analyzing the impact on survival according to the expression of these markers, a relationship was found in the expression of MYC and BCL2 separately, as well as their simultaneous expression, however, the expression of BCL6 did not significantly impact survival.

These findings are correlated with those reported by other groups, which warns of the importance of performing an immunohistochemical search for MYC and BCL2 expression since these patients may have an aggressive course of the disease and lower overall survival. Although it would be desirable to have tests to detect MYC, BCL2 and BCL6 rearrangements, in our context it is often not possible, and although the expression of these proteins is not related to a genetic translocation, it gives us valuable information on the prognosis of patients with diffuse large cell lymphoma.

CONCLUSION

This study has identified a group of patients with DLBCL at high clinical risk who overexpress the MYC, BCL2. The expression of BCL6 in our population was low and was not related to a decrease in overall survival.

REFERENCES

1. Hernandez-Ruiz E, Alvarado-Ibarra M, Lien-Chang LEJ, et al. Epidemiology and clinical characteristics of non-Hodgkin lymphoma in Mexico. World J Oncol 2021; 12 (1): 28-33. doi: 10.14740/wjon1351
2. Susanibar-Adaniya S, Barta SK. 2021 Update on Diffuse large B cell lymphoma: A review of current data and potential applications on risk stratification and management. Am J Hematol 2021; 96 (5): 617-29. doi: 10.1002/ajh.26151
3. Hans CP, Weisenburger DD, Greiner TC, et al. Confirmation of the molecular classification of diffuse large B-cell lymphoma by immunohistochemistry using a tissue microarray. Blood 2004; 103 (1): 275-82. doi: 10.1182/blood-2003-05-1545
4. Morrison VA. Frontline therapy with R-CHOP for diffuse large B-cell lymphoma: Where have we come (or not come)? A perspective. J Geriatr Oncol 2021; 12 (2): 320-5. doi: 10.1016/j.jgo.2020.09.015
5. Swerdlow SH, Campo E, Pileri SA, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms Blood 2016; 127: 2375-90. http://dx.doi.org/10.1182/blood-2016-01-643569
6. Swerdlow SH. Diagnosis of "double hit" diffuse large B-cell lymphoma and B-cell lymphoma, unclassifiable, with features intermediate between DLBCL and Burkitt lymphoma: when and how, FISH versus IHC. Hematology 2014; 2014: 90-9. https://dx.doi.org/10.1182/asheducation-2014.1.90
7. Johnson NA, Slack GW, Savage KJ, Connors JM, et al. Concurrent expression of MYC and BCL2 in diffuse large B-cell lymphoma treated with rituximab plus cyclophosphamide, doxorubicin, vincristine, and prednisone. J Clin Oncol 2012; 30 (28): 3452-9. doi: 10.1200/JCO.2011.41.0985
8. Ye Q, Xu-Monette ZY, Tzankov A, et al. Prognostic impact of concurrent MYC and BCL6 rearrangements and expression in de novo diffuse large B-cell lymphoma. Oncotarget 2016; 7 (3): 2401-16.
9. Zhang Y, Wang H, Ren C, et al. Correlation between C-MYC, BCL-2, and BCL-6 protein expression and gene translocation as biomarkers in diagnosis and prognosis of diffuse large B-cell lymphoma. Front Pharmacol 2018; 9: 1497. https://doi.org.10.3389/fphar.2018.01497
10. Cai Q, Medeiros LJ, Xu X, Young KH. MYC-driven aggressive B-cell lymphomas: biology, entity, differential diagnosis and clinical management. Oncotarget 2015; 6 (36): 38591-616. doi: 10.18632/oncotarget.5774
11. Horn H, Ziepert M, Becher C, et al. MYC status in concert with BCL2 and BCL6 expression predicts outcome in diffuse large B-cell lymphoma. Blood 2013; 121 (12): 2253-63. doi: 10.1182/blood-2012-06-435842
12. Ma Z, Niu J, Cao Y, et al. Clinical significance of "double-hit" and "double-expression" lymphomas. J Clin Pathol 2020; 73 (3): 126-38. doi: 10.1136/jclinpath-2019-206199
13. Huang S, Nong L, Wang W, et al. Prognostic impact of diffuse large B-cell lymphoma with extra copies of MYC, BCL2 and/or BCL6: comparison with double/triple hit lymphoma and double expressor lymphoma. Diagn Pathol 2019; 14 (1): 81. doi: 10.1186/s13000-019-0856-7.
14. Ting C-Y, Chang K-M, Kuan J-W, et al. Clinical significance of BCL2, C-MYC, and BCL6 genetic abnormalities, Epstein-Barr virus infection, CD5 protein expression, germinal center B cell/non-germinal center B-cell subtypes, co-expression of MYC/BCL2 proteins and co-expression of MYC/BCL2/BCL6 proteins in diffuse large B-cell lymphoma: A clinical and pathological correlation study of 120 patients. Int J Med Sci 2019; 16: 556-66. http://dx.doi.org/10.7150/ijms.27610
15. Staiger AM, Ziepert M, Horn H, et al. Clinical impact of the cell-of-origin classification and the MYC/BCL2 dual expresser status in diffuse large B-cell lymphoma treated within prospective clinical trials of the German High-Grade Non-Hodgkin's Lymphoma Study Group. J Clin Oncol 2017; 35: 2515-26. http://dx.doi.org/10.1200/jco.2016.70.3660