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Molecular Classification of Large B-Cell Lymphoma and High-Grade B-Cell Lymphoma Cases and Association with Outcomes in Morocco

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Abstract: Background: High-grade B-cell lymphoma with *c-MYC* and *BCL2* and/or *BCL6* rearrangements (HGBL-DHL/THL) is a recently identified category in the most recent World Health Organization (WHO) classification. For all tumors displaying the appearance of diffuse large B-cell lymphoma (DLBCL) or high-grade B-cell lymphoma (HGBL), it is necessary to perform fluorescence in situ hybridization (FISH) in order to achieve an accurate diagnosis. The findings of FISH and immunohistochemistry (IHC) examinations from 50 DLBCL/HGBL samples obtained from Hassan II University Hospital in Fez/Morocco are reported. **Methods:** This retrospective study included 50 patients diagnosed with DLBCL/HGBL over a period of nine years (2013–2022) and treated with RCHOP chemotherapy protocol. All patients underwent a histological study followed by an immunohistochemical study to confirm the diagnosis and to classify patients according to cell of origin into non-GCB and GCB subtypes; then, a cytogenetic study using FISH was performed to classify patients according to the presence or absence of rearrangements in the *c-MYC*, *BCL2* and *BCL6* genes. A comparison was made between the molecular subtypes of DLBCL/HGBL in relation to clinicopathological features and outcomes. **Results:** Among the 50 cases studied in our population, we found 5 cases of HGBL with DLBCL morphology and 45 cases of DLBCL, which consisted of 13 cases (28.89%) of GCB subtype and 32 cases (71.11%) of non-GCB subtype based on the immunohistochemistry Hans algorithm. After FISH testing of all cases, we found three cases of double-hit lymphoma (DHL) and one case of triple-hit lymphoma (THL). Thus, HGBL-DHL/THL accounted for 8% of the cases. Furthermore, two cases were detected with only one rearrangement in the *BCL2* gene and one case harboring a rearrangement in the *BCL6* gene. DHL and THL patients and patients with a single rearrangement (*BCL2* or *BCL6*) have a worse prognosis than patients with no rearrangement. **Conclusions:** DHL and THL are an aggressive entity of HGBL with poorer outcomes in comparison to DLBCL/HGBL NOS. First-line treatment with the RCHOP chemotherapy protocol may not be effective for all aggressive DLBCL cases. More targeted treatment is crucial for better patient outcomes.

Keywords: high-grade B-cell lymphoma; double-hit lymphoma; triple-hit lymphoma; germinal center B cell; FISH; immunohistochemistry; Hans algorithm

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1. Introduction

Several studies demonstrated the impact of the cell of origin (COO) in the classification of diffuse large B-cell lymphoma (DLBCL). Gene expression profiling defines three subgroups (GCB, ABC and unclassified), while classification via immunohistochemistry adopting the Hans algorithm dissects DLBCL down into two subtypes: germinal center B cell (GCB) and non-germinal center B cell (non-GCB) [1]. This classification has clinical relevance, in that patients with DLBCL of ABC origin (non-GCB) have a worse prognosis than those with the CGB subtype. In addition to the role of cell of origin in predicting the prognostic value of immunohistochemical subtypes, fluorescence in situ hybridization analysis (FISH) can detect rearrangements in DLBCL and has shown that GCB and ABC tumors have different molecular profiles. Adopting a precision medicine strategy based on molecular discoveries in DLBCL is the most optimal way to develop novel therapeutic targets.

In this regard, the World Health Organization 2017 Classification of Tumors of Hematopoietic and Lymphoid Tissues has introduced a new entity: high-grade B-cell lymphoma with *c-MYC* and *BCL2* and/or *BCL6* rearrangements (HGBL-DHL/THL) [1]. Several previous studies have shown that this entity is associated with poor prognosis. The proportion of HGBL-DHL/THL among tumors with diffuse large B-cell lymphoma (DLBCL) morphology is estimated to be 1–12% [2–4]. Patients diagnosed with these lymphomas have an aggressive clinical course characterized by advanced-stage disease, extranodal involvement, high serum lactate dehydrogenase (LDH) levels and high-intermediate–high IPI score [5,6]. Paradoxically, cases of *c-MYC/BCL2* or *BCL6* DHL and *c-MYC/BCL2/BCL6* THL have a favorable GCB cell of origin [7,8].

Myelocytomatosis viral oncogene homolog (*c-MYC*), situated at 8q24, is a nuclear protein playing a role of a transcription factor that regulates the expression of around 10% of genes involved in cellular differentiation, proliferation and programmed cell death. Chromosomal abnormalities of the *c-MYC* gene are associated with poor prognostic outcomes; hence, the *c-MYC* proto oncogene is qualified as a negative prognostic parameter [9]. *c-MYC* rearrangements can occur in 4–14% of DLBCLs and can affect both the GCB and ABC subtypes [10].

The B-cell lymphoma 2 (*BCL2*) gene is situated at 18 q21. It is a member of *BCL2* family genes that encode the synthesis of proteins responsible for the regulation control of programmed cell death (PCD)—that is, apoptosis induced via the mitochondrial pathway [11].

The B-cell lymphoma 6 (*BCL6*) gene is located at 3q27, encoding protein synthesis, which is a transcription factor playing an important role in the formation and normal functioning of germinal centers, prevention of DNA double-stranded break-induced apoptosis in B lymphocytes and cell cycle regulation [9].

Several researchers have evaluated the predictive significance of *BCL2* or *BCL6* rearrangement in patients with DLBCL. The investigations have presented conflicting findings, indicating that these rearrangements may or may not have a prognostic influence in patients diagnosed with DLBCL [12,13].

The aim of this study was to classify DLBCL/HGBL cases based on molecular criteria according to the presence or absence of rearrangements in the *c-MYC*, *BCL2* and *BCL6* genes. The relationships between molecular subtypes and clinicopathological aspects and prognosis in the Moroccan context were analyzed.

2. Materials and Methods

2.1. Patients

This is a retrospective study including 50 patients diagnosed with DLBCL/HGBL according to the World Health Organization (WHO) classification between 2013 and 2022 at Hassan II University Hospital, Fez, Morocco.

The choice of patients was based, on the one hand, on the availability of a good-quality specimen and clinical and follow-up data; on the other hand, it was based on the availability of the FISH technique.

Staging was based on the Ann Arbor staging system, which is commonly used to classify the extent of disease in patients with DLBCL. The system consists of four stages: stage I involves a single lymph node region or a single extranodal site; stage II includes two or more lymph node regions on the same side of the diaphragm or with limited extranodal involvement; stage III involves lymph node regions on both sides of the diaphragm, possibly with spleen or localized extranodal involvement; and stage IV indicates diffuse or disseminated involvement of one or more extralymphatic organs, such as the liver, bone marrow or lungs.

Regarding the response criteria, the Lugano classification was used to assess treatment response in lymphoma patients. It classifies responses into four categories, including complete remission (CR), partial remission (PR), stable disease (SD) and progressive disease (PD).

2.2. Morphological Features

The tissue samples were fixed in formalin, normally treated, embedded in paraffin and cut into 4 µm slices before being stained with hematoxylin and eosin (HE). After reviewing the HE-stained slides from each tumor block, representative regions with the highest level of tumor cells were chosen for an immunohistochemical and FISH analysis (Figure 1).

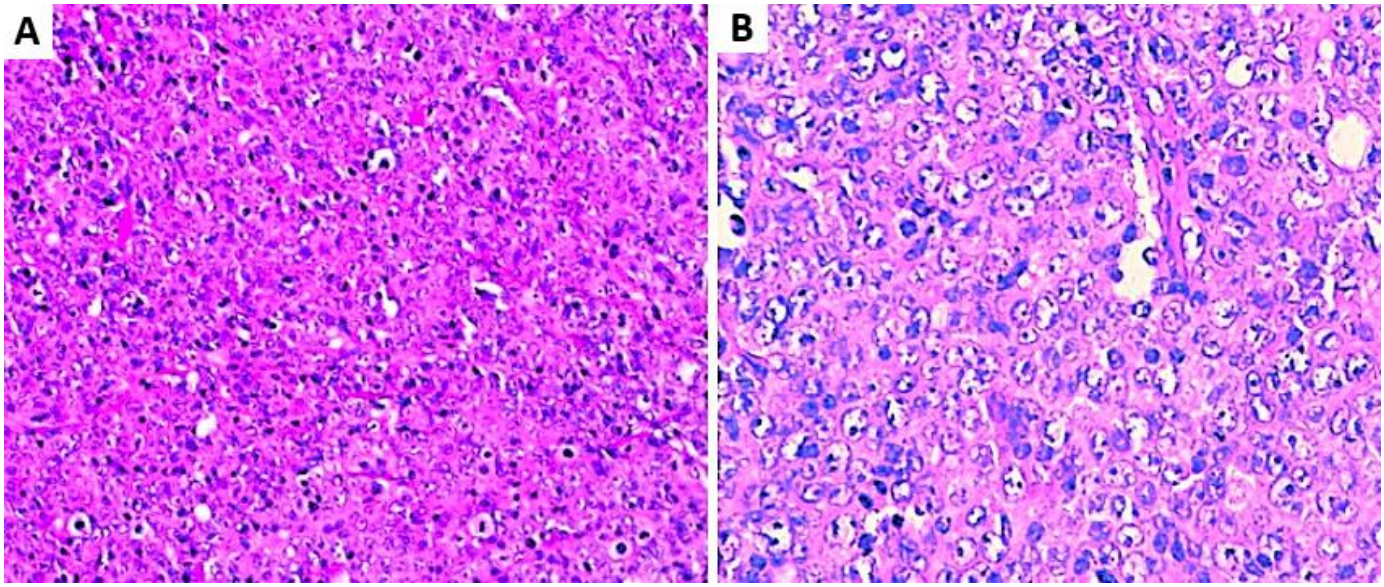

Figure 1. Morphological appearance of DLBCL cells. (**A**) Diffuse proliferation of large B cells (HES×200). (**B**) Large tumor cells (HES×400).

2.3. Immunohistochemistry Study

Formalin-fixed, paraffin-embedded tissue sections were used for the immunohistochemistry process. *CD10* (clone 56C6), *BCL6* (clone LN22) and *MUM1* (clone MUM1p) were used as markers. The immunoreaction was performed in an automated Dako Cover Stainer for these antibodies.

The classification as GCB versus non-GCB subtype was based on the Hans algorithm, using *CD10*, *BCL6* and *MUM1* expression with a cutoff of 30% of positive cells (Figure 2) [14].

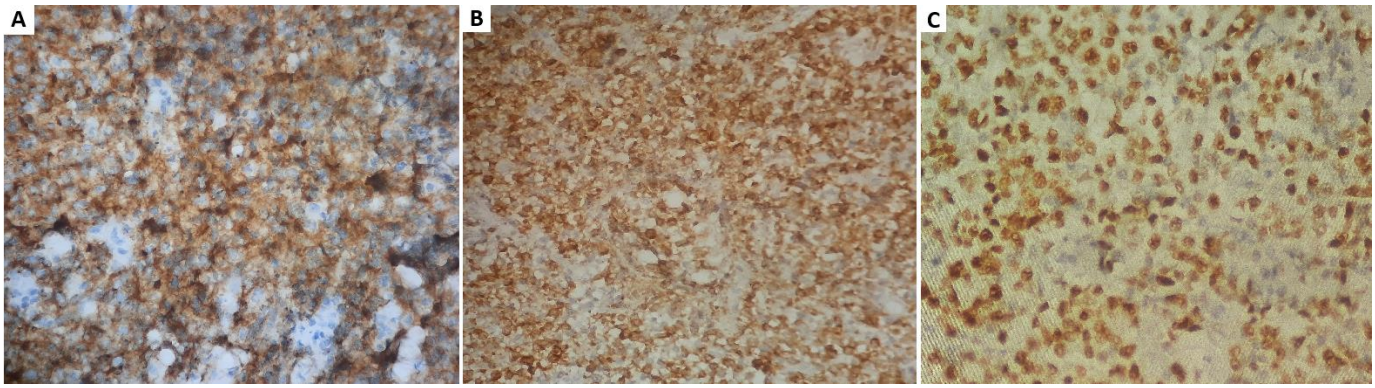

Figure 2. Immunostaining with cluster of differentiation (*CD10*), B-cell lymphoma 6 (*BCL6*) and multiple myeloma oncogene 1 (*MUM1*) in diffuse large B-cell lymphoma cellblocks. (**A**) *CD10*-positive, (**B**) *BCL6*-positive, (**C**) *MUM1*-positive. (Immunostaining, ×400).

2.4. Cytogenetic Study

The FISH analysis was performed on 4 µm tumor microarray tissue using break-apart FISH DNA probes for *c-MYC*/8q24. 21, *BCL2*/18q21. 33 and *BCL6*/3q27 (probes PL49, PL150 and PL136; Zyto Light). Paraffin-embedded tissue sections were deparaffined and pretreated with a heat pretreatment solution at 98 °C for 15 min. The slides were then subjected to enzymatic digestion using pepsin solution at 37 °C for 15 min. Slides were denatured for 10 min at 75 °C and hybridized overnight at 37 °C in Hybrite equipment. Following hybridization, the coverslips were removed, and the slides were washed at 37 °C for 2 × 5 min in 1 × wash buffer. After dehydration, the samples were counterstained with DuraTeC/Dapi solution and covered for stockage at 2–8 °C in obscurity.

For the detection of *c-MYC*/*BCL2*/*BCL6* rearrangements, a signal pattern consisting of one orange/green fusion signal, one orange signal and a separate green signal indicates one normal locus and one locus affected by rearrangement (Figure 3).

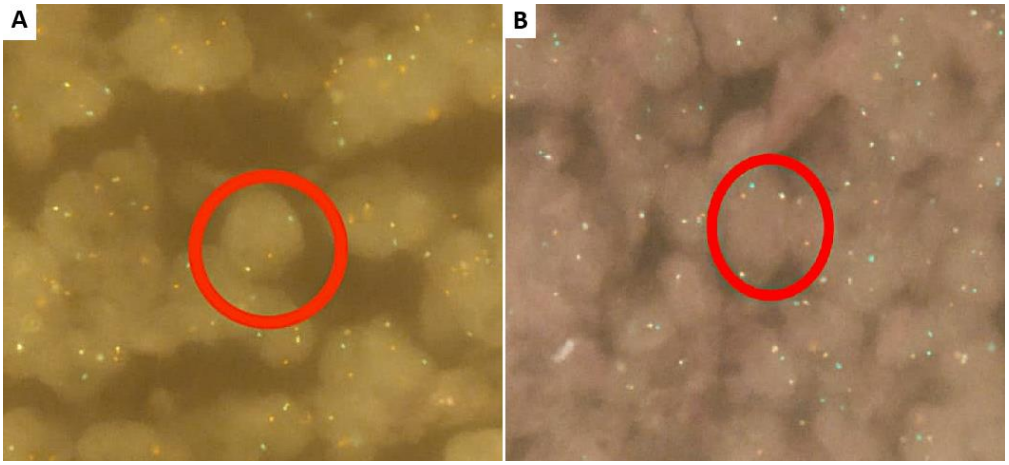

Figure 3. FISH testing with break-apart FISH DNA probes in diffuse large B-cell lymphoma cells. (**A**) *c-MYC* gene rearranged, (**B**) *BCL6* gene rearranged.

Cases with break-apart signals in more than 5% of nuclei were considered positive for the presence of rearrangements.

2.5. Survival Analysis

From the date of diagnosis to the first occurrence of progression, relapse or death, the event-free survival (EFS) period was determined. From the date of diagnosis to the date of death, the overall survival (OS) period was calculated. For some patients who

had an antecedent history of diffuse large B-cell lymphoma prior to the first year of our study (2013), survival was calculated from the date of initial diagnosis. These patients were included in the study due to disease progression or relapse during the 2013–2022 study period, making their cases relevant for the analysis of treatment and survival outcomes.

3. Results

In our population, there were 26 males (52%) and 24 females (48%) (sex ratio: 1.08), with a median age of 59.5 years (range: 11–92 years), and 21 patients (42%) were aged over 60 years. Moreover, 43 patients (86%) in our series exhibited an elevated LDH level; 40 patients (80%) had a high Ann Arbor stage III/IV; and 37 patients (74%) had an appearance of B symptoms. In addition, 28 patients (56%) had a high IPI score ($\geq$3), and 35 patients (70%) had an extranodal involvement of at least one site.

Regarding the tumor locations, it was noted that 24 patients (48%) had primary nodal involvement. Among these cases, the cervical location was the most frequent in 22 patients (91%). Primary extranodal involvement was observed in 26 patients (52%). Among these cases, the digestive location was the most frequent, with a percentage of 27% (seven cases).

In our series, we found 5 cases of HGBL with DLBCL morphology (10%) and 45 cases of DLBCL (90%). DLBCL cases were classified into two immunohistochemical subtypes based on the Hans algorithm: 13 patients (28.89%) had the GCB subtype, and 32 patients (71.11%) had the non-GCB subtype.

In our series, it was observed that *Ki67* was high (>90%) in 14 cases (28%).

By using the molecular cytogenetic test via FISH, three cases (6%) of DHL and a single case (2%) of THL were detected in our series of studies. DHL and THL represented 8% of all cases. Two cases (4%) were detected with a rearrangement in the *BCL2* gene only, and only one case (2%) was detected with a rearrangement in the BCL6 gene. The 46 cases with no rearrangement in the *c-MYC* gene were classified as DLBCL NOS/HGBL NOS.

In our series, all patients received chemotherapy based on the RCHOP protocol as a first line of treatment. Response to treatment was assessed after eight cycles of chemotherapy. Treatment was therefore completed for 30 patients (60%), who achieved complete remission, while for patients who had a partial remission/progression/relapse or stable disease, treatment was supplemented with a second line of chemotherapy using the RDHAOX protocol in the majority of cases; these patients represented 30% of the cases (15 patients). The median age of patients who received the RDHAOX chemotherapy protocol was 60 years (from 51 to 68 years). The remaining five patients (10%) died before continuing their cycles of chemotherapy.

In our cohort, the median of overall survival was 40.5 months (range: 0.25–216 months), and the median of event-free survival was estimated at 30 months (range: 0.25–110 months) (Figure 4).

The sociodemographic, clinical, morphological, immunohistochemical and cytogenetic data are shown in Table 1.

In addition to DHL/THL, we found three cases harboring only one rearrangement without *c-MYC* rearrangement: two cases with *BCL2* rearranged and one case with *BCL6* rearranged.

The first case with *BCL2* rearranged was a 70-year-old male patient who had elevated Ann Arbor stage (IV), high IPI score (4) and high LDH level. The localization of his tumor was primarily intranodal, with secondary pulmonary and bone localizations. This patient had a GCB subtype and a *Ki67* expression at 80%. He received eight cycles of RCHOP chemotherapy, and a re-assessment CT scan showed a progression of his pathology. A second-line treatment was prescribed, but his state of health deteriorated severely, and he refused to continue treatment. One month later, the patient died. Thus, the patient's overall survival was 18 months, with an estimated event-free survival of 7 months.

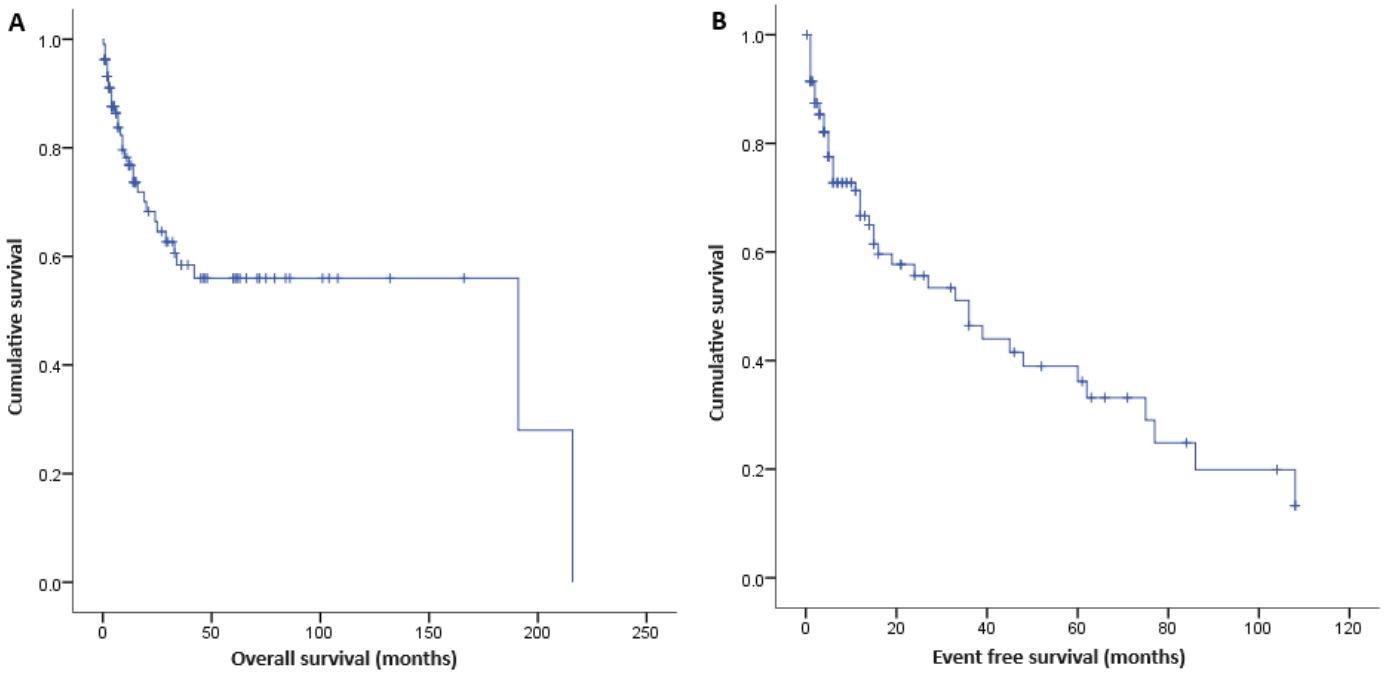

Figure 4. (**A**) Kaplan–Meier overall survival for all patients, (**B**) Kaplan–Meier event-free survival for all patients.

Table 1. Sociodemographic, clinical, immunohistochemical and cytogenetic data of the diagnostic groups after FISH testing.

	Diagnostic Groups after FISH Testing		
	THL (n = 1) n (%)	**DHL (n = 3) n (%)**	**DLBCL NOS/HGBL NOS (n = 46) n (%)**
Sociodemographic data:			
Female	0 (0%)	3 (100%)	21 (45.65%)
Male	1 (100%)	0 (0%)	25 (54.35%)
Aged over 60 years	1 (100%)	1 (33%)	18 (39.13%)
Prognostic markers:			
B symptoms	1 (100%)	1 (33%)	34 (73.91%)
High OMS index (3 or 4)	1 (100%)	0 (0%)	7 (15.22%)
High LDH level	1 (100%)	3 (100%)	40 (86.95%)
High Ann Arbor stage (III/IV)	1 (100%)	2 (67%)	37 (80.43%)
High IPI score (>2)	1 (100%)	2 (67%)	25 (54.34%)
Extranodal involvement	1 (100%)	2 (67%)	32 (69.56%)
COO classification:			
GCB subtype	0 (0%)	0 (0%)	14 (34.15%)
Non-GCB subtype	1 (100%)	3 (100%)	27 (65.85%)
Ki 67 > 90%	1 (100%)	1 (33%)	12 (26.09%)
Ki 67 < 90%	0 (0%)	2 (67%)	34 (73.91%)
Cytogenetic test:			
c-MYC R	1 (100%)	3 (100%)	0 (0%)
BCL2 R	1 (100%)	0	2 (4.35%)
BCL6 R	1 (100%)	3 (100%)	1 (2.17%)

Table 1. *Cont.*

	Diagnostic Groups after FISH Testing		
	THL (n = 1) n (%)	**DHL (n = 3) n (%)**	**DLBCL NOS/HGBL NOS (n = 46) n (%)**
Response to treatment:			
Complete remission	0 (0%)	2 (67%)	28 (60.87%)
Partial remission	1 (100%)	1 (33%)	10 (21.74%)
Stable disease	0 (0%)	0 (0%)	2 (4.35%)
Progression	0 (0%)	0 (0%)	6 (13.04%)
Relapse	0 (0%)	2 (67%)	5 (10.86%)
Survival:			
OS (median)	5 months	31 months	40.5 months
EFS (median)	5 months	19 months	30 months

The second case with *BCL2* rearranged was a 51-year-old female who had high Ann Arbor stage (IV), high IPI score (4) and high LDH level. Her tumor was located primarily in the cavum, with secondary lymph node and bone localizations. Immunohistochemistry confirmed a diagnosis of HGBL, with high expression of Ki67 (95%). The patient died before starting chemotherapy. The OS and EFS for this patient was 2 months.

The third case with *BCL6* rearranged was an 80-year-old female patient with high Ann Arbor stage (III), high IPI score (3) and high LDH level. The localization of her tumor was intranodal, with cervical and inguinal adenopathies without extranodal involvement. The immunohistochemistry test showed that it was a non-GCB subtype, with an expression of *Ki67* at 70%. The patient received chemotherapy and Rituximab. A re-evaluation CT scan showed complete tumor regression. The patient had an overall survival of 22 months, and she is still alive.

For DHL and THL patients, the sociodemographic, clinical, immunophenotypic and outcome data are presented in Table 2.

Table 2. Clinical, immunophenotypic and outcome data of patients with HGBL/DHIT-THIT.

	DHL (*c-MYC*/*BCL6*) Case 1	**DHL (*c-MYC*/*BCL6*) Case 2**	**DHL (*c-MYC*/*BCL6*) Case 3**	**THL (*c-MYC*/*BCL2*/*BCL6*) Case 4**
Age (years)	46	48	75	92
Sex	F	F	F	M
Ann Arbor stage	II	IV	IV	IV
IPI score	0	4	3	4
LDH level	High	High	High	High
Intranodal localizations	Cervical poly-adenopathies	Cervical, mediastinal, abdominal, iliac and inguinal adenopathies	Cervical, abdominal and iliac adenopathies	Cervical adenopathies
Extranodal localizations	Absent	Digestive	Cavum and amygdale	Orbit, upper lip and tongue
Ki67	70%	90%	80%	95%
COO	Non-GCB	Non-GCB	Non-GCB	Non-GCB
Response to treatment	Complete response	Stable disease after first line of treatment Complete response after second line of treatment	Relapse	Death
Status	Still alive	Died	Died	Died
OS	92 months	19 months	31 months	5 months
EFS	92 months	19 months	6 months	5 months

4. Discussion

The aim of our study was to perform a molecular classification of DLBCL and HGBL patients in order to evaluate the prognosis of molecular subtypes and to compare it with previous studies. The issue with this classification is the incidence of DHL and THL subtypes, which do not exceed 10% in the majority of studies (Table 3).

c-MYC rearrangement was detected in 8% of our cases, a proportion approximately comparable with previously published data [7,8,15].

In our series, all cases of *c-MYC* rearrangement were of the non-GCB subtype. However, in other series, *c-MYC* rearrangement was more frequent in GCB DLBCLs compared with non-GCB DLBCLs [16].

BCL2 and *BCL6* rearrangements were detected in 6% and 10% of cases, respectively. In other series, *BCL2* and *BCL6* rearrangements were more frequent in comparison to our results [7,15]. Similarly to *c-MYC* rearrangements, *BCL2* rearrangements were more frequent in the GCB subtype, in contrast to *BCL6* rearrangement, which was more frequent in the non-GCB subtype in some studies. In other studies, *BCL6* rearrangements were present with the same frequency in both subgroups. In our series, all *BCL6* rearrangements were of the non-GCB subtype, and *BCL2* rearrangement was present in one case of the GCB subtype and two cases of the non-GCB subtype. Moreover, the two patients with rearrangement of the *BCL2* gene only had inferior outcomes in comparison to the patient with *BCL6* rearrangement, who had a better response to treatment and better OS. Several researchers have evaluated the predictive significance of *BCL2* or *BCL6* rearrangement in patients with DLBCL. The investigations have presented conflicting findings, indicating that these rearrangements may or may not have a prognostic influence in patients diagnosed with DLBCL [12,13,17].

HGBL-DHL and THL represented 8% of the cases in our series, which is concordant with other series [4,8,18]. All of these cases were of the non-GCB subtype. However, previous series have shown that the HGBL-DHL and THL subtypes are more likely associated with the GCB subtype [8]. Other series showed no significant difference between GCB and non-GCB subtypes regarding the molecular subtype of HGBL (DHL and THL) [7]. Moreover, patients with *c-MYC*-DHL that involved *BCL2* and those with *c-MYC*-THL almost exclusively fell into the GCB DLBCL subgroup in Rosenwald et al.'s study, whereas those with MYC-DHL that involved BCL6 were found in both COO subgroups [16].

In our series, all HGBL-DHL patients had *c-MYC*/*BCL6* rearrangement. DHL with *c-MYC*/*BCL2* rearrangements was slightly higher in comparison to DHL with *c-MYC*/*BCL6* rearrangements in previous series (Table 3). Other series found no cases of DHL with BCL2 rearrangement in the non-GCB subtype [7], which is comparable with our results.

Patients with DHL or THL in our series presented with aggressive clinical features: they all had an elevated LDH level, and 75% of them had a high Ann Arbor stage, high IPI score and extranodal involvement. These findings align with previous studies [19,20].

In our series, the DHL and THL cases were less responsive to chemotherapy in comparison to DLBCL NOS/HGBL NOS cases; complete remission was obtained in only one case of DHL who had favorable tumor characteristics (low Ann Arbor stage, low IPI score and no extranodal involvement). Except for the first case of DHL, all cases had an inferior OS in comparison to the median of OS in DLBCL NOS/HGBL NOS cases. These results allow us to conclude that DHL/THL are aggressive and have worse outcomes than DLBCL NOS/HGBL NOS in the Moroccan context. Several previous studies have shown worse outcomes for DHL/THL (Table 3).

The prognostic value of *c-MYC* gene rearrangement has been established by many studies. *c-MYC* rearrangement was associated with an inferior OS and EFS in several studies [3,4,8,16]. Similarly, DHL and THL have been associated with poor prognosis after standard RCHOP chemotherapy (Table 3). More precisely, the combination of *c-MYC* and *BCL2* rearrangements affects the outcome. When combined together, these two genes have a synergistic clinical effect: *c-MYC* as a cellular proliferation regulator and *BCL2* as a blocker of programmed cell death and apoptosis [9]. On the other hand, the prognostic

implication of DHL with *c-MYC* and *BCL6* rearrangements is controversial [3,7]. Therefore, a study conducted by Rosenwald et al. investigated 2383 respondents who received RCHOP chemotherapy. The study found that patients who had the *c-MYC* rearrangement associated with *BCL2* and/or *BCL6* rearrangements had lower progression-free survival (PFS) and overall survival (OS) [16]. A retrospective analysis conducted by Laude et al. included 160 patients with HGBL (81% DHL and 19% THL). The study found that patients who received intense chemotherapy had a significantly better PFS compared to those treated with RCHOP [21]. Another study conducted by Zeremski et al. indicated that intensified regimens could possibly improve 2-year OS and 2-year PFS in HGBL-DHL/THL patients [22]. DHL and THL are therefore considered aggressive lymphomas, which are less responsive to standard chemotherapy protocols; these entities require a more intensive therapeutic approach or a more personalized treatment based on the molecular subtype. Patients with these lymphomas should be involved in clinical trials, the subject of which is the examination of targeted therapies supporting the key mechanism of pathogenesis of *c-MYC* and *BCL2* activation. Intensive chemotherapy should be given to patients who are appropriate for this treatment. A personalized approach should be adopted considering every molecular subtype in order to improve the prognosis for these patients.

Table 3. Incidence and outcome for DHL/THL lymphomas in other studies.

	N	DHL (*c-MYC-BCL2*)	DHL (*c-MYC-BCL6*)	THL	All DHL and THL Cases	Poor Outcome for DHL/THL Lymphoma Cases
S. Barrans et al. [4]	303	8%	1%	3%	12%	Yes
E. C. Obermann, M. Csato et al. [2]	333	0.45%	0.45%	0%	≈1%	Yes
S. O. Yoon et al. [23]	186	1%	1%	1%	3%	Yes
M. G. Tibiletti et al. [24]	74	7%	7%	1%	12%	Yes
Q. Ye et al. [7]	898	2.8%	2.9%	NA	NA	Yes
A. Tzankov et al. [25]	563	1.42%	0.71%	0.35%	2.49%	Yes
N. A. Johnson et al. [15]	167	8.38%	NA	NA	NA	Yes
C. Visco et al. [26]	327	2.45%	NA	NA	NA	Yes
N. Akyurek et al. [27]	239	1.67%	0.84%	0.42%	2.93%	Yes
Mostafa M. Amer et al. [28]	30	10%	0%	0%	NA	Yes
S. Huang et al. [29]	130	3%	3%	1.5%	7.69%	Yes
C. C. Oliveira et al. [30]	120	1.6%	0.8%	0%	NA	Yes
Our series	50	0%	6%	2%	8%	Yes

Limitations: The limitation of our study is the small sample size, which does not allow us to correlate the various clinicopathological factors of the molecular subtypes with patient prognosis. This is due to the unavailability of the FISH kit to expand the sample size.

5. Conclusions

Based on the analysis of the prognostic value of rearrangements in the *c-MYC*, *BCL2* and *BCL6* genes and the relationship between the molecular subtypes of HGBL and DLBCL and outcomes in previous studies and our own, we can conclude that DHL/THL- HGBL are an aggressive entity with poor outcomes in comparison to DLBCL NOS/HGBL NOS cases. Therefore, it is crucial to perform additional research on the fundamental physiological processes that contribute to this specific category of lymphoproliferative diseases as well as to comprehend the interaction between genetic alterations and the development of lymphomas in order to develop more targeted therapies for each molecular subtype. In this

context, several current research projects show promise, particularly those focusing on the *c-MYC* and *BCL2* genes. Recent studies have shown that it is crucial to use new treatment approaches, such as the newly approved anti-*CD19* monoclonal antibodies and chimeric antigen receptor (CAR) T cells, for patients with high-risk NHL DLBCL and HGBL who do not respond completely to previous treatments.

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Informed Consent Statement: The authors certify that they obtained written informed consent forms from the patients' families for the publication of this article.

Data Availability Statement: The data presented in this study are available on request from the corresponding author.

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