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– ORIGINAL PAPER –

Real-World Outcomes and Morphological Patterns of Aggressive Non-Hodgkin Lymphoma in Moldova: A Five-Year Retrospective Cohort Study

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Abstract

Objectives: *The addition of rituximab to CHOP chemotherapy has significantly improved outcomes in aggressive non-Hodgkin lymphoma (ANHL). However, data from resource-constrained healthcare systems remain limited. This study aimed to evaluate the clinical characteristics and treatment outcomes of ANHL in the Republic of Moldova.*

Materials and Methods*: This retrospective cohort study included 54 patients diagnosed with ANHL and treated at the Oncological Institute of Moldova between 2018 and 2022. Disease classification followed the 2022 World Health Organization criteria. Overall survival (OS) was assessed using the Kaplan–Meier method, and differences between treatment groups were evaluated using the log-rank test.*

Outcomes: *The median age was 57.4 years. Diffuse large B-cell lymphoma was the most frequent subtype (64.8%), followed by lymphoblastic lymphoma (29.6%). Rituximab-based regimens were associated with higher complete remission rates compared with CHOP alone. Median OS was longer in the R-CHOP group (32 months) than in the CHOP group (17 months), with a statistically significant difference (log-rank p = 0.004).*

Conclusions*: Rituximab-based therapy was associated with improved treatment response and survival in this cohort. These findings support the use of standard immunochemotherapy in resource-limited settings while highlighting the need to strengthen diagnostic infrastructure and access to advanced disease characterization.*

Keywords: *Lymphoma, Non-Hodgkin; Lymphoma, Large B-Cell, Diffuse; Antineoplastic Combined Chemotherapy Protocols; Rituximab; Survival Analysis; Moldova; Healthcare Disparities.*

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Introduction

Aggressive non-Hodgkin lymphomas (ANHLs) represent a heterogeneous group of lymphoid malignancies characterized by rapid progression and the need for prompt, intensive treatment [1]. Diffuse large B-cell lymphoma (DLBCL) is the most common aggressive subtype, with well-established diagnostic and therapeutic approaches described in international guidelines and contemporary reviews [2–4]. The incorporation of the

anti-CD20 monoclonal antibody rituximab into the cyclophosphamide, doxorubicin, vincristine, and prednisone (CHOP) regimen has significantly improved progression-free and overall survival, and remains the standard of care [5,6].

However, the translation of these outcomes into routine clinical practice is variable, particularly in resource-constrained settings where disparities in diagnostic capacity, access to immunotherapy, and supportive care persist. In addition, survival outcomes and disease burden may vary across populations and healthcare systems [7,8].

In the Republic of Moldova, the management of ANHL is influenced by systemic limitations in healthcare infrastructure. Although international standards recommend advanced molecular profiling and PET–CT–guided response assessment, access to these modalities remains limited. Furthermore, lymphoma biology is heterogeneous, and emerging insights into molecular subtypes continue to refine disease classification and risk stratification [9].

Recent updates in lymphoma classification, including the 5th edition of the World Health Organization classification, emphasize the importance of accurate morphological and molecular subtyping for risk-adapted therapy [1]. In settings with constrained diagnostic resources, understanding the local distribution of lymphoma subtypes is particularly important, as it may influence both treatment decisions and clinical outcomes.

This study addresses these gaps by evaluating the morphological distribution, clinical characteristics, and treatment outcomes of patients with ANHL treated at the Oncological Institute of Moldova. By comparing rituximab-based therapy with chemotherapy alone using survival analysis, we aim to provide real-world evidence on treatment outcomes and to characterize the clinicopathological profile of ANHL in this setting.

Materials and Methods

Study Design and Patient Selection

We conducted a retrospective, single-center cohort study including 54 adult patients diagnosed with aggressive non-Hodgkin lymphoma (ANHL) and treated at the Oncological Institute of the Republic of Moldova between January 2018 and April 2022. Inclusion criteria were: (1) histologically confirmed ANHL; (2) age ≥18 years; and (3) availability of complete clinical and follow-up data.

The study was conducted in accordance with the Declaration of Helsinki [10] and approved by the Ethics Committee of Nicolae Testemitanu State University of Medicine and Pharmacy. As this was a retrospective study based on the review of the existing medical records, individual informed consent was waived.

Diagnostic Procedures and Classification

Morphological classification was performed according to the 2022 World Health Organization (WHO) Classification of Tumours of Haematopoietic and Lymphoid Tissues. Diagnosis was established based on histopathological examination, with immunohistochemistry (IHC) performed when available. Clinical staging was assessed using the Ann Arbor system and supported by computed tomography (CT) of the chest, abdomen, and pelvis. Bone marrow involvement was evaluated by trephine biopsy.

Treatment Protocols

Patients were allocated to treatment groups based on clinical indications and treatment availability. The administered regimens included: (1) CHOP (cyclophosphamide, doxorubicin, vincristine, and prednisone); (2) R-CHOP (CHOP plus rituximab); (3) CHOP followed by consolidative radiotherapy (RT); and (4) R-CHOP followed by consolidative radiotherapy.

Rituximab was administered intravenously at a standard dose of 375 mg/m² per cycle. Radiotherapy was delivered as consolidative treatment based on clinical indications and was not standardized across patients, which may have introduced treatment selection bias.

Statistical Analysis

Overall survival (OS) was defined as the time from diagnosis to death from any cause or last follow-up. Survival analysis was performed using the Kaplan–Meier method, and differences between groups were assessed using the log-rank (Mantel–Haenszel) test.

Given the limited sample size, age was analyzed as a dichotomous variable (≤60 vs >60 years) in the primary survival analysis to improve statistical power. Additionally, age was evaluated as a categorical variable (18–39, 40–64, and ≥65 years) for descriptive and exploratory analyses.

Associations between treatment regimen and response were evaluated using the chi-square test across all four treatment groups. Proportions were presented with corresponding 95% confidence intervals (CIs), calculated using the exact (Clopper–Pearson) method based on binomial distribution. A two-sided p-value <0.05 was considered statistically significant.

Statistical analyses were performed using MedCalc software (MedCalc Software Ltd., Ostend, Belgium; accessed March 15, 2026). Confidence intervals for proportions were additionally verified using an independent online calculator (https://sample-size.net; accessed April 12, 2026). References were managed and formatted using Zotero (Corporation for Digital Scholarship, USA).

Results

Demographic and Environmental Characteristics

A total of 54 patients diagnosed with aggressive non-Hodgkin lymphoma between 2018 and 2022 were included. The majority of patients were aged 40–64 years (57.4%, 95% CI: 43.2–70.7; n=31), followed by those aged ≥65 years (28.0%, 95% CI: 16.4–41.6; n=15), while patients aged 18–39 years accounted for 14.8% (95% CI: 6.6–27.1; n=8). Overall, females were more frequently affected than males (55.6% vs. 44.4%) (Table 1).

Table 1. *Demographic characteristics of the study population by age group and sex.*

Age group (years)	Number of patients	Frequency (%)	Males, n (%)	Females, n (%)
18-39	8	14.8	5 (62.5)	3 (37.5)
40-64	31	57.4	13 (42.0)	18 (58.0)
>65	15	28.0	6 (40.0)	9 (60.0)
Total	54	100.0	24 (44.4)	30 (55.6)

Abbreviations: *CI, confidence interval; n, number of patients. Percentages are presented within each age group. Exact binomial 95% confidence intervals were calculated.*

Regarding place of residence, most patients originated from rural areas (76.0%, 95% CI: 62.3–86.5; n=41), while 24.0% (95% CI: 13.4–37.6; n=13) were from urban settings (Table 2).

Age-stratified analysis showed that patients aged 40–64 years were predominantly from rural areas (83.8%, 95% CI: 66.2–94.5; n=26), whereas only 16.2% (95% CI: 5.4–33.7; n=5) were from urban settings. A similar pattern was observed among patients aged ≥65 years (73.4%, 95% CI: 44.9–92.2 vs. 26.6%, 95% CI: 7.7–55.1 for rural and urban areas, respectively). In contrast, patients aged 18–39 years were equally distributed between rural and urban environments (50.0% each; 95% CI: 15.7–84.3) (Table 2).

Table 2. *Demographic and residential distribution of patients with aggressive non-Hodgkin lymphomas.*

Age group (years)	Number of patients	Living environment	
		Rural, n (%)	Urban, n (%)
18-39	8	4 (50.0)	4 (50.0)
40-64	31	26 (83.8)	5 (16.2)
>65	15	11 (73.4)	4 (26.6)
Total	54	41 (76.0)	13 (24.0)

Abbreviations: *CI, confidence interval; n, number of patients. Percentages are presented within each age group. Exact binomial 95% confidence intervals were calculated.*

Immunohistochemical Variants and Disease Distribution

The distribution of immunohistochemical subtypes by age group is presented in Table 3. Overall, diffuse large B-cell lymphoma (DLBCL) was the most frequent subtype, accounting for 64.8% (95% CI: 50.6–77.3; n=35). It predominated in patients aged 40–64 years (67.7%, 95% CI: 48.6–83.3; n=21) and ≥65 years (80.0%, 95% CI: 51.9–95.6; n=12).

In patients aged 18–39 years, diffuse large B-cell lymphoma (DLBCL) was the most common subtype (62.5%, 95% CI: 24.4–91.4; n=5), followed by

lymphoblastic lymphoma (25.0%, 95% CI: 3.2–65.0; n=2).
Mantle cell lymphoma (3.7%, 95% CI: 0.45–12.7; n=2) and anaplastic lymphoma (1.8%, 95% CI: 0.05–9.8; n=1) were infrequent and observed only in isolated cases.

A statistically significant association was observed between age group and morphological subtype (χ^2 = 13.30, df = 6, p = 0.038).

Table 3. *Distribution of patients by age group and morphological subtype.*

Age group (years)	Number of patients	Morphologic variant			
		DLBCL, n (%)	Lymphoblastic lymphoma, n (%)	Mantle cell lymphoma, n (%)	Anaplastic lymphoma, n (%)
18-39	8	5 (62.5)	2 (25.0)	0	1 (12.5)
40-64	31	21 (67.7)	8 (25.8)	2 (6.4)	0
>65	15	12 (80.0)	3 (20.0)	0	0
Total	54	35 (64.8)	16 (29.6)	2 (3.7)	1 (1.8)

Abbreviations: *CI, confidence interval; n, number of patients. Percentages are presented within each age group. Exact binomial 95% confidence intervals were calculated.*

Clinical Staging

The distribution of clinical stage by age group is presented in Table 4. Advanced-stage disease (stage III–IV) predominated, with stage IV being the most frequent stage (53.7%, 95% CI: 39.6–67.3; n=29).
In patients aged 40–64 years, stage IV disease was the most common (51.6%, 95% CI: 33.1–69.8; n=16), followed by stage II (25.8%, 95% CI: 11.8–44.6; n=8).

Among patients aged ≥65 years, stage IV predominated (66.6%, 95% CI: 38.3–88.1; n=10). In the 18–39 year age group, stage distribution was more balanced, although stage IV remained the most frequent (37.5%, 95% CI: 8.5–75.5; n=3).
Overall, early-stage disease (stage I–II) was less common than advanced-stage disease.

Table 4. *Distribution of patients by age group and disease stage.*

Age group (years)	Number of patients	Disease stage			
		I n (%)	II n (%)	III n (%)	IV n (%)
18-39	8	2 (25.0)	2 (25.0)	1 (12.5)	3 (37.5)
40-64	31	4 (12.9)	8 (25.8)	3 (9.7)	16 (51.6)
>65	15	1 (6.7)	3 (20.0)	1 (6.7)	10 (66.6)
Total	54	7 (13.0)	13 (24.0)	5 (9.3)	29 (53.7)

Abbreviations: *CI, confidence interval; n, number of patients. Percentages are presented within each age group. Exact binomial 95% confidence intervals were calculated.*

Therapeutic Response and Survival Outcomes

Treatment response analysis demonstrated a statistically significant association between treatment regimen and response distribution (Table 5). The overall complete remission (CR) rate for the cohort was 63.0% (95% CI: 48.7–75.7; n=34).
Rituximab-containing regimens were associated with higher response rates compared with non-rituximab regimens. The R-CHOP+RT group demonstrated the highest CR rate (91.6%, 95% CI: 61.5–99.7; n=11), followed by R-CHOP (74.0%, 95% CI: 51.5–89.7; n=17). Among patients treated without rituximab, the CHOP+RT group achieved a CR rate of 50.0% (95% CI: 15.7–84.3; n=4), whereas CHOP alone had the lowest CR rate (18.2%, 95% CI: 2.2–51.7; n=2).

A statistically significant association was observed between treatment regimen and response distribution ($\chi^2 = 19.56$, df = 6, p = 0.003).

Survival analysis according to treatment group demonstrated a statistically significant difference in overall survival between rituximab-based and non-rituximab regimens. Median overall survival was 32 months in the R-CHOP group compared with 17 months in the CHOP group (log-rank $\chi^2 = 8.51$, p = 0.004) (Figure 1).

***Table 5**. Treatment outcomes by regimen (overall).*

Regimen	Number of patients	Complete remission, n (%)	Partial remission, n (%)	No response/progressive disease, n (%)
CHOP	11	2 (18.2)	1 (9.1)	8 (72.7)
R-CHOP	23	17 (74.0)	1 (4.3)	5 (21.7)
CHOP-RT	8	4 (50.0)	2 (25.0)	2 (25.0)
R-CHOP-RT	12	11 (91.6)	0	1 (8.3)
Total	54	34 (63.0)	4 (7.4)	16 (29.6)

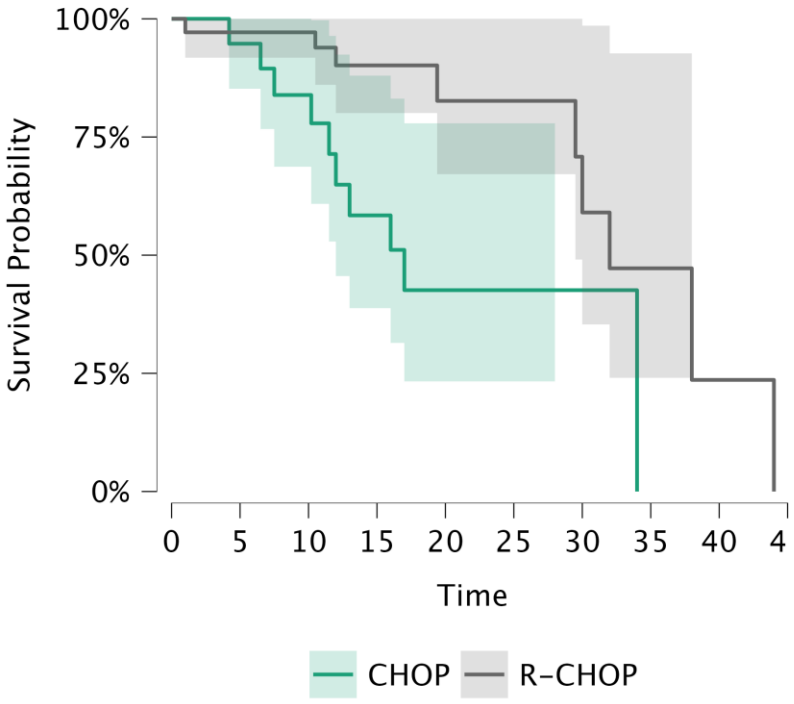


***Figure 1.** Kaplan–Meier overall survival curves according to treatment group*

***Legend:** Kaplan–Meier curves showing overall survival in patients treated with CHOP versus rituximab-based regimens (R-CHOP). Patients were grouped irrespective of the use of consolidative radiotherapy due to the limited sample size. Median overall survival was 17 months in the CHOP group and 32 months in the R-CHOP group. The difference between groups was statistically significant (log-rank, $\chi^2 = 8.51$, $p = 0.004$). Shaded areas represent 95% confidence intervals.*

Survival analysis stratified by age (≤60 vs >60 years) showed a trend toward improved survival in younger patients. Median overall survival was 34.0 months in patients aged ≤60 years compared with 29.5 months in those aged >60 years. However, this difference was not statistically significant (log-rank $\chi^2 = 0.71$, p = 0.399) (Figure 2).

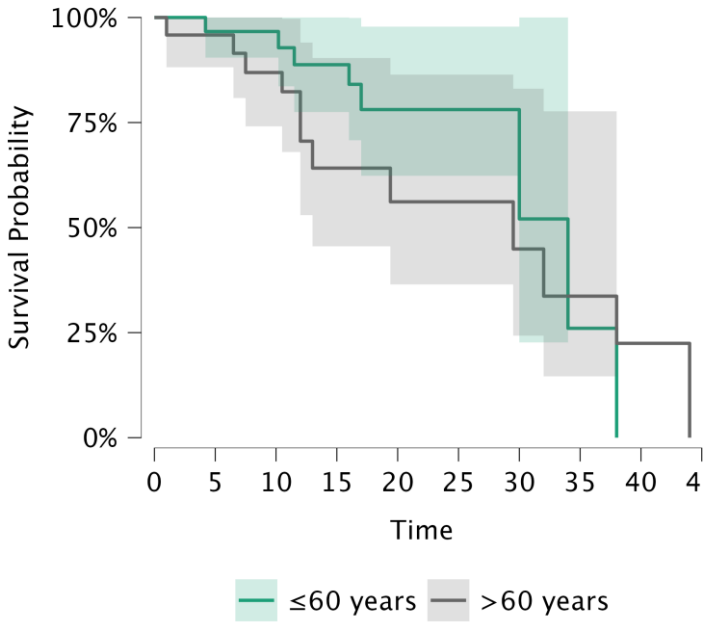
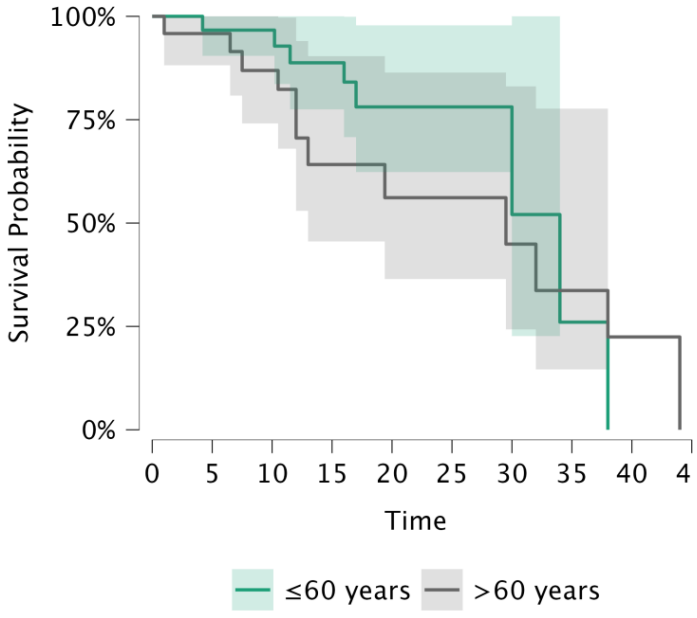


Figure 2. *Kaplan–Meier overall survival curves stratified by age group (≤60 vs >60 years).*
Legend: *Patients aged ≤60 years showed a trend toward improved survival compared with those aged >60 years; however, the difference was not statistically significant (log-rank $\chi^2 = 0.71$, $p = 0.399$). Shaded areas represent 95% confidence intervals.*

Additional exploratory analysis using three age groups (18–39, 40–64, ≥65 years) did not demonstrate statistically significant differences in survival (log-rank $\chi^2 = 2.96$, df = 2, p = 0.228) (Supplementary Figure 1).



Supplementary Figure 1. *Kaplan–Meier overall survival curves by three age groups (18–39, 40–64, ≥65 years).*

Legend: *No statistically significant differences in overall survival were observed between age groups (log-rank $\chi^2 = 2.96$, df = 2, p = 0.228). Shaded areas represent 95% confidence intervals.*

Discussion

This retrospective study provides real-world data on aggressive non-Hodgkin lymphoma in the Republic of Moldova, a setting characterized by limited diagnostic resources and restricted access to advanced oncological care. The cohort demonstrated a predominance of diffuse large B-cell lymphoma (DLBCL), a high proportion of advanced-stage disease at diagnosis, and a majority of patients originating from rural areas. These findings reflect the clinical and healthcare context in which lymphoma is diagnosed and managed in this region.

Stage at presentation and healthcare access

A substantial proportion of patients presented with advanced-stage disease (stage III–IV), while early-stage disease was less common. Although causal relationships cannot be established in this study, this pattern may reflect delayed diagnosis and limited access to specialized oncological services — well-recognized challenges in resource-constrained healthcare systems [8,9]. The predominance of patients from rural areas further supports this interpretation, suggesting potential disparities in healthcare access. Similar patterns have been reported in comparable settings, where differences in diagnostic infrastructure and access to care may influence stage at presentation and overall disease burden [8,11]. Additionally, population-based analyses have demonstrated variability in survival outcomes across lymphoma subtypes and healthcare settings, reflecting differences in access to care and treatment delivery [8].

Therapeutic outcomes and the role of rituximab

The analysis demonstrated a statistically significant association between treatment regimen and response distribution, with higher complete remission rates observed in patients receiving rituximab-containing regimens. Survival analysis also showed improved overall survival in patients treated with R-CHOP compared with CHOP alone.

These findings are consistent with randomized clinical trials and long-term follow-up studies demonstrating improved outcomes with the addition of rituximab to CHOP chemotherapy [3,5,6,12]. However, given the observational design of this study, these results should be interpreted as associations rather than causal effects.

Survival and age-related patterns

In the present study, survival analysis stratified by age (≤60 vs >60 years) did not demonstrate statistically significant differences between groups. Although a non-significant trend toward improved survival in younger patients was observed, this did not reach statistical significance.

Similarly, exploratory analysis using three age groups (18–39, 40–64, ≥65 years) did not show statistically significant differences in survival. These findings may be influenced by the relatively small sample size and limited number of events, which reduce statistical power. Larger studies have identified age as a prognostic factor in aggressive lymphomas, but such effects may not be detectable in smaller cohorts [11,13].

Radiotherapy bias and interpretation of survival analysis

An important methodological consideration is the potential confounding effect of radiotherapy. Radiotherapy was administered based on clinical indication rather than a standardized protocol, introducing the possibility of treatment selection bias. Patients receiving radiotherapy may have differed in disease characteristics, including stage, localization, or response to therapy.

To address this limitation, survival analyses were performed by grouping patients into CHOP-based and R-CHOP-based categories irrespective of radiotherapy use. While this approach improves statistical robustness, it limits the ability to evaluate the independent effect of radiotherapy. Therefore, survival differences should primarily be interpreted in relation to rituximab-based treatment.

Challenges and future directions

Despite improved outcomes observed with rituximab-based therapy, important challenges remain. In high-resource settings, treatment strategies have expanded to include targeted therapies, CAR T-cell therapy, and advanced supportive care [11]. In contrast, in resource-limited settings, priorities may include improving early diagnosis, strengthening pathology infrastructure, and ensuring access to standard immunochemotherapy [11,14].

Limitations

This study has several limitations. Its retrospective design and single-center setting may introduce selection bias and limit generalizability. The relatively small sample size restricted subgroup analyses, including age-stratified survival comparisons.

Incomplete access to immunohistochemical and molecular diagnostics may have influenced subtype classification. Additionally, the independent effect of radiotherapy could not be evaluated due to non-standardized treatment allocation. These limitations should be considered when interpreting the findings.

Conclusions

Aggressive non-Hodgkin lymphoma remains a clinical challenge in resource-limited settings. In this Moldovan cohort, patients frequently presented with advanced-stage disease, and diffuse large B-cell lymphoma was the predominant subtype. Rituximab-based therapy was associated with higher response rates and improved survival compared with chemotherapy alone, supporting its role in first-line treatment.

However, survival findings should be interpreted with caution due to the limited sample size and the potential confounding effect of non-standardized radiotherapy use. Improving outcomes in this setting may depend on strengthening diagnostic capacity, expanding access to immunochemotherapy, and promoting earlier detection. Further studies incorporating larger cohorts and standardized diagnostic approaches are needed to better characterize regional disease patterns and inform clinical practice.

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A preliminary version of this study was previously published as an abstract in a conference proceedings volume. Differences in subtype distribution compared with the current manuscript reflect subsequent data verification, case inclusion updates, and refinement of morphological classification according to the latest WHO criteria.

Conflict of Interest Statement

We, the undersigned, certify that we do not have any financial or personal relationships that might bias the content of this work.

Ethics Approval and Consent to Participate:

This study was approved by the Ethics Committee of Nicolae Testemițanu State University of Medicine and Pharmacy (Protocol No. 78, approved on 16 September 2024). Due to the retrospective design, the requirement for informed consent was waived.

Author Contributions:

Artur Antosel: study conception and design, data collection, statistical analysis, and manuscript drafting.

Cristina Dudnic: study conception, data collection and analysis support.

Maria Robu: study design and critical revision of the manuscript.

Sanda Buruiana: methodological support and manuscript revision.

Victor Tomacinschii: clinical supervision and data interpretation.

All authors read and approved the final manuscript.

Artificial Intelligence Statement:

An AI-based language tool was used for grammatical and linguistic refinement of the manuscript. All scientific content, data analysis, interpretation, and conclusions are the sole responsibility of the authors.

Data Availability Statement:

The datasets generated and/or analyzed during the current study are not publicly available due to patient confidentiality and data protection regulations, but are available from the corresponding author upon reasonable request, subject to institutional approval.

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