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ORIGINAL PAPERS

Immunotherapy with Rituximab in Follicular Lymphomas

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ABSTRACT

Background: *Non-Hodgkin Lymphomas (NHL) represent a recent and fascinating domain of hemato-oncology, in which remarkable progress has been made. The conventional treatments of indolent lymphomas do not extend the survival rate, nor do they cure. Recent directions are centered on using several new drugs that are capable of overcoming the mechanisms that are resistant to recovery. The initiation of immunotherapy (Rituximab in 1997) seems to have changed the natural evolution of follicular lymphomas (FL). It is possible that resistance to healing in follicular lymphomas may be neutralized with Rituximab by suppressing STAT-1 positive macrophages that are present in the cellular microenvironment.Thereinafter, the re-evaluation of recent models of prognostic and therapeutic paradigmas that were used in FL became compulsory.*

The purpose of the paper *is to compare the evolution of patients with follicular lymphoma and the period of response, according to the treatments.*

Material and method: *The study group consisted of the 71 patients diagnosed with follicular lymphoma, out of a total of 767 malignant lymphatic proliferations with B cells, for a period of 7 years (2002-2008), at the Hematology Department, Hospital Coltea, Bucharest and Hematology Department, Universitary Hospital, Bucharest*

Results and conclusions: *Combining chemotherapy with Rituximab had better results compared to the same chemotherapy, administered alone, both in induction and in case of relapse. The overall response rate in our study group was 74.7%, out of which 42.3% complete remissions. The overall response rate was 84.61% in the Rituximab group, compared to 68.88% in patients without Rituximab.*

Keywords: follicular, treatment, immunotherapy

INTRODUCTION

In the last decade, passive immunotherapy with anti-CD20 monoclonal antibody, Rituximab, modified the therapeutic approach of B cell non-Hodgkin lymphoma (NHL) (1,2).

Conventional chemotherapy does not determine the healing of follicular lymphoma. Moreover, the absence of a plateau in the survival curve for this therapy, suggests that most patients will have a relapse of disease (3).

Combining chemotherapy with Rituximab has superior results to the same chemotherapy,

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but administered alone, so that the concomitant immuno - chemotherapy administration has become standard first-line therapy in follicular lymphoma, resulting in prolonged survival without relapse of disease and an improvement of overall survival (4,5).

The use of Rituximab (MabThera) in clinical practice was approved by the Food and Drug Administration in 1997, initially only for the treatment of indolent NHL relapse, especially follicular lymphoma (6).

It is currently the most widely used antibody in non-Hodgkin lymphoma, both in the induction therapy or the maintenance of disease remission and the relapses (6-8).

According to Alvaro T. (9,10) and Canioni D (11), STAT-1 positive macrophages are involved in suppressing the immune response of the host by T cell deletion. The presence of an increased number of these macrophages is of negative impact on the overall survival of patients with FL. Alvaro T. and collaborators have proved that the adverse effect of STAT-1 positive macrophages is inhibited by the association of Rituximab to chemotherapy, and thus overcoming the resistance to healing in follicular lymphomas. ❑

MATERIAL AND METHOD

The study group consisted of the 71 patients diagnosed with follicular lymphoma (FL), out of a total of 767 malignant lymphatic proliferations with B cells, for a period of 7 years (2002-2008), at the Hematology Hospital Coltea, Bucharest and Hematology Universitary Hospital, Bucharest.

The type of survey was retrospective and prospective, since diagnosis until death or completion of the study or loss of evidence. The main clinical and biological characteristics of patients with follicular lymphoma included in the study are presented in Table 1.

Characteristics	Patients
Age	60 years
Male/female	37/34
Urban/rural	52/19
Localized/advanced form	21/50
FL 1st/2nd/3rd degree	28/17/26
Adenopathy absent/<5cm/5-10 cm/>10cm	5/45/14/7
Hepatomegaly absent/present	27/44
Splenomegaly absent/present	42/29
Hemoglobine (g/dL) <7/7-10/10-12/>12	1/2/16/52
White Blood Cells (/µL)<4000/4000-9000/>9000	6/48/17
Platelets (/µL) <150000/150000-450000/>450000	8/60/3
ESR(mm/h) <60/60-100/>100	63/7/1
Fibrinogen (mg/dL) <400/400-600/>600	43/24/4
C reactive protein (mg/dL) <0.3/>0.3	41/30

TABLE 1. Clinical characteristics of the study group

Statistical analysis

The statistical analysis was performed on the personal computer, in SPSS 16.0 software for Windows. For the parameters or variables used in this paper the range, mean and standard deviation, coefficient of variation and standard error were presented. We used the Kolmogorov-Smirnov and Shapiro-Wilk tests, rejecting the assumption of normality was determined by values of $p < 0.05$. The determination of correlations between different studied variables was based on using the Pearson correlation coefficient in all cases where the variables had a normal distribution, and it was considered that a certain correlation is statistically significant if $p < 0.05$.

The objectives of our study were:

- Tracking the clinical course and response to treatment in follicular lymphoma.
- Comparing the evolution of patients and the duration of response, according to treatment. ❑

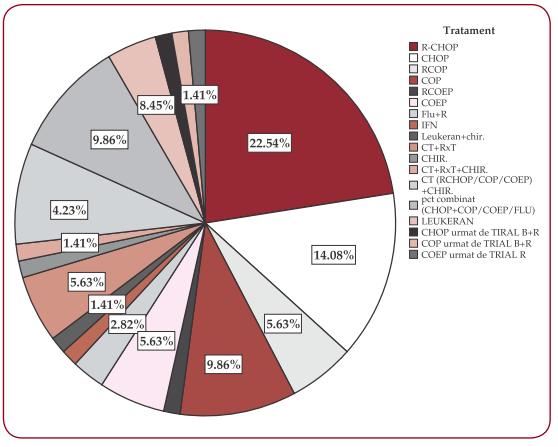

FIGURE 1. Distribution of patients with follicular lymphoma according to their treatment

R-Rituximab, C-Cyclophosphamide, H-Doxorubicine, O-Oncovin/Vincristine, P-Prednisone, E-Etoposide, Flu-Fludarabine, IFN-Interferone

RESULTS AND DISCUSSIONS

The therapy administered to patients with FL is shown in Figure 1. During the 7 years of follow up, immuno-chemotherapy was applied to 26 patients (36.1%), while "single" chemotherapy, without Rituximab was administered to 45 patients (63.3%). The most commonly used treatment chart was R-CHOP (16 cases, 22.5%). Three of the study group patients (4.2%) were included in the evaluation trial of proteosome inhibitor therapy (Bortezomib, B) with or without Rituximab. Other therapy methods were: radiotherapy combined with polychemotherapy, or radiotherapy + polychemotherapy + surgical treatment (partial or total removal of the tumour).

Evaluation of treatment response

Complete remission (CR) was obtained in 42.3% (30 patients) and partial remission in 32.4% (23 patients) (Figure 2). The overall response rate was 74.7% (53 patients), slightly increased in comparison to other groups described in specialty literature where the response rates have been 61-73%. A percentage of 25.4 (18 patients) did not respond to treatment or the disease progressed (non-responsive).

The comparative analysis of the type of response to treatment in patients with localized forms of FL and in advanced forms as well is shown in Figure 3. The share of complete remissions was two times higher in those with advanced forms (20 patients, 66.7%) compared to the localized forms of FL (10 patients, 33.4%). On the other hand, in the case of non-responders also, the share of advanced forms of FL was double (12 patients, 66.7%) compared to the localized forms of disease (6 patients, 33.4%). Therefore, the stage of the disease did not influence the response to treatment in patients with follicular lymphoma.

We analyzed the influence of early decrease in tumour mass on treatment response and found that the highest rate of complete remission was obtained in patients who showed a decrease of > 50% in tumour mass after the first treatment.

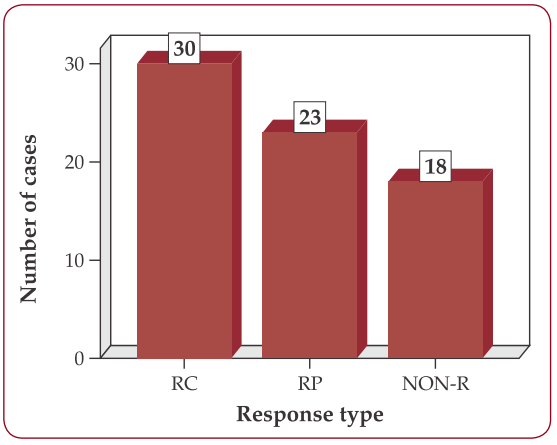

FIGURE 2. The distribution of response to treatment

RC-Complete response, RP-Partial response, NON-R-No response

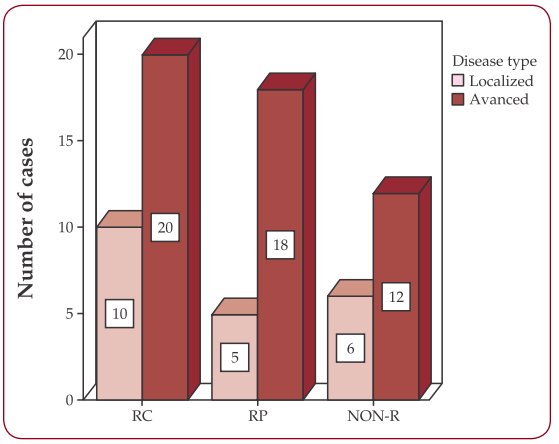

FIGURE 3. The distribution of pacients with FL according to the type of response to treatment, and according to the disease type - localized and avanced

RC-Complete response, RP-Partial response, NON-R-No response

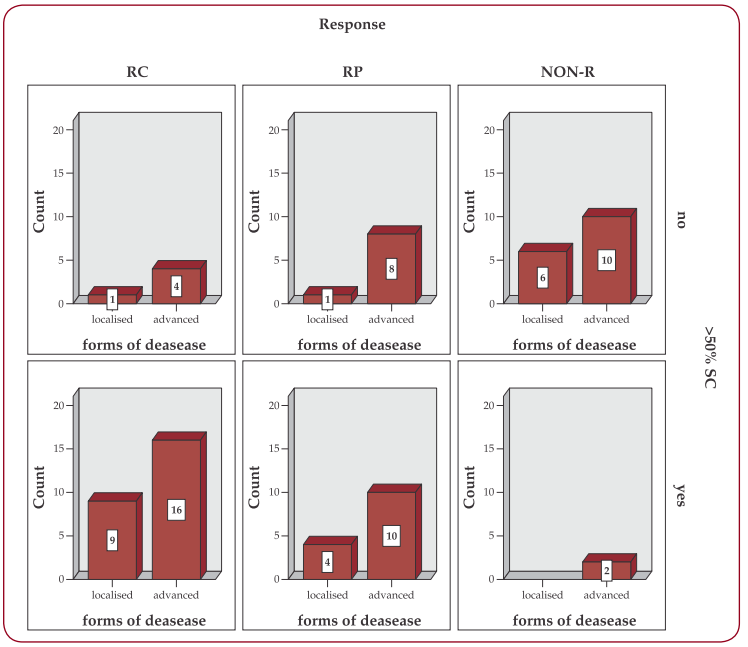

FIGURE 4. The influence of decreasing tumour mass and that of the form of disease on treatment response

RC-Complete response, RP-Partial response, NON-R-No response

A percentage of 83.3 (25 patients) of those with a complete response showed an early decrease in tumour mass, compared to only 16.6% (5 patients) with complete response, who did not show a decrease in tumour mass of > 50% after the first treatment (Figure 4), the ratio being 5:1.

Most non-responders came from the group that did not obtain an early response to therapy (16 cases, 88.9%) - compared to only 2 cases (11.2%) with no remission even though they presented an early reduction of tumour mass.

The comparative analysis of the type of response to treatment in patients with FL who received immuno-chemotherapy and those who received "simple" chemotherapy is represented in Figure 5. In the group of patients who have received immuno-chemotherapy 53.85% (14 patients) achieved complete remission, unlike the group of patients without Rituximab, in which complete remission was obtained in 35.55% (16 cases). Although the group of patients who received immuno-chemotherapy is much smaller compared to the group of patients with conventional chemotherapy, the percentage of those with CR is 1.5 times higher in the case of immuno-chemotherapy. Also, the percentage of non-responders is significantly higher in patients who did not benefit from Rituximab: 31.11% compared to 15.38% in patients with Rituximab.

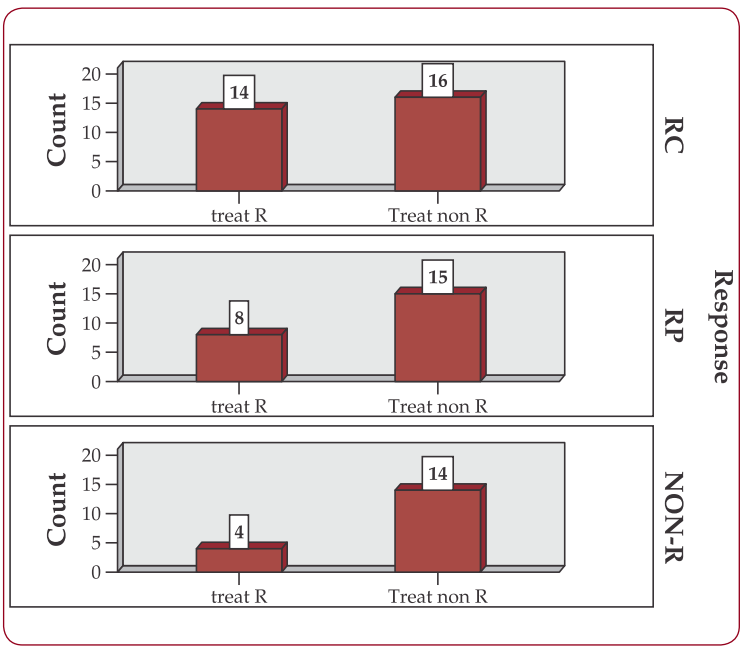

FIGURE 5. The influence of Rituximab(R) therapy on treatment response

RC-Complete response, RP-Partial response, NON-R-No response

The overall response rate was 84.61% (22 cases) in the Rituximab group, compared to 68.88% (31 cases) in patients without Rituximab.

The average duration of response to induction treatment was 20.25 +/- 26.328 months, with limits between 4 -164 months (Figure 6).

Disease relapse

The relapse rate in patients with follicular lymphoma was 41.5% (22 patients), out of which 6 patients (11.3%) showed histological conversion to diffuse large cell NHL.

Although the overall response rate to induction therapy was high (74.7%), it is noted that almost half of those who responded to the initial therapy, showed relapses of the disease, indicating that although they initially respond very well, relapses in FL patients are frequent.

The comparative analysis of disease relapse, according to treatment (with or without an association with Rituximab), is shown in Figure 7.

Prolonged remission of disease (more than 24 months) was almost double (31%) in patients who received immuno-chemotherapy, compared to patients who received simple chemotherapy (16%).

An early relapse of disease (less than 12 months) was present only in the group that did not receive immunotherapy (16% vs. 0%).

The percentage of non-responders was double (31%) in the group of patients with FL who did not receive immunotherapy, compared to the group of patients with FL who received chemotherapy associated with Rituximab (16%).

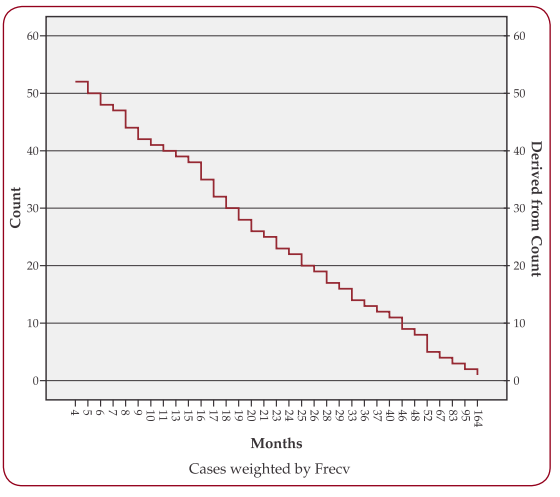

FIGURE 6. Period of remission

The disease relapse was strongly influenced by:

- a reduction of tumour mass with > 50% after the first course (p <0.001). In patients with significant response after the first course, the relapse rate was much lower.
- the type of response (complete, partial, non-responders) (p = 0.03). In case of a complete response, the duration of the remission was longer.

The relapse with transformation into aggressive lymphoma was present only in those with a partial response and in none of the patients with a complete response.

In conclusion, both in induction and in the case of relapse, the association of chemotherapy and Rituximab had superior results to the same chemotherapy, but administered alone.

The response to treatment correlated with the other studied parameters

We tried to determine the influence of various clinical and biological variables on the response to treatment.

- The factor with the highest predictive value for obtaining complete remission was the risk group which the patient entered at diagnosis, FLIPI (Follicular Lymphoma International Prognostic Index), p < 0.001. In the study group most patients who achieved complete response, were part of the low risk group at diagnosis, FLIPI =0-1 (Figure 8).
- The next factor in decreasing order of statistical significance was the International Prognostic Index score IPI (p <0.01). Most of the patients who achieved complete response, were part of the low risk group at diagnosis (IPI=0-1). No patient with CR was part of the high risk group (IPI=4-5). On the other hand, most non-responsive patients, presented at diagnosis an IPI ≥3 (Figure 9).
- The response to treatment is significantly directly correlated with the performance status of the patients (p <0.01), with the same intensity as IPI. Most of the patients who achieved complete response were asymptomatic at diagnosis (ECOG = 0). On the contrary, in the non-responders group, half of the patients had

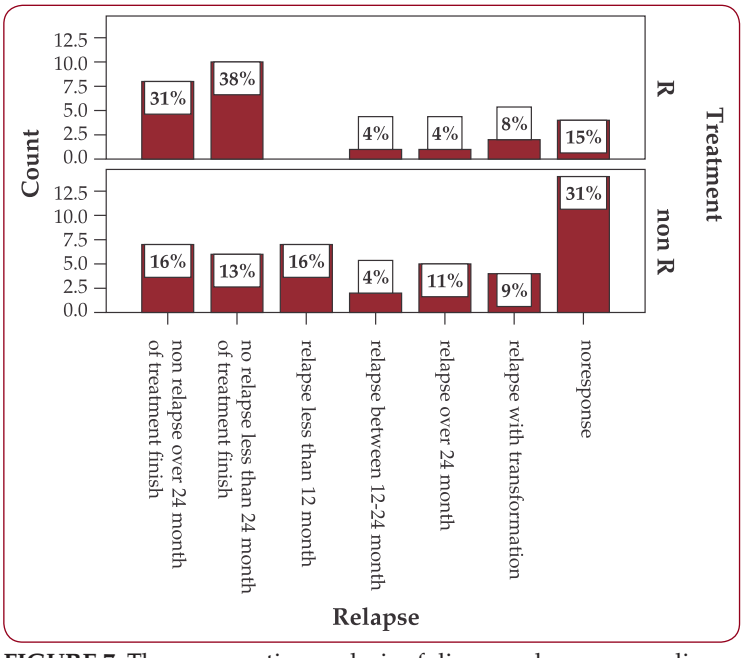

FIGURE 7. The comparative analysis of disease relapses, according to the treatment

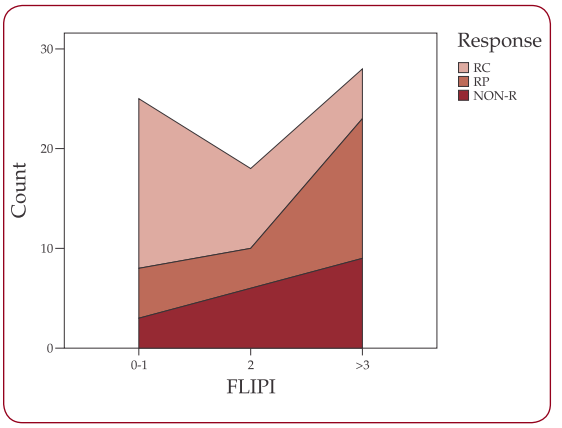

FIGURE 8. The correlation between the type of response and FLIPI

RC-Complete response, RP-Partial response, NON-R-No response

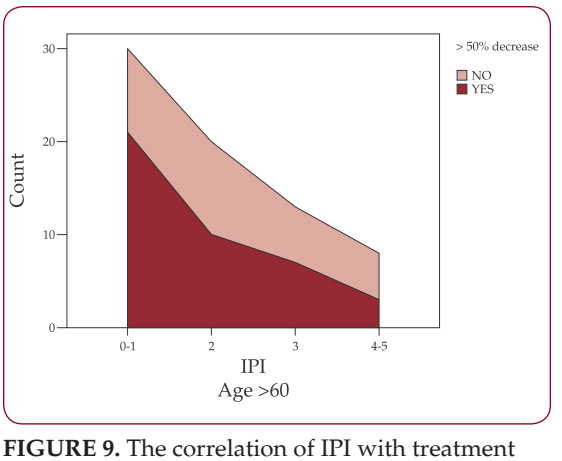

FIGURE 9. The correlation of IPI with treatment response

a poor performance status in the beginning, ECOG ≥2 (Figure 10).
- The location (superficial or deep) and dimensions of adenopathies influence the response to treatment. The largest number of complete remissions was obtained in patients with superficial lymph nodes and smaller than 5 cm.
- Most patients who showed a tumour decrease of more than 50% after the first course obtained complete remission.
- The response rate to treatment was correlated with the serum LDH level and the presence of thrombocytosis was associated with an unsatisfactory response.
- There is a statistically nonsignificant correlation between the response to treatment and the degree of hepatomegaly (p= 0.296), and the presence of medullary damage (p= 0.208).

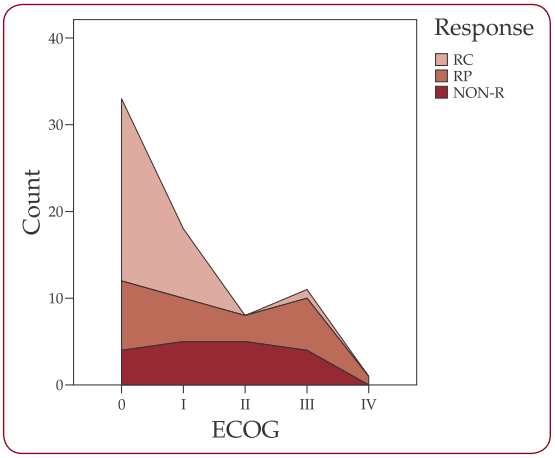

FIGURE 10. The correlation ECOG - response to treatment

RC-Complete response, RP-Partial response, NON-R-No response

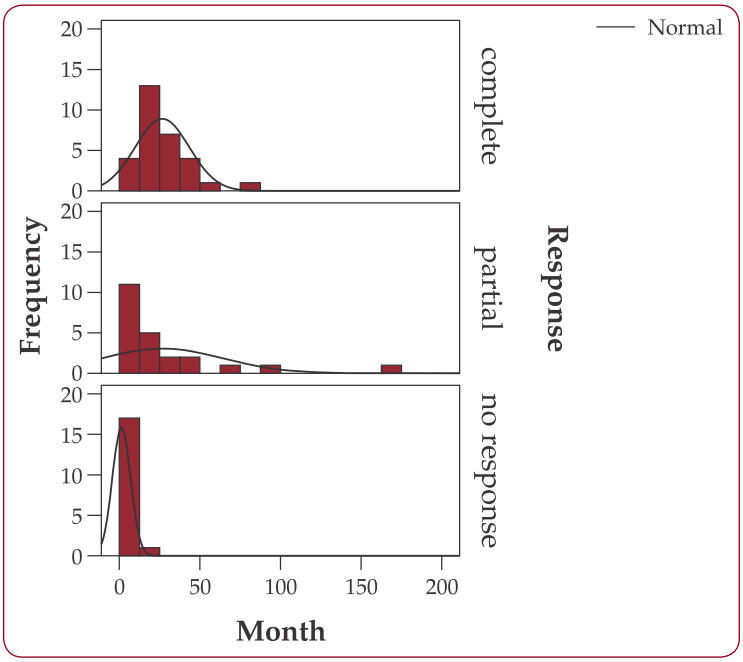

FIGURE 11. The correlation of the remission period with the type of response

Evaluation of treatment response
- The factors with the highest predictive value for a prolonged remission were getting a complete remission after induction therapy (Figure 11) and a decrease in tumour mass with more than 50% after the first course (Figure 12).
- The haemoglobin level influences the duration of remission (p= 0.02).
- The serum level of lactate dehydrogenase (LDH) was associated with a tendency for early relapse (p =0.007), Figure 13.

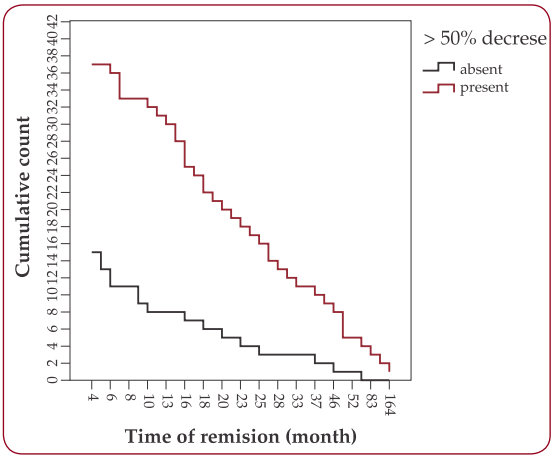

FIGURE 12. The correlation of remission duration with a decrease in tumour mass of > 50%

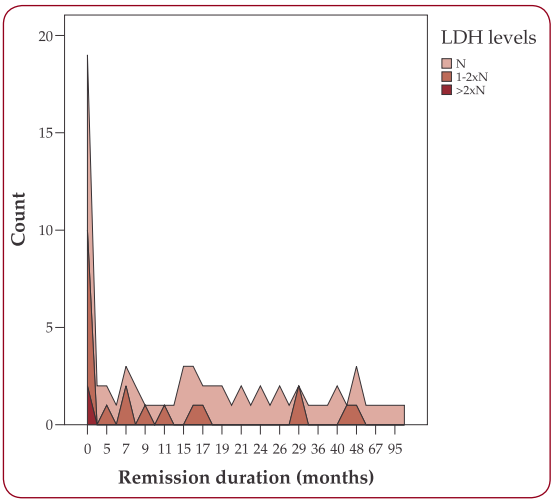

FIGURE 13. The correlation of remission duration with the LDH level

N - normal

- The FL histology type significantly correlated with the duration of disease remission. The 1st degree FL patients had a duration of remission and overall survival rate that was superior to 3rd degree FL.
- In contrast, the histology type of FL does not correlate with a tumour mass reduction after the first course, the type of response or the occurrence of a disease relapse.
- The highest average duration of remission, was found in patients with 2nd degree FL (17 patients) = 27.24 months + / - 25.133 months, followed by 1st degree FL (28 patients) = 22.46 months + / -34.539 months. The shortest period of remission duration was found in 3rd degree FL patients (26 patients) = 13.31 months + / - 26.328 months.
- The type of treatment significantly influences the duration of response from a statistical point of view (p=0.012). The average duration of response is 21.02 months + / -10.768 months in the case of patients with immunotherapy compared to18.92 months + / - 16.487 months in patients who did not receive immunotherapy.

Report Time of response (months)			
Treatment	Mean	Number	Std dev
Treatment R	21.02	26	16.48
Treatment nonR	18.92	45	10.6

- In the study group the average duration of response to induction treatment was 20.25 + / - 26.328 months, with the limits 4 months and 164 months.
- In case of a relapse, the duration of response to re-induction therapy is significantly lower, 9.92 months (compared to 20.25 months in case of induction), with a range of 0 - 5 months.
- The factor with the highest predictive value for the occurrence of relapse is the decrease of tumour mass by more than 50% after the first course ($p < 0.001$), (Figure 14).
- The occurrence of relapse is significantly correlated with the type of response to treatment ($p = 0.03$). In patients who achieved complete response, the relapse rate was lower (Figure 15).
- In patients with follicular lymphoma, who had a complete response, the share of those who did not have a relapse is higher than that of those who have relapsed.
- The proportion of patients who relapsed is higher in patients with a partial remission, than in those with a complete remission, and the relapse with transfor-

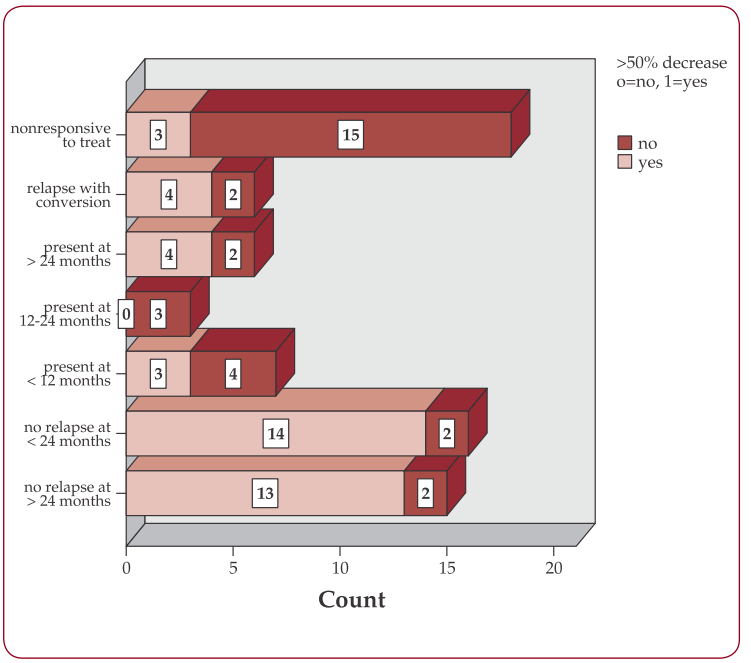

FIGURE 14. The correlation of FL relapses with a decrease in tumour mass

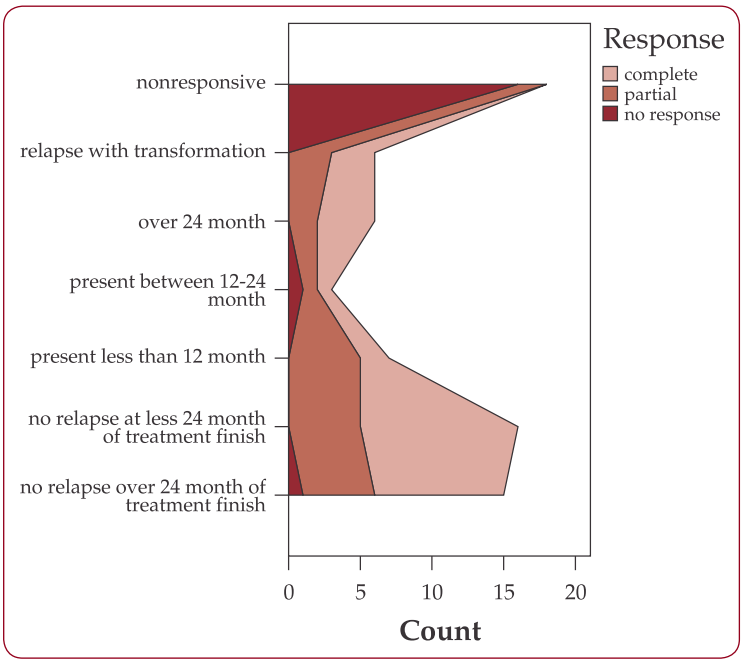

FIGURE 15. The correlation of occurrence of disease relapses with the response to treatment

mation into an aggressive lymphoma is present only in those showing a partial response and in none of the patients with a complete response.
- Thus, obtaining a CR and healing is associated with a lower risk of relapse, similar to M. Shipp's study.
- The remission duration influences the appearance of relapse (p=0.05). The longer the remission duration, the lower the chance of a relapse.
- The presence of thrombocytosis at diagnosis ($p < 0.008$). The onset of thrombocytosis correlates inversely to the duration of remission.
- Surprisingly, there is a weak correlation between LDH value and disease relapse (p= 0.053). ❑

CONCLUSIONS

- In patients with follicular lymphoma, the treatment with anti-CD20 monoclonal antibodies (Rituximab) has proven its superiority both in the induction therapy and maintenance of remission as in case of diseases relapses. We can say that the natural evolution of follicular lymphoma has been modified along with the introduction of immunotherapy.
- In the group of patients receiving immunotherapy combined with chemotherapy, 53.85% (14 cases) had a complete remission, as opposed to the group of patients without Rituximab, in which a CR was obtained in 35.55% (16 cases).
- The percentage of non-responders is significantly higher in patients who did not benefit from Rituximab: 31.11% compared to 15.38% in patients with Rituximab.
- The overall response rate was 84.61% (22 cases) in the Rituximab group, compared to 68.88% (31 cases) in patients without Rituximab.
- Prolonged remission (more than 24 months) was almost double (31%) in patients who received immuno-chemotherapy, compared to patients who received simple chemotherapy (16%).
- Early relapse of disease (less than 12 months) was present only in the group that did not receive immunotherapy (16% vs. 0%) and the percentage of non-responders was double (31%) in those who did not receive immunotherapy compared with those who received treatment with monoclonal antibodies (15%).
- The conventional treatments of indolent lymphomas do not extend the survival rate, nor do they cure. Until recently considered a utopia, the healing of lymphoma is the goal of the present therapy. Recent directions are centred on using several new drugs that are capable of overcoming the mechanisms that are resistant to recovery. The introduction of immunotherapy (Rituximab in 1997) and radio-immunotherapy (Tositumomab in 2002, Ibritumomab in 2003) seems to have changed the natural progression of follicular lymphoma.

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