World Journal of Hepatology

BRIEF ARTICLE

Hepatitis C virus-related B cell subtypes in non Hodgkin's lymphoma

Adriano M Pellicelli, Massimo Marignani, Valerio Zoli, Mario Romano, Aldo Morrone, Lorenzo Nosotti, Giuseppe Barbaro, Antonio Picardi, Umberto Vespasiani Gentilucci, Daniele Remotti, Cecilia D'Ambrosio, Caterina Furlan, Fabrizio Mecenate, Ettore Mazzoni, Ignazio Majolino, Roberto Villani, Arnaldo Andreoli, Giorgio Barbarini

Adriano M Pellicelli, Cecilia D'Ambrosio, Roberto Villani, Arnaldo Andreoli, Liver Unit, Azienda Ospedaliera San Camillo Forlanini, Circonvallazione Gianicolense 87, 00149 Rome, Italy
Massimo Marignani, Department of Digestive and Liver Disease, Azienda Ospedaliera Sant'Andrea, Via Grottarossa 1035/1039, 000189 Rome, Italy
Valerio Zoli, Ignazio Majolino, Hematology and Bone Marrow Transplantation, Azienda Ospedaliera San Camillo Forlanini, Circonvallazione Gianicolense 87, 00149 Rome, Italy
Mario Romano, Liver Unit, Ospedale Sandro Pertini, Via dei Monti Tiburtini 385, 00157 Rome, Italy
Aldo Morrone, Azienda Ospedaliera San Camillo Forlanini, Circonvallazione Gianicolense, 87, 00149 Rome, Italy
Lorenzo Nosotti, Medicine of Migration, National Institute for Migrant Health and Poverty, Via di S. Gallicano 25/a, 00153 Rome, Italy
Giuseppe Barbaro, Department of Medical Pathophysiology, University of Rome "La Sapienza", Viale del Policlinico 155, 00161 Rome, Italy
Antonio Picardi, Umberto Vespasiani Gentilucci, Liver Unit, Campus Biomedico University, Via Álvaro del Portillo 21, 00128 Rome, Italy
Daniele Remotti, Histopathology Unit, Azienda Ospedaliera San Camillo Forlanini, Circonvallazione Gianicolense 87, 00149 Rome, Italy
Caterina Furlan, Department of Infectious and Tropical Disease, University of Rome "La Sapienza", Viale del Policlinico 155, 00161 Rome, Italy
Fabrizio Mecenate, Liver Unit, Ospedale Villa Betania, Via Niccolò Piccolomini 27, 00165 Rome, Italy
Ettore Mazzoni, Liver Unit, Policlinico Casilino, Via Casilina 1049, 00169 Rome, Italy
Giorgio Barbarini, Infectious and Parasitic Diseases, Policlinico San Matteo P.zzale Golgi 2, 2710 Pavia, Italy

Abstract

AIM: To evaluate if indolent B cell-non Hodgkin's lymphoma (B-NHL) and diffuse large B-cell lymphoma (DLBCL) in hepatitis C virus (HCV) positive patients could have different biological and clinical characteristics requiring different management strategies.

METHODS: A group of 24 HCV related B-NHL patients (11 indolent, 13 DLBCL) in whom the biological and clinical characteristics were described and confronted. Patients with DLBCL were managed with the standard of care of treatment. Patients with indolent HCV-related B-NHL were managed with antiviral treatment pegylated interferon plus ribavirin and their course observed. The outcomes of the different approaches were compared.

RESULTS: Patients with DLBCL had a shorter duration of HCV infection and a higher prevalence of HCV genotype 1 compared to patients with indolent B-NHL in which HCV genotype 2 was the more frequent genotype. Five of the 9 patients with indolent HCV-related

B-NHL treated with only antiviral therapy, achieved a complete response of their onco-haematological disease (55%). Seven of the 13 DLBCL patients treated with immunochemotheraphy obtained a complete response (54%).

CONCLUSION: HCV genotypes and duration of HCV infection differed between B-NHL subtypes. Indolent lymphomas can be managed with antiviral treatment, while DLBCL is not affected by the HCV infection.

Key words: Hepatitis C virus infection; Diffuse large B cell lymphoma; Indolent lymphoma; Pegylated interferon; Lymphomagenesis

INTRODUCTION

The relationship between lymphoproliferative disorders and infectious agents has been studied for many decades. Epidemiological studies have linked hepatitis C virus (HCV) infection to B-cell non Hodgkin's Lymphomas (B-NHL)[1,2]. The majority of these studies were conducted in Italy, where the prevalence of HCV infection is particularly high[3-5]. However, studies conducted in countries with a lower prevalence of HCV infection also found a possible positive association between HCV and risk of developing B-NHL[6]. In a large pooled analysis of combined data from several countries, de Sanjose *et al*[7] demonstrated that presence of HCV infection was linked not only to marginal zone lymphoma, considered an indolent course B-NHL, but also to diffuse large B-cell lymphoma (DLBCL), a high-grade B-NHL. However, the strongest argument for a causative role of HCV infection in lymphoproliferative disease derives from interventional studies where antiviral regimen directed to HCV were successful in achieving the cure of HCV-related B-NHL[8-11].

In our study, the authors have analyzed and compared the biological and clinical features of HCV-related indolent B-NHL versus DLBCL. Furthermore, the authors have evaluated the outcomes of the different treatment approaches used in the management of these two types of B-NHL, and evaluated the influence of HCV infection on disease course.

MATERIALS AND METHODS

Patients

One hundred and twenty-five consecutive patients with B-NHL referred to our institution between January 2008 and January 2009 were included in the study and analyzed retrospectively. The diagnosis of lymphoma was established according to the 2008 World Health Organization (WHO) classification of tumours of haematopoietic and lymphoid tissues[12]. Total body computed tomography (CT) scan, bone marrow biopsy (BOM), lesion biopsies and physical examination were used to assess the stage and extranodal involvement according to Costwolds modification of the Ann Arbor classification[13]. Indolent B-NHL was defined by the 2008 WHO classification of Tumours of haematopoietic and lymphoid tissue[12] and included: marginal zone lymphoma (nodal, splenic and extranodal), lymphoplasmocytoid lymphoma and follicular lymphoma. Clinically, indolent course lymphoma was also defined as having a lesion doubling time greater than 1 year, no B symptoms and no masses > 10 cm (bulky disease). The study was approved by the local institutional review board.

Laboratory analysis

Enzyme-linked immunosorbent assays were used to determine anti-HCV antibodies in the 125 patients. All patients anti-HCV antibodies positive, underwent HCV-RNA testing [reverse transcription polymerase chain reaction (RT-PCR); detection limit <15 UI/mL)]. HCV-RNA determination was performed at disease diagnosis and subsequently at one, three, six and twelve months during antiviral treatment, and at six and twelve month after the its completion. HCV genotyping was performed in all viremic HCV patients (Immunogenetics Line Probe Assay, INNO-lipa HCV, Innogenetics, Ghent, Belgium). All HCV patients were tested for the presence of cryoglobulins. In all HCV-positive patients alanine aminotransferase (ALT) plasma levels were determined and expresses as UI/L. All HCV positive patients were tested for hepatitis B virus (HBV) antibodies and HBsAg (AxSYM, Abbott Laboratories, North Chicago, IL, United States), and for antibodies to human immunodeficiency virus (HIV) using an HIV-1 third-generation assay (AxSYM HIV 1/2, Abbott Laboratories). Presence of Bcl2 and Bcl6 were evaluated in biopsy samples or peripheral and medullary blood mononuclear cells using immunohistochemistry in all cases of HCV related B-NHL. The presence of HBV, HIV infection or concomitant neoplastic diseases excluded patients from the study. In HCV positive patients, demographic information such as periods of first and last exposure to injecting drug use or blood transfusion, tattoos and occupational exposure, needlestick injuries were recorded and considered as valid surrogate timepoints to define duration of HCV infection, expressed as mean ± SD.

Liver Biopsy

Fifteen of the 24 patients with HCV-related B-NHL gave informed consent to perform liver biopsy. Histologic

evaluation was carried out according to the Ishak score[14].

Antiviral treatment

Patients with indolent HCV-related B-NHL were offered treatment with antiviral therapy on the basis that previous interventional studies had demonstrated the efficacy of this approach in inducing a complete response of onco-hematological diseases (CR)[8-11]. In detail, pegylated interferon alpha 2a (180 μg) was administered subcutaneously once a week. It was combined with oral ribavirin: 800 mg/d when the patient weighed < 65 kg, 1000 mg/d when the weight was between 65 and 85 kg and 1200 mg/d when the patient was > 85 kg. Treatment was scheduled for 1 year if the patient had HCV genotype 1 or 4, and for 6 mo if the genotype was 2 or 3. Epoetin alfa was given at the dosage of 40 000 IU/week subcutaneously if haemoglobin levels (Hb) decreased by more than 2 g/100mL as compared to baseline in the first 4 wk of treatment, or when Hb was below 10 g/100 mL during treatment. Toxicity of antiviral treatment was evaluated according National Cancer Institute Common Terminology Criteria for Adverse Events; treatment dose was reduced in the case of grade 2 development, or withheld in the case of grade 3 toxicity (until toxicity had resolved to grade 2). Treatment was stopped in the case of grade 4 toxicity. Sustained virologic response (SVR) was defined as HCV-RNA negativity (< 12 IU/mL) 24 wk after stopping antiviral treatment. Patients were categorized as non-responders if HCV-RNA was positive after three months from the beginning of antiviral treatment. Relapser status was defined as the reappearance of HCV-RNA after antiviral treatment stoppage.

Haematologic Response

Antiviral treatment: In lymphoma patients managed with the antiviral treatment, haematological response was evaluated at the end of antiviral therapy, and every 3 mo thereafter.

Immunochemotherapy: Immunochemotherapy was reserved for patients with DLBCL, an aggressive form of lymphoma. The multi-drug regimen used in all patients was Cyclophoshamide, Doxorubicin, Vincristine and Prednisone associated with Rituximab (CHOP-R).

The haematological response was evaluated by means of physical examination, biochemical evaluation, CT scan and BOM when indicated according to standard response criteria[13]. CR was defined as no evidence of lymphoma. In patients with marginal splenic lymphoma, CR was defined as resolution of splenomegaly, absence of peripheral circulating villous lymphocytes, and normalization of platelet and white blood cells counts. When the BOM was initially positive, a negative BOM evaluation was an additional required criterion for confirming CR. Partial haematological response (PR) was defined as a ≥ 50% decrease in the size of all measurable lesions. The criteria for progressive haematological disease (PD) was a > 25% size increase in a previously documented lesion or the appearance of new lesions[15].

Statistical analysis

Continuous data were expressed as mean and standard deviation, and analysed using the *t*-test for independent samples with 95% confidence intervals. Categorical data were analysed using the χ^2 test, with Yates's correction and the Fisher's exact test. The significance level was set at a two-tailed *P* value < 0.05.

RESULTS

Of 125 consecutive patients presenting with B-NHL, 24 patients were HCV antibody positive, and all were viremic. The prevalence of HCV-related B-NHL in our population was 19.2%. Of these patients, 13 (54%) had DLBCL, while 11 (46%) had an indolent, HCV-related B-NHL (Table 1).

As compared to the indolent HCV-related B-NHL group, the DLBCL group had significantly more male patients, had a short duration of HCV infection, and a preponderance of patients with HCV genotype 1 infection. No differences in HCV-RNA titres, ALT levels, and histologic grading and staging between the two group of B-NHL were detected (Table 2). All DLBCL patients were treated with immunochemotherapy. Antiviral treatment was proposed to all 11 patients with indolent HCV related B-NHL. Nine out of 11 (81%) agreed to be treated with the antiviral treatment.

Antiviral treatment outcome

During antiviral treatment, 4 patients experienced grade 2 anaemia and were treated with epoetin alfa. One patient developed depression, and treatment with sertraline (50 mg/d) was started. However, all nine indolent, HCV-related B-NHL patients treated with antiviral treatment completed the scheduled course. All the patients had an end treatment response. Six months after antiviral treatment completion, 7 patients had a SVR, while 2 patients had a relapse of HCV infection (Table 3).

Haematologic outcome in patients treated with antiviral treatment

Complete response of onco-hematological diseases was obtained in 5 of the 7 patients with SVR (71%), while the remaining 2 patients with SVR had a PR. The two patients not responding to antiviral treatment developed PD. After a median of (14.2 ± 2) mo, the 5 patients originally obtaining SVR were still in CR (Table 3).

Haematological outcome in patients treated with immunochemotherapy

Thirteen patients with DLBCL were treated with immunochemotherapy as first line therapy. A CR was observed in 7 patients. In 2 of the 13 DLBCL patients treated with chemotherapy, there was an increase in ALT value (> 1.5 normal value) during treatment. None of the HCV-positive DLBCL patients had to stop treatment because of liver related events.

Table 1 Clinical and biological characteristics of 24 hepatitis C virus patients with low and high grade B cell-non Hodgkin's lymphoma

	Sex	Age (yr)	HCV RNA log₁₀ (UI/mL)	HCV genotype	Duration of HCV-infection (mo)	Ishak (grading)	Ishak (staging)	Type of lymphoma	Stage	Extranodal	Cryoglobulin	Bcl2/ Bcl6
1	F	70	5.90	2a/2c	21	4	0	Marginal extranodal (MALT)	IV	Orbit/BM	Positive	+/-
2	F	69	5.40	2a	19	12	6	Marginal extranodal (MALT)	IV	Parotid/BM	Positive	-/-
3	F	64	nd	2	25	nd	nd	Marginal extranodal (MALT)	IV	Orbit/BM	Positive	na
4	M	36	nd	2	7	6	1	Splenic marginal	IV	Spleen/BM	Negative	-/-
5	F	72	6.00	1b	22	8	2	Splenic marginal	III	Spleen	Negative	-/-
6	F	65	nd	2a/2c	nd	nd	nd	Splenic marginal	IV	Spleen/BM	Negative	-/-
7	F	70	5.90	2	nd	8	1	Marginal nodal	IV	BM	Positive	-/-
8	F	55	5.07	2a/2c	31	17	6	Follicular lymphoma	III	None	na	+/+
9	M	67	6.80	2a	30	6	0	Follicular lymphoma	IV	Liver/BM	Negative	+/-
10	F	59	nd	2	20	16	6	Follicular lymphoma	III	None	Negative	+/+
11	F	78	nd	2a	30	nd	nd	Lymphoplasmocytoid lymphoma	IV	BM	Positive	na
12	M	61	nd	1b	18	5	2	DLBCL	IV	BM	na	na
13	M	29	7.80	1a	4	6	0	DLBCL	II	None	Negative	+/+
14	M	66	6.70	1b	nd	12	6	DLBCL		Liver, Lung	na	na
15	M	62	6.90	1a	10	nd	nd	DLBCL	IV	BM	na	na
16	M	56	5.1	1b	nd	nd	nd	DLBCL	IV	BM	na	na
17	F	55	5.4	1b	nd	nd	nd	DLBCL	IV	Liver	na	na
18	F	59	6.4	1a/1b	nd	nd	nd	DLBCL	IV	Lung/BM	na	+/+
19	M	65	5.8	1b	11	10	2	DLBCL	IV	Lung	Negative	+/+
20	M	46	nd	1b	6	8	1	DLBCL	IV	Liver	Negative	+/+
21	M	51	6.3	1b	15	10	2	DLBCL	IV	Lung/BM	Negative	-/-
22	M	49	5.9	3	15	nd	nd	DLBCL	IV	Gastric	Negative	-/-
23	M	78	6.2	2a/2c	19	10	1	DLBCL	III	None	Negative	+/+
24	F	74	5.0	1	16	10	4	DLBCL	IV	Palatine Tonsil/BM	Negative	-/-

DLBCL: Diffuse large B cells lymphoma; nd: Not determined; Extranodal: Extranodal involvement; BM: Bone marrow; na: Not available; MALT: Mucosal associated lymphoid tissue; HCV: Hepatitis C virus.

Table 2 Comparison of biological, virological and clinical characteristics of hepatitis C virus-related low *vs* high grade B cell-non Hodgkin's lymphoma

	DLBCL (*n* = 13)	Indolent B-NHL (*n* = 11)	95% CI	*P* value
Age (yr)[1]	57 ± 12	64 ± 11	-16.8 to 2.8	0.153
Sex (M/F)	10/3	2/9	-	0.014
Duration of HCV infection (yr)[1]	12 ± 5	22 ± 7	-15.1 to -4.9	< 0.001
ALT value (UI/L)[1]	58 ± 40	40 ± 27	-11.5 to 47.5	0.219
Genotype 1/not 1	10/3	2/9	-	0.014
HCVRNA log₁₀ (UI/mL)[1]	6.1 ± 0.6	5.8 ± 0.3	-0.11 to 0.71	0.147
Ishak (grading)[1]	8.4 ± 2.1	9.3 ± 5.0	-4.05 to 2.25	0.560
Ishak (staging)[1]	1.5 ± 1.2	2.75 ± 2.7	-2.97 to 0.47	0.146

[1]Means ± SD; HCV: Hepatitis C virus; B-NHL: B cell-Non Hodgkin's lymphoma; DLBCL: Diffuse large B cells lymphoma; CI: Confidence interval.

DISCUSSION

In accordance with de Sanjose *et al*[7], we found a high prevalence of HCV-positives (19%) among DBLC and indolent lymphoma patients. We observed that DLBCL patients had a history suggestive of a short duration of HCV infection, and a higher prevalence of genotype 1, as compared to patients with indolent, low-grade B-NHL, who had a higher prevalence of genotype 2. Various clinical studies failed to demonstrate an association between B-NLH and specific HCV genotypes, although the possible association between specific HCV genotypes and particular subtypes of B-NHL was not considered in these studies[3,9,16].

Several studies have demonstrated differences between infection due to HCV genotypes 1 and 2[17,18]. In HCV genotype 2 infection, HCV-RNA titre was lower, there were more patients with normal ALT values, and the patients had a longer duration of HCV infection as compared to genotype 1 patients. Conversely, chronic HCV hepatitis progression appears to have a more rapid and severe course in genotype 1 as compared to genotype 2[17]. Because HCV genotype 2 is associated with a longer duration of viral infection, it can be speculated that over time it might induce a persistent immunostimulation of B cells. Zignego *et al*[19,20] have found that type II mixed crioglobulinemia and *bcl-2* expression in HCV genotype 2 patients was the more frequent cause of a prolonged immunostimulation of the B cell exerted by HCV over time. In our study HCV genotype 2 was detected in 5 out of 8 patients who had *bcl-2* positivity and in all the patients where a long duration of HCV infection was found. We believe that *bcl-2* expression in indolent lymphoma, such as, follicular lymphoma and marginal zone lymphoma, is the expression of a chronic B cell proliferation in response to antigenic stimulation or polyclonal activation in genotype 2 patients with and without cryoglobulins.

It has been demonstrated that the HCV envelope

Table 3 Results of antiviral treatment in patients with low grade B cell-non Hodgkin's lymphoma

	Sex	Age (yr)	Histology	Stage	HCVRNA log₁₀ (UI/mL)	Genotype	Staging (Ishak)	Treatment	Treatment duration (mo)	Response to treatment	Remission lymphoma
1	F	70	Marginal extranodal	IV	5.9	2a/2c	0	Peg 180 µg/wk + RBV 800 mg/d	6	SVR	Complete hematological response
2	F	69	Marginal extranodal	IV	5.4	2a	6	Peg 180 µg/wk + RBV 800 mg/d	6	SVR	Complete hematological response
3	F	64	Marginal extranodal	IV	5.6	1b	nd	Peg 180 µg/wk + RBV 1000 mg/d	12	Relapser	Progressive disease
4	M	36	Marginal splenic	IV	5.5	2	1	Peg 180 µg/wk + RBV 800 mg/d	6	SVR	Partial response
5	F	72	Marginal splenic	III	6.0	1b	2	Peg 180 µg/wk + RBV 1000 mg/d	12	SVR	Partial response
6	F	65	Marginal splenic	IV	5.6	2a/2c	nd	Peg 180 µg/wk + RBV 1200 mg/d	6	SVR	Complete hematological response
7	F	70	Marginal nodal	IV	5.9	2	2	Peg 180 µg/wk + RBV 800 mg/d	6	SVR	Complete hematological response
8	F	55	Follicular lymphoma	III	5.0	2a/2c	6	Peg 180 µg/wk + RBV 800 mg/d	6	Relapser	Progressive disease
9	M	67	Follicular lymphoma	IV	6.8	2a	0	Peg 180 µg/wk + RBV 800 mg/d	6	SVR	Complete hematological response

MU: Million of units; RBV: Ribavirin; SVR: Sustained virological response.

glycoprotein E2 interacts with a specific B cell receptor associated with the CD19/CD21/CD81 complex. This interaction lowers the threshold for B cell activation and induces the proliferation of benign B cells. However, this prolonged exposure to stimuli may render B cells at risk for additional events leading to malignant transformation[21]. This mechanism may offer a clue to interpreting lymphomagenesis in genotype 2 patients affected by indolent HCV-related B-NHL in whom a prolonged exposure to immunostimulation of the B-cell compartment seems to be a characteristic.

On the contrary, in our study direct lymphocyte transformation could be hypothesized in HCV genotype 2 patients on the basis of the shorter duration of HCV infection. Furthermore we found positivity of *bcl-6* in five DLBCL patients, while *bcl-6* negativity was found in only three patients. Positivity of *bcl-6* in DLBCL is found in typical DLBCL and not in DLBCL transformed from indolent lymphoma. We believe that this data reinforces the hypothesis of a direct lymphomagenesis of HCV in DLBCL[22]. B cell receptors can bind HCV and efficiently internalize the virus, possibly causing genomic instability[21,23-25]. This mechanism might be involved in a possible scenario of direct lymphomagenesis. Direct lymphocyte transformation has been demonstrated for Epstein-Barr virus, human herpes virus 8, and human T lymphotropic virus 1. However this mechanism, even if intriguing, has not been conclusively demonstrated for HCV[26].

Because the etiopathogenetic mechanisms underlying HCV-related low and high-grade B-NHL may differ, the optimal management approach also differs. Indolent, HCV-related B-NHL is a subset of neoplasms characterized by an unrelenting course that requires chemotherapy only if an aggressive behaviour develops. In this subset of lymphomas, antiviral treatment alone may eradicate the HCV infection and stop the chronic antigen-driven lymphoproliferation. Clinical studies have demonstrated the efficacy of antiviral treatment in these patients[9-11]. A systematic review of Gisbert *et al*[8] has shown that in 65 HCV infected patients with lymphoproliferative disorders treated with antiviral regimen, CR was achieved in 75% of cases. In contrast, HCV negative patients did not respond to interferon, indicating that CR in HCV positive patients was not merely due to the antiproliferative action of interferon.

Our study confirms that combined antiviral therapy is effective in inducing CR in indolent, HCV-related B-NHL, and that reaching SVR seems to be crucial in maintaining it. In our studies most patients reaching SVR achieved a CR, while 2 patients relapsing after treatment had a progression of haematologic disease. However, two of the HCV positive, indolent B-NHL reaching SVR had a PR, suggesting that other mechanisms may intervene in determining CR. Obviously, the high percentage of SVR obtained in this group is also associated to the elevated prevalence of genotype 2, a well known easier-to-treat genotype. However, antiviral therapy of HCV-positive indolent B-NHL is an attractive therapeutic option, even if SVR is the objective.

For HCV-positive DLBCL, immunochemotherapy is necessary. A recently published paper observed that HCV-positive status is a risk factor for the development of hepatitis flare in patients treated with rituximab-containing regimens. In this retrospective study none of the HCV-negative lymphoma patients receiving rituximab–containing therapy developed hepatitis flares as compared with a significant percentage in the HCV positive group. However, even if no liver-related deaths deriving from the hepatitis flare were observed in this paper, it was impossible to define the exact mechanism of toxicity[27]. In our

study we treated all HCV-positive DLBCL patients with CHOP-R regimen, and CR was obtained and maintained over the period in 7 of 13 patients despite the persistence of HCV infection. Hepatotoxicity, expressed by an increase in transaminases and bilirubin, was noted in 2 patients, with liver biopsy showing cirrhosis in both (patients 14 and 24). However, in our study none of the patients had to stop or modify treatment because of liver-related complications.

In conclusion, indolent HCV-related B-NHL and DLBCL have different biological and clinical features: the former is associated with a higher prevalence of HCV genotype 2 and a longer exposure to HCV infection, whereas the latter more frequently shows infection with genotype 1 and a shorter duration of HCV exposure. These characteristics and the differential response of indolent HCV-related lymphomas to antiviral treatment suggest that these two groups might follow different pathways of lymphomagenesis. Thus, we strongly believe that antiviral treatment should be the first line of treatment to be offered in these patients. As far as the treatment of DLBCL patients is concerned, we observed that liver-related complications can develop in HCV-positive cases, but these are marginal and do not require modifications to the onco-hematological treatment schedule, thus not affecting the opportunity of obtaining CR. Further studies are needed to determine the utility of antiviral treatment as consolidation therapy after cytostatic treatment for high-grade B-NHL.

COMMENTS

Background
Non Hodgkin's Lymphoma (NHL) is the hematologic malignancy with the highest prevalence worldwide. Among the risk factor for NHL are primary and acquired immune deficiency as well as several infectious agents such as hepatitis C virus (HCV). A positive association between HCV and NHL has been confirmed in a large number of studies. It has been reported that clearance of HCV infection by antiviral treatment led to regression of the tumor burden in indolent HCV-related NHL, while the therapeutic approach for HCV-related high grade NHL could be different. In this study the authors have analyzed and compared the biological and clinical features of HCV-related indolent B cell-non Hodgkin's lymphoma (B-NHL) *vs* HCV-related high grade lymphomas, such as diffuse large B cell lymphoma. Furthermore, the authors have evaluated the outcomes of the different treatment approaches used in the management of these two types of B-NHL, and evaluated the influence of HCV infection on disease course.

Research frontiers
HCV-related lymphomagenesis could be due to different immunopathologic mechanisms. While in indolent HCV-related lymphoma an indirect role of HCV in lymphomagenesis is hypothesized, in HCV-related high grade lymphoma a direct mechanism could be possible. A deep understanding of the mechanism of HCV lymphomagenesis is essential for the development of further therapeutic approaches.

Innovations and breakthroughs
This is the first study that has analyzed and compared the biological and clinical features of two subtypes of HCV-related B cell NHL. Characteristics of HCV infection such as viral genotype, duration of HCV infection, and histopathology could be of fundamental importance to understand the different treatment approaches used in the management of these two subtypes of HCV-related B cell NHL.

Applications
The observations could have clinical importance for future therapeutic approaches in HCV-related B cell NHL.

Terminology
HCV-related B cell NHL is a lymphoma that is linked to HCV infection
Peer review
This is a study that analyzes the mechanism of HCV-related lymphomagenesis and then the rationale of therapeutic approaches.

REFERENCES

1   **Gisbert JP**, García-Buey L, Pajares JM, Moreno-Otero R. Prevalence of hepatitis C virus infection in B-cell non-Hodgkin's lymphoma: systematic review and meta-analysis. *Gastroenterology* 2003; **125**: 1723-1732
2   **Giordano TP**, Henderson L, Landgren O, Chiao EY, Kramer JR, El-Serag H, Engels EA. Risk of non-Hodgkin lymphoma and lymphoproliferative precursor diseases in US veterans with hepatitis C virus. *JAMA* 2007; **297**: 2010-2017
3   **Mele A**, Pulsoni A, Bianco E, Musto P, Szklo A, Sanpaolo MG, Iannitto E, De Renzo A, Martino B, Liso V, Andrizzi C, Pusterla S, Dore F, Maresca M, Rapicetta M, Marcucci F, Mandelli F, Franceschi S. Hepatitis C virus and B-cell non-Hodgkin lymphomas: an Italian multicenter case-control study. *Blood* 2003; **102**: 996-999
4   **Vallisa D**, Bertè R, Rocca A, Civardi G, Giangregorio F, Ferrari B, Sbolli G, Cavanna L. Association between hepatitis C virus and non-Hodgkin's lymphoma, and effects of viral infection on histologic subtype and clinical course. *Am J Med* 1999; **106**: 556-560
5   **Cocco P**, Piras G, Monne M, Uras A, Gabbas A, Ennas MG, Palmas A, Murineddu M, Collu S, Melis M, Rais M, Todde P, Cabras MG, Angelucci E, Massarelli G, Nieters A. Risk of malignant lymphoma following viral hepatitis infection. *Int J Hematol* 2008; **87**: 474-483
6   **Schöllkopf C**, Smedby KE, Hjalgrim H, Rostgaard K, Panum I, Vinner L, Chang ET, Glimelius B, Porwit A, Sundström C, Hansen M, Adami HO, Melbye M. Hepatitis C infection and risk of malignant lymphoma. *Int J Cancer* 2008; **122**: 1885-1890
7   **de Sanjose S**, Benavente Y, Vajdic CM, Engels EA, Morton LM, Bracci PM, Spinelli JJ, Zheng T, Zhang Y, Franceschi S, Talamini R, Holly EA, Grulich AE, Cerhan JR, Hartge P, Cozen W, Boffetta P, Brennan P, Maynadié M, Cocco P, Bosch R, Foretova L, Staines A, Becker N, Nieters A. Hepatitis C and non-Hodgkin lymphoma among 4784 cases and 6269 controls from the International Lymphoma Epidemiology Consortium. *Clin Gastroenterol Hepatol* 2008; **6**: 451-458
8   **Gisbert JP**, García-Buey L, Pajares JM, Moreno-Otero R. Systematic review: regression of lymphoproliferative disorders after treatment for hepatitis C infection. *Aliment Pharmacol Ther* 2005; **21**: 653-662
9   **Vallisa D**, Bernuzzi P, Arcaini L, Sacchi S, Callea V, Marasca R, Lazzaro A, Trabacchi E, Anselmi E, Arcari AL, Moroni C, Bertè R, Lazzarino M, Cavanna L. Role of anti-hepatitis C virus (HCV) treatment in HCV-related, low-grade, B-cell, non-Hodgkin's lymphoma: a multicenter Italian experience. *J Clin Oncol* 2005; **23**: 468-473
10 **Hermine O**, Lefrère F, Bronowicki JP, Mariette X, Jondeau K, Eclache-Saudreau V, Delmas B, Valensi F, Cacoub P, Brechot C, Varet B, Troussard X. Regression of splenic lymphoma with villous lymphocytes after treatment of hepatitis C virus infection. *N Engl J Med* 2002; **347**: 89-94
11 **Mazzaro C**, De Re V, Spina M, Dal Maso L, Festini G, Comar C, Tirelli U, Pozzato G. Pegylated-interferon plus ribavirin for HCV-positive indolent non-Hodgkin lymphomas. *Br J Haematol* 2009; **145**: 255-257
12 **Swerdlow SH**, Campo E, Harris NL, Jaffe ES, Pileri SA, Stein H, Thiele J, Vardiman JW eds. 2008 World Health Organization (WHO): classification of tumours. Tumours of haematopoietic and lymphoid tissue. Lyon: IARC Press, 2008: 267-268
13 **Lister TA**, Crowther D, Sutcliffe SB, Glatstein E, Canellos GP,

Young RC, Rosenberg SA, Coltman CA, Tubiana M. Report of a committee convened to discuss the evaluation and staging of patients with Hodgkin's disease: Cotswolds meeting. *J Clin Oncol* 1989; **7**: 1630-1636

14   **Ishak K,** Baptista A, Bianchi L, Callea F, De Groote J, Gudat F, Denk H, Desmet V, Korb G, MacSween RN, Histological grading and staging of chronic hepatitis. *J Hepatol* 1995; **22:** 696-699

15   **Cheson BD**, Horning SJ, Coiffier B, Shipp MA, Fisher RI, Connors JM, Lister TA, Vose J, Grillo-López A, Hagenbeek A, Cabanillas F, Klippensten D, Hiddemann W, Castellino R, Harris NL, Armitage JO, Carter W, Hoppe R, Canellos GP. Report of an international workshop to standardize response criteria for non-Hodgkin's lymphomas. NCI Sponsored International Working Group. *J Clin Oncol* 1999; **17**: 1244

16   **Silvestri F**, Pipan C, Barillari G, Zaja F, Fanin R, Infanti L, Russo D, Falasca E, Botta GA, Baccarani M. Prevalence of hepatitis C virus infection in patients with lymphoproliferative disorders. *Blood* 1996; **87**: 4296-4301

17   **Kobayashi M**, Tanaka E, Sodeyama T, Urushihara A, Matsumoto A, Kiyosawa K. The natural course of chronic hepatitis C: a comparison between patients with genotypes 1 and 2 hepatitis C viruses. *Hepatology* 1996; **23**: 695-699

18   **Prati D**, Capelli C, Zanella A, Mozzi F, Bosoni P, Pappalettera M, Zanuso F, Vianello L, Locatelli E, de Fazio C, Ronchi G, del Ninno E, Colombo M, Sirchia G. Influence of different hepatitis C virus genotypes on the course of asymptomatic hepatitis C virus infection. *Gastroenterology* 1996; **110**: 178-183

19   **Zignego AL**, Ferri C, Giannini C, Monti M, La Civita L, Careccia G, Longombardo G, Lombardini F, Bombardieri S, Gentilini P. Hepatitis C virus genotype analysis in patients with type II mixed cryoglobulinemia. *Ann Intern Med* 1996; **124**: 31-34

20   **Zignego AL**, Ferri C, Giannelli F, Giannini C, Caini P, Monti M, Marrocchi ME, Di Pietro E, La Villa G, Laffi G, Gentilini P. Prevalence of bcl-2 rearrangement in patients with hepatitis C virus-related mixed cryoglobulinemia with or without B-cell lymphomas. *Ann Intern Med* 2002; **137**: 571-580

21   **Suarez F**, Lefrere F, Besson C, Hermine O. Splenic lymphoma with villous lymphocytes, mixed cryoglobulinemia and HCV infection: deciphering the role of HCV in B-cell lymphomagenesis. *Dig Liver Dis* 2007; **39** Suppl 1**:** S32-S37

22   **Besson C**, Canioni D, Lepage E, Pol S, Morel P, Lederlin P, Van Hoof A, Tilly H, Gaulard P, Coiffier B, Gisselbrecht C, Brousse N, Reyes F, Hermine O. Characteristics and outcome of diffuse large B-cell lymphoma in hepatitis C virus-positive patients in LNH 93 and LNH 98 Groupe d'Etude des Lymphomes de l'Adulte programs. *J Clin Oncol* 2006; **24**: 953-960

23   **Marcucci F**, Mele A. Hepatitis viruses and non-Hodgkin lymphoma: epidemiology, mechanisms of tumorigenesis, and therapeutic opportunities. *Blood* 2011; **117**: 1792-1798

24   **Levy S.** Alteration of B cell function by hepatitis C virus. *Haematol Rep* 2005; **1:** 55-58

25   **Quinn ER**, Chan CH, Hadlock KG, Foung SK, Flint M, Levy S. The B-cell receptor of a hepatitis C virus (HCV)-associated non-Hodgkin lymphoma binds the viral E2 envelope protein, implicating HCV in lymphomagenesis. *Blood* 2001; **98**: 3745-3749

26   **Suarez F**, Lortholary O, Hermine O, Lecuit M. Infection-associated lymphomas derived from marginal zone B cells: a model of antigen-driven lymphoproliferation. *Blood* 2006; **107**: 3034-3044

27   **Marignani M,** Mangone M, Cox MC, Angeletti S, Veggia B, Ferrari A, di Fonzo M, Begini P, Gigante E, Laverde G, Aloe-Spiriti A, Monarca B, Delle Fave G. HCV-positive status and hepatitis flares in patients with B-cell non-Hodgkin's lymphoma treated with rituximab-containing regimens. *Dig Liver Dis* 2011; **43**: 139-142