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Efficacy and Tolerability of Rituximab With or Without PEGylated Interferon Alfa-2b Plus Ribavirin in Severe Hepatitis C Virus–Related Vasculitis

A Long-Term Followup Study of Thirty-Two Patients

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Objective. **To report on the long-term followup of a cohort of patients with hepatitis C virus (HCV)–related vasculitis treated with rituximab with or without PEGylated interferon alfa-2b (PEG–IFN alfa-2b) plus ribavirin.**

Methods. **The study group comprised 32 HCV RNA–positive patients with HCV-related vasculitis: 20 patients were treated with rituximab and PEG–IFN alfa-2b (9 of whom had not previously received antiviral treatment and 11 of whom had experienced disease resistance to or relapse with antiviral treatment), and 12 antiviral-intolerant patients were treated with rituximab alone.**

Results. **Treatment with rituximab and PEG–IFN alfa-2b plus ribavirin induced a complete clinical response and a partial clinical response in 80% and 15% of patients, respectively, a complete immunologic response and a partial immunologic response in 67% and 33% of patients, respectively, and a sustained virologic response in 55% of patients. Treatment with rituximab alone induced a complete clinical response and a partial clinical response in 58% and 9% of patients, respectively, and a complete immunologic response and a partial immunologic response in 46% and 36% of patients, respectively. B cell depletion was achieved in 96% of patients, and B cell recovery began after a median delay of 12 months. After a mean ± SD followup period of 23 ± 12 months, 22% of patients experienced a clinical relapse, and 34% of patients experienced an immunologic relapse. All relapses were associated with the absence of virologic control, and 78% of relapses were associated with B cell recovery. Six patients were re-treated with rituximab. All 6 of these patients had a complete clinical response, 50% had a complete immunologic response, and 50% had a partial immunologic response. Rituximab was well tolerated overall.**

Conclusion. **Rituximab is an effective treatment of severe and/or refractory HCV-related vasculitis. Relapses were consistently associated with the absence of virologic control. The clinical and immunologic efficacy of rituximab after repeated infusion appeared to be the same as that observed after induction therapy.**

Mixed cryoglobulinemia (MC) vasculitis is a systemic vasculitis that affects mainly the small and, less

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frequently, medium-sized vessels (1) and is attributable to the expansion of B cells producing pathogenic IgM with rheumatoid factor (RF) activity (2,3). MC leads to clinical manifestations ranging from the so-called MC syndrome (purpura, arthralgia, and asthenia) to more serious lesions with neurologic and renal involvement (4). More than 80% of MC vasculitis cases are associated with hepatitis C virus (HCV) infection (5–7).

Treatment of HCV-related MC with severe organ involvement may target either the viral trigger (HCV) or the downstream B cell arm of autoimmunity (8,9). The main goals of treatment are to induce a sustained virologic and clinical response and to minimize the use of immunosuppressive drugs. Treatment with PEGylated interferon alfa-2b (PEG–IFN alfa-2b) plus ribavirin has resulted in sustained clinical and virologic responses in up to 60% of cases (10). Cryoglobulin clearance was also observed in half of patients, and antiviral therapy has been shown to reverse bone marrow monoclonal B cell expansion in patients with HCV-related MC (11). Although this approach provides a satisfactory response rate, additional therapy may be needed in MC patients with severe organ involvement or in those who do not experience an early virologic response (10,12). Corticosteroids in combination with cytotoxic agents may be useful for controlling life-threatening organ involvement while awaiting the generally slow response to antiviral treatment.

More recently, preliminary data on the efficacy of rituximab, an anti-CD20 monoclonal antibody, in a small cohort of patients with MC-related vasculitis have been reported (13–19). A complete clinical response was observed in 60–70% of patients, with cryoglobulin clearance in one-third of patients. However, the absence of efficacy for HCV clearance supports the need for combined antiviral therapy to block the HCV infection trigger and obviate long-term liver complications of such a chronic infection. We previously reported the results of a pilot study using rituximab combined with PEG–IFN alfa-2b plus ribavirin in severe refractory HCV-associated MC vasculitis. More than 90% of the treated patients showed clinical improvement, and two-thirds of these patients experienced a complete clinical response. HCV RNA and serum cryoglobulin became undetectable in all of the patients who had a complete clinical response. Treatment was well tolerated, with no infectious complications (20).

The aim of the current study was to describe the efficacy and tolerance of rituximab, with and without combined antiviral therapy, in a large cohort of patients with HCV-related vasculitis and long-term followup.

PATIENTS AND METHODS

Patients. The study group comprised 32 consecutive patients treated with rituximab for HCV-related vasculitis, with (n = 20) or without (n = 12) combined antiviral therapy with PEG–IFN alfa-2b plus ribavirin. All of the patients were seen between 2004 and 2008. Patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin included 9 who were naive in the present study included 9 who had not previously received antiviral therapy and 11 of 16 patients included in a previous study (20) whose disease was resistant to or had relapsed after combination antiviral therapy, for whom 14 months of additional followup is reported (the 5 remaining patients in that study died [n = 1] or were lost to followup [n = 4]). Twelve patients were treated with rituximab alone because of inefficacy of ≥2 previous and well-conducted antiviral therapies (including PEG–IFN alfa-2b plus ribavirin for at least 12 months) (n = 6), poor tolerance (i.e., fever, myalgia, cytopenia) to previous antiviral therapy received for 5, 6, and 9 months, respectively (n = 3), or contraindications to IFN because of a psychiatric disorder (n = 3). The study conformed to the ethics guidelines of the Declaration of Helsinki. Patients had a serum cryoglobulin level of >0.05 gm/liter on at least 2 occasions that was associated with purpura, arthralgia, and renal or neurologic involvement (except 3 patients without detectable cryoglobulin in the serum). All patients were positive for serum HCV RNA.

Inclusion criteria for the study were as follows: 1) chronic active HCV infection, 2) severe vasculitis-related organ involvement (i.e., renal, digestive, or neurologic), and 3) a minimum of 6 months of followup after initiation of rituximab treatment. Exclusion criteria were the presence of hepatitis B surface antigen or anti–human immunodeficiency virus antibodies. Twenty-five patients had histologically confirmed systemic vasculitis (nerve [n = 13], kidney [n = 9], and skin [n = 3]). Among those without histologically proven vasculitis (n = 7), patients with purpura were considered to have small-vessel vasculitis, and those with HCV infection and MC were classified as having MC-related vasculitis.

None of the patients had received previous treatment with rituximab. The therapy schedule was as follows: 1) weekly administration of 4 intravenous infusions of rituximab at a dose of 375 mg/m^2 (on days 1, 8, 15, and 22) over a 1-month period (n = 27) and 2) administration of 2 intravenous infusions of rituximab at a dose of 1,000 mg (on days 1 and 15) (n = 5). The patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin (n = 20) received a combination of PEG–IFN alfa-2b (1.5 μg/kg/week subcutaneously) plus ribavirin (600–1,200 mg/day orally) 1 month after the last rituximab infusion, for a median duration of 12 months (range 3–20 months). To avoid cumulative side effects, rituximab and the antiviral therapy were not started at the same time.

Eleven patients received corticosteroids in association with rituximab because of life-threatening vasculitis (n = 7), acute renal failure related to membranoproliferative glomerulonephritis (n = 2), immune thrombocytopenia (n = 1), or rituximab-related serum sickness (n = 1). Three patients underwent plasmapheresis because of acute renal failure related to membranoproliferative glomerulonephritis (n = 2) or life-threatening vasculitis (n = 1). No patients received cyclophosphamide in association with rituximab as first-line treat-

Table 1. Baseline characteristics of the patients with chronic active HCV-related vasculitis, according to treatment*

Characteristic	All patients (n = 32)	Rituximab with PEG–IFN alfa-2b plus ribavirin (n = 20)	Rituximab only (n = 12)
No. of men/no. of women	14/18	6/14	8/4
Age, mean ± SD years	59 ± 12	61 ± 11	57 ± 13
HCV related			
Genotype 1	16 (52)	12 (63)	4 (34)
Genotype 2	7 (23)	4 (21)	3 (25)
Genotype 3	5 (16)	2 (11)	3 (25)
Genotype 4	1 (3)	0 (0)	1 (8)
Genotype 5	2 (6)	1 (5)	1 (8)
Genotype not available	1	1	0
HCV RNA, mean ± SD log copies/IU	5.9 ± 0.6	5.8 ± 0.5	6.1 ± 0.8
Vasculitis duration, mean ± SD months	31 ± 42	28 ± 46	36 ± 35
METAVIR score, mean ± SD			
Activity score	1.4 ± 0.9	1.4 ± 1.0	1.4 ± 0.5
Fibrosis score	2.2 ± 1.4	2.3 ± 1.4	2.0 ± 1.4
MC related			
Cryoglobulin positivity	29 (91)	18 (90)	11 (92)
Cryoglobulin level, mean ± SD gm/liter	1.03 ± 0.78	1.25 ± 0.78†	0.66 ± 0.63
Type II cryoglobulins	28 (97)	18 (100)	10 (91)
Monoclonal kappa	27 (96)	18 (100)	9 (90)
Monoclonal lambda	1 (4)	0 (0)	1 (10)
Type III cryoglobulins	1 (3)	0 (0)	1 (9)
C4, mean ± SD gm/liter	0.08 ± 0.09	0.08 ± 0.10	0.09 ± 0.09
RF positivity	28/31 (90)	17/19 (89)	11/12 (92)
RF, mean ± SD IU/liter	236 ± 387	182 ± 224	329 ± 573
IgM, mean ± SD gm/liter	2.5 ± 3.2	2.0 ± 1.1	3.1 ± 4.4
Vasculitis-related organ involvement			
Purpura	22 (69)	14 (70)	8 (67)
Peripheral nervous system	22 (69)	16 (80)	6 (50)
Arthralgia	17 (53)	11 (55)	6 (50)
Kidney	14 (44)	10 (50)	4 (33)
Myalgia	6 (19)	3 (15)	3 (25)
Gastrointestinal tract	3 (9)	3 (15)	0 (0)
Heart	3 (9)	2 (10)	1 (8)
Central nervous system	2 (6)	2 (10)	0 (0)
B cell non-Hodgkin's lymphoma	8 (25)	4 (20)	4 (33)
Treatment			
Rituximab with PEG-IFN alfa-2b plus ribavirin	20 (63)	20	0
Antiviral-naive patients	9	9	–
Antiviral-resistant or antiviral-relapser patients	11	11	–
Rituximab only	12 (37)	–	12

* Except where indicated otherwise, values are the number (%) of patients. HCV = hepatitis C virus; PEG–IFN alfa-2b = PEGylated interferon alfa-2b; MC = mixed cryoglobulinemia; RF = rheumatoid factor.
† $P = 0.04$ versus rituximab only.

ment. Comorbidities included arterial hypertension (n = 5), diabetes mellitus (n = 5), coronary artery disease (n = 3), cutaneous lupus (n = 1), antiphospholipid syndrome (n = 1), and polycystic kidney disease (n = 1).

For each patient, clinical and laboratory data were recorded at the time of the initial evaluation, at 3 months, 6 months, 9 months, and 12 months, and then every 6 months and at the end of followup. The diagnosis of non-Hodgkin's lymphoma was based on the World Health Organization criteria (21).

Immunologic and virologic markers. Cryoglobulins were measured and classified as previously described (22,23). The glomerular filtration rate was determined as previously described by Cockcroft and Gault (24). A 24-hour urine collection was also performed in order to quantify daily protein excretion. Flow cytometric analysis of the CD19+ B cell marker was evaluated in peripheral blood mononuclear cells at baseline, every 3 months for 1 year, and then every 6 months. Serum levels of HCV RNA were measured by reverse transcription–polymerase chain reaction, with a detection threshold of 2.7 log copies/ml (15 IU/ml). Liver biopsy specimens or noninvasively obtained biologic markers of liver involvement were evaluated according to the previously validated METAVIR scoring system (25).

Assessment of treatment response. The response to treatment was defined as previously described (10), by com-

paring clinical, immunologic, and virologic parameters at the initial evaluation, at 3 months, 6 months, 9 months, and 12 months, and then every 6 months and at the end of followup. Clinical response was defined by analyzing the progression of the following main clinical signs: skin involvement (absence of purpura and/or leg ulcer), peripheral neuropathy (clinical and electrophysiologic improvement on 2 successive examinations), renal involvement (normalization of the serum creatinine level and disappearance of proteinuria and/or hematuria), and the absence of arthralgia. A complete clinical response in MC-related vasculitis was defined as improvement in all baseline clinical manifestations. A partial response was defined as improvement in at least half of the baseline clinical manifestations. All other patients were classified as clinical nonresponders. A sustained virologic response was defined as the absence of detectable serum HCV RNA 6 months after cessation of antiviral therapy; the remaining patients were classified as virologic nonresponders. A complete immunologic response was defined as the absence of serum cryoglobulin, and a partial immunologic response was defined as a >50% decrease in the baseline cryoglobulin level.

A clinical relapse was defined as the reappearance of clinical signs of vasculitis, a virologic relapse was defined as the reappearance of detectable HCV RNA, and an immunologic relapse was defined as the reappearance of serum cryoglobulin. A lymphoma response was defined by physical examination (presence of lymphadenopathy and/or splenomegaly) and peripheral blood and computed tomography analyses. Complete remission of lymphoma was defined as the total disappearance of the lymphomatous mass, and a partial remission was defined as a 50% reduction in the mass.

Statistical analysis. Data are presented as the mean ± SD for continuous variables and the percentage for qualitative variables. Fisher's exact test was used to compare qualitative variables, and the nonparametric Mann-Whitney U test was used to compare continuous variables. Wilcoxon's test was used to compare paired continuous variables. *P* values less than 0.05 were considered significant. Statistical analyses were performed using GraphPad Prism version 4.0 and InStat version 3.0 for Windows (GraphPad Software, San Diego, CA).

RESULTS

Patient characteristics. Two groups of patients with severe HCV-related vasculitis were assessed according to the treatment used: 1) rituximab and PEG–IFN alfa-2b plus ribavirin for antiviral-naive patients (n = 9) and patients who were resistant to or experienced a relapse after receiving previous antiviral therapy (n = 11), and 2) rituximab alone for patients for whom antiviral therapy was contraindicated or in whom tolerance to antiviral therapy was poor (n = 12).

The main characteristics of the patients are shown in Table 1. Thirty-two patients with HCV-related vasculitis (14 men and 18 women, mean ± SD age 59 ± 12 years [range 36–84 years]) were included. The mean ± SD duration of HCV-related vasculitis was 31 ± 42 months (range 2–192 months, median 12.5 months). Eight patients (25%) had a non-Hodgkin's B cell lymphoma (marginal zone [n = 4], lymphocytic [n = 1], lymphoplasmacytic [n = 1], diffuse large B cell [n = 1], and splenic with villous lymphocytes [n = 1]). All but 3 patients had MC, with a mean ± SD cryoglobulin level of 1.03 ± 0.78 gm/liter (range 0.06–2.95). The mean ± SD serum C4 level was 0.08 ± 0.09 gm/liter (range 0.01–0.35), the mean ± SD serum RF level was 236 ± 387 IU/liter (range 5–1,927), and the mean ± SD serum IgM level was 2.5 ± 3.2 gm/liter (range 0.35–16). Fifteen (47%) of 32 patients had elevated serum alanine aminotransferase (ALT) levels, and the mean ALT concentration was 1.3-fold the upper limit of the normal value. No significant difference was observed between patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin and those treated with rituximab alone, except for a higher cryoglobulin level in patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin (*P* = 0.04).

Efficacy of treatment. The clinical, immunologic, and virologic efficacy of rituximab in the 2 groups of patients is summarized in Table 2. Clinical improvement was observed after a mean ± SD period of 6.8 ± 4.7 months for patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin and 3.5 ± 1.3 months for those treated with rituximab alone; an immunologic response in these patients was observed after a mean ± SD period of 7.0 ± 3.3 months and 5.0 ± 2.1 months, respectively. No difference was observed regarding the clinical, immunologic, and virologic efficacy of rituximab and PEG–IFN alfa-2b plus ribavirin between antiviral-naive pa-

Table 2. Clinical, immunologic, and virologic efficacy of therapy*

	Rituximab with PEG–IFN alfa-2b plus ribavirin (n = 20)	Rituximab only (n = 12)
Clinical response		
Complete	16/20 (80)	7/12 (58)
Partial	3/20 (15)	1/12 (9)
Nonresponder	1/20 (5)	4/12 (33)
Relapse	3/20 (15)	4/12 (33)
Immunologic response		
Complete	12/18 (67)	5/11 (46)
Partial	6/18 (33)	4/11 (36)
Nonresponder	0/18 (0)	2/11 (18)
Relapse	5/18 (28)	6/12 (50)
Virologic response		
Sustained	11/20 (55)	0/12 (0)
Nonresponder	9/20 (45)	12/12 (100)

* Values are the number (%) of patients. None of the differences were significant. PEG–IFN alfa-2b = PEGylated interferon alfa-2b.

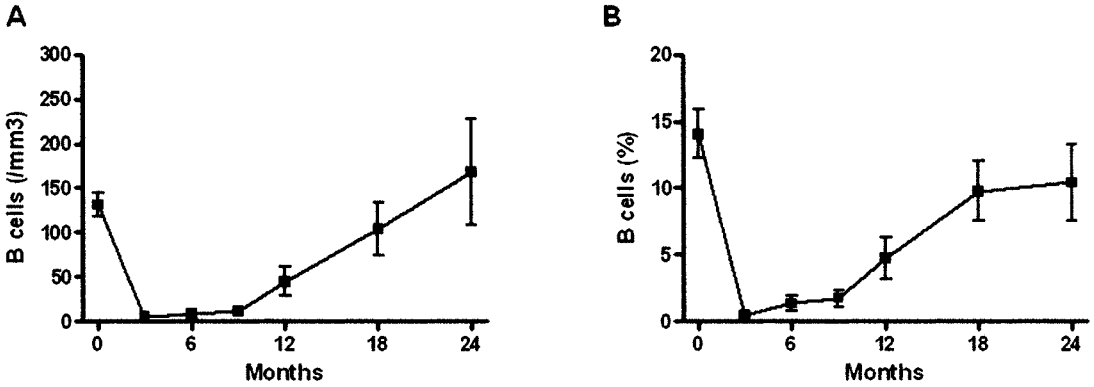

Figure 1. Dynamics of CD19+ B cell depletion and recovery during and after treatment with rituximab in patients with hepatitis C virus–related vasculitis. Values are the mean ± SD absolute number of CD19+ cells per mm^3 (**A**) and the mean ± SD percentage of CD19+ cells among total lymphocytes (**B**).

tients and antiviral-resistant or antiviral-relapser patients, except for a trend toward more frequent complete immunologic responses in antiviral-resistant or antiviral-relapser patients (complete response in 90% and partial response in 10%) than in antiviral-naive patients (complete response in 38% and partial response in 62%; $P = 0.07$).

B cell depletion in the peripheral blood was achieved in 26 (96%) of 27 patients (data were not available for 5 patients). The mean ± SD number of CD19+ cells at baseline (131 ± 73/mm^3 [14 ± 9%]) dropped to 5 ± 10/mm^3 (0.5 ± 0.8%) 3 months after administration of the first rituximab infusion. Recovery of the B cell count always began after 6 months (median delay 12 months [mean ± SD 10 ± 2 months]) (Figure 1). The mean and median delays until B cell recovery were slightly longer in patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin than in those treated with rituximab alone (11 ± 2 months versus 9 ± 4 months and 12 months versus 9 months, respectively; $P = 0.47$).

The time course of serum cryoglobulin, C4, RF, and IgM levels in the whole cohort of 32 patients is detailed in Figure 2. The mean ± SD cryoglobulin level decreased from 1.03 ± 0.78 gm/liter to 0.23 ± 0.39 gm/liter at 6 months ($P < 0.001$) and to 0.26 ± 0.33 gm/liter at the end of followup ($P < 0.001$). Serum cryoglobulin disappeared in 17 patients (59%) and decreased by more than 50% from the baseline level in 10 patients (34%). The C4 complement level increased from 0.08 ± 0.09 gm/liter at baseline to 0.17 ± 0.10 gm/liter at 6 months ($P < 0.001$) and to 0.16 ± 0.09 gm/liter at the end of followup ($P < 0.001$). RF levels decreased from 236 ± 387 IU/liter at baseline to 86 ± 136 IU/liter at 6 months ($P = 0.03$) and to 76 ± 155 IU/liter at the end of followup ($P = 0.003$). IgM levels decreased from 2.5 ± 3.2 gm/liter at baseline to 0.9 ± 0.4 gm/liter at 6 months ($P = 0.004$) and to 1.3 ± 1.2 gm/liter at the end of followup ($P = 0.02$).

Figure 3 shows the time course of the cryoglobulin and C4 levels in patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin and those treated with rituximab alone. Interestingly, in patients treated with rituximab alone, after a slight increase between 6 months and 12 months, the mean cryoglobulin level decreased from 0.60 gm/liter at 12 months to 0.24 gm/liter at 18 months ($P = 0.17$); the cryoglobulin level was reassessed at 18 months in 5 of 12 patients, 3 of whom were re-treated with rituximab at months 10, 12, and 17.

The time course of the HCV load in patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin and those treated with rituximab alone is shown in Figure 4. The combination of rituximab and PEG–IFN alfa-2b plus ribavirin induced a significant decrease in the HCV load, from a mean ± SD of 5.8 ± 0.5 log copies/ml at baseline to 2.3 ± 2.7 log copies/ml at the end of followup ($P < 0.001$), with 11 (55%) of 20 patients showing a sustained virologic response. In patients treated with rituximab alone (without antiviral therapy), the HCV load was 6.1 ± 0.8 log copies/ml at baseline, 5.4 ± 0.8 log copies/ml at 12 months, and 6.0 ± 0.5 log copies/ml at 24 months (P not significant). None of the patients in this group had a sustained virologic response.

ALT levels remained stable during followup (mean ± SD 1.4 ± 1.0–fold the upper limit of normal at baseline versus 1.3 ± 0.9–fold at the end of followup) in

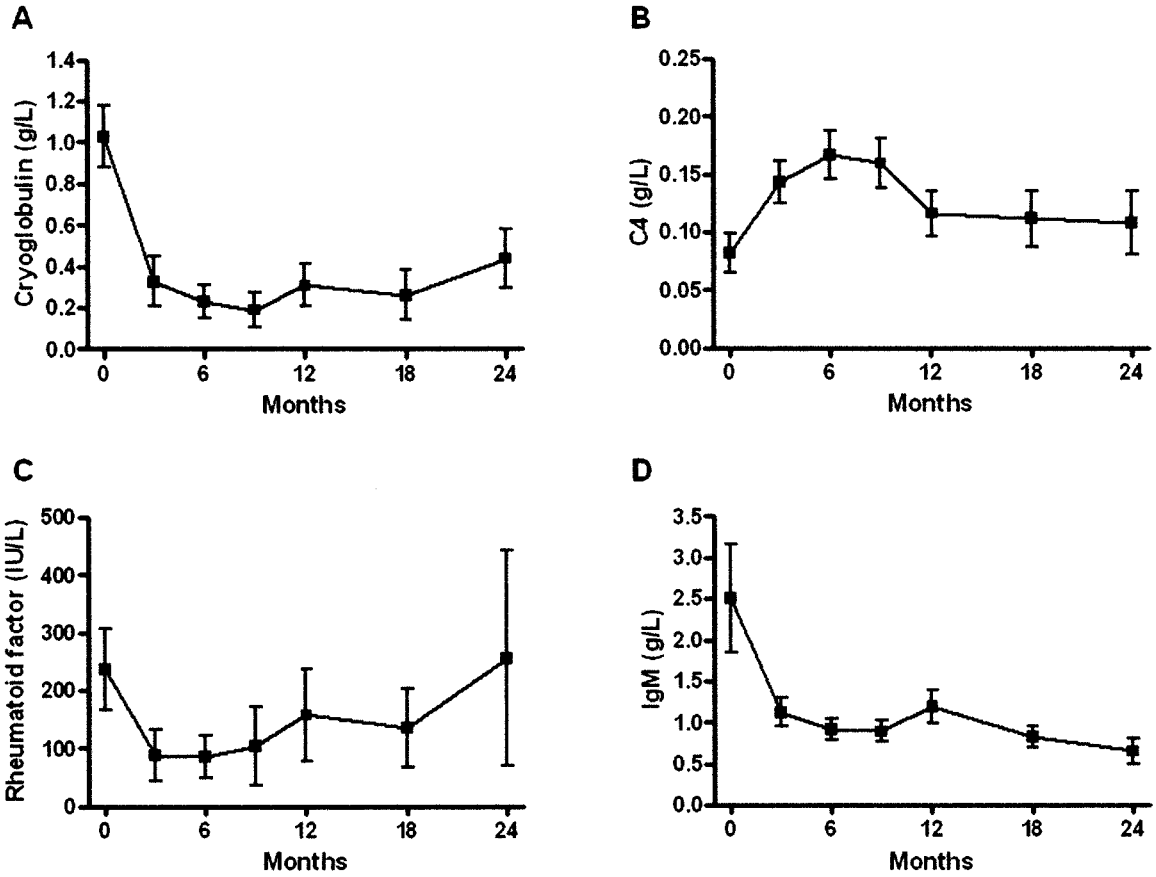

Figure 2. Time course of serum cryoglobulin (**A**), C4 complement (**B**), rheumatoid factor (**C**), and IgM (**D**) levels in the cohort of 32 patients with hepatitis C virus–related vasculitis treated with rituximab with or without PEGylated interferon alfa-2b plus ribavirin. Values are the mean ± SD.

the whole cohort. In patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin, the ALT level decreased from 1.3 ± 0.7–fold the upper limit of normal at baseline to 1.1 ± 0.4 at the end of followup, while in those treated with rituximab alone, the ALT level increased slightly, from 1.5 ± 1.4–fold the upper limit of normal at baseline to 1.7 ± 1.4–fold at the end of followup. A total of 3 patients died (1 died of hepatocarcinoma, 1 died of progressive deterioration of the general health status, and 1 died of an unknown cause).

Clinical and immunologic relapse and re-treatment with rituximab. After a mean ± SD followup of 23 ± 12 months (range 6–44 months), 7 patients experienced a clinical relapse (22%), including 3 (15%) of 20 patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin and 4 (33%) of 12 patients treated with rituximab alone ($P = 0.34$), a mean ± SD 18 ± 7 months after the initiation of rituximab therapy. Eleven patients experienced an immunologic relapse (34%), including 5 patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin and 6 patients treated with rituximab alone, 15 ± 6 months after the initiation of rituximab therapy. All clinical relapses were associated with an immunologic relapse. All clinical and immunologic relapsers (n = 11) were HCV RNA positive and were either virologic nonresponders after treatment with rituximab and PEG–IFN alfa-2b plus ribavirin (n = 5) or had been treated with rituximab alone without antiviral therapy (n = 6). Among the clinical and/or immunologic relapsers, 7 (78%) of 9 experienced B cell recovery starting after a mean ± SD delay of 11 ± 4 months, with a B cell count of $>50/mm^3$ reached after 16 ± 3 months; 2 (22%) of 9 patients did not experience B cell recovery, and data were not available during followup for the 2 remaining patients.

Six patients were re-treated with rituximab. Among these 6 patients, 5 had an immunologic and clinical relapse, and 1 had only an immunologic relapse.

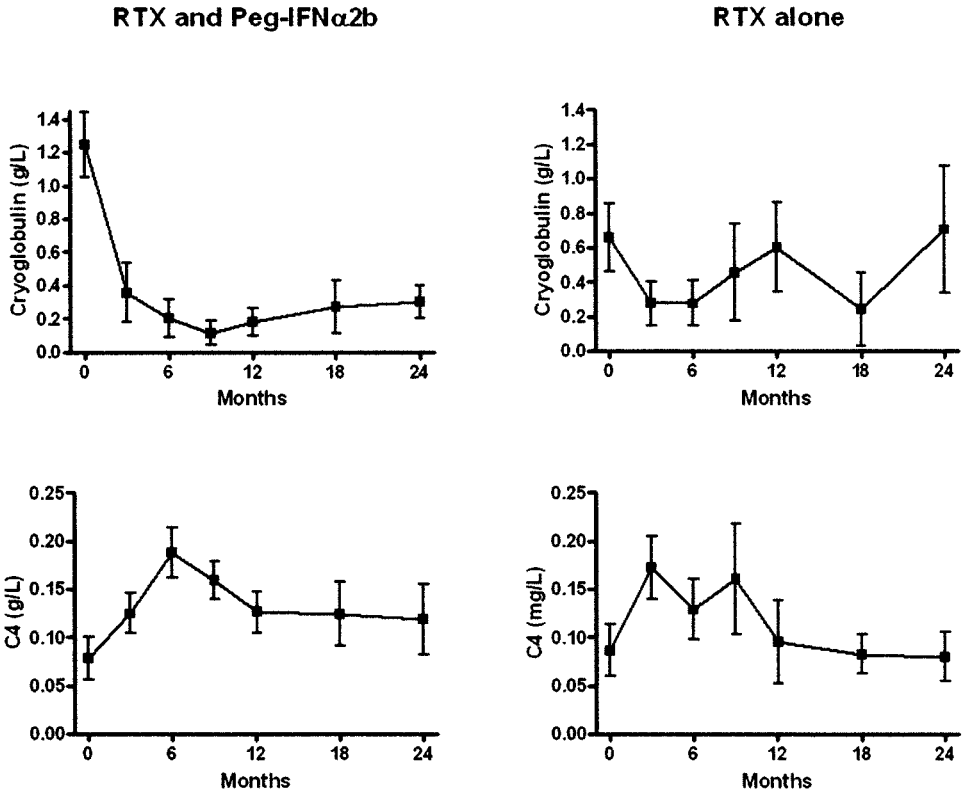

Figure 3. Time course of serum cryoglobulin and C4 complement levels in patients treated with rituximab (RTX) and PEGylated interferon alfa-2b (PEG–IFN alfa-2b) plus ribavirin and those treated with rituximab alone. Values are the mean ± SD.

Four of these patients had been initially treated with rituximab alone, and 2 had received initial treatment with rituximab and PEG–IFN alfa-2b plus ribavirin. The re-treatment schedules for these patients were as follows: 4 intravenous infusions of rituximab at a dose of 375 mg/m^2 on days 1, 8, 15, and 22 (n = 2), 1 intravenous infusion of rituximab at a dose of 1,000 mg (n = 2), 2 intravenous infusions of rituximab at a dose of 1,000 mg

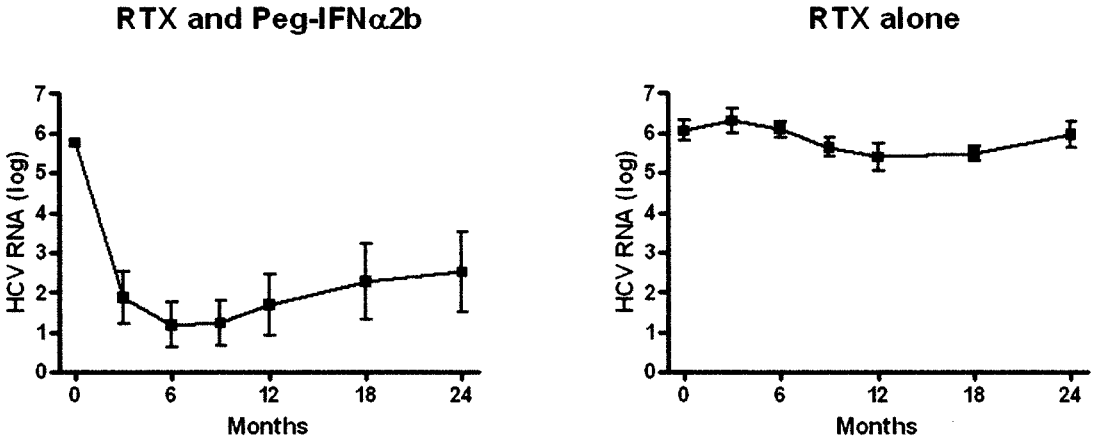

Figure 4. Time course of the hepatitis C virus (HCV) load in patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin and those treated with rituximab alone. Values are the mean ± SD. See Figure 3 for other definitions.

on days 1 and 15 (n = 1), and 1 intravenous infusion of rituximab at a dose of 375 mg/m^2 (n = 1). Rituximab was given along with PEG–IFN alfa-2b plus ribavirin in 2 patients, corticosteroids in 2 patients, cyclophosphamide in 1 patient, and methotrexate in 1 patient. After re-treatment with a mean ± SD followup of 9.7 ± 3.6 months (range 3–13 months), all patients experienced a complete clinical response, 3 (50%) of 6 patients had a complete immunologic response, and 3 patients (50%) had a partial immunologic response.

Tolerability of treatment. Rituximab was well tolerated in 24 (75%) of 32 patients. In the 8 remaining patients, side effects included serum sickness (n = 6), early-onset neutropenia (n = 1), late-onset neutropenia (n = 1), varicella zoster virus infection (n = 1), and subcutaneous extravasation of rituximab (n = 1). Among those treated with rituximab and PEG–IFN alfa-2b plus ribavirin, interruption of antiviral therapy was required in 5 patients, because of hematologic toxicity in 2 patients (at months 3 and 9, respectively), a flare of skin psoriasis in 1 patient (at month 8), hepatocarcinoma in 1 patient (at month 7), and poor compliance with antiviral therapy in 1 patient (at month 9).

DISCUSSION

In this study, the most striking observations were the good efficacy and tolerability of rituximab in a large series of patients with severe and/or refractory HCV-related vasculitis with long followup, the consistent association between relapses and the absence of virologic control, and the similarly good efficacy of rituximab after repeated infusions. We enrolled 11 patients previously reported by our group (20) and for whom we report 14 additional months of followup (33 months of followup versus 19 months in the previous study) and 21 previously unreported patients. This additional followup is particularly interesting in terms of assessing the clinical response to treatment, notably the outcome in patients with neurologic involvement, the rates of clinical and immunologic relapses, and the efficacy of repeated rituximab infusions in patients who experienced a relapse. Indeed, among the 11 previously reported patients, 5 exhibited an immunologic relapse, and 3 had a clinical relapse during the extended followup period.

We observed good efficacy of rituximab in both groups of patients, with a better response in those treated with rituximab and PEG–IFN alfa-2b plus ribavirin. Among patients treated with rituximab and PEG–IFN alfa-2b plus ribavirin, 95% exhibited a clinical response (complete response in 80%), 100% exhibited an immunologic response (complete response in 67%), and 55% showed a sustained virologic response. Among patients treated with rituximab alone, 67% exhibited a clinical response (complete response in 58%), 82% exhibited an immunologic response (complete response in 46%), and no patient exhibited a virologic response. Moreover, our data showed that the efficacy of rituximab and PEG–IFN alfa-2b plus ribavirin was similar in antiviral-naive patients and antiviral-resistant or antiviral-relapser patients, suggesting similar therapeutic management in both groups of patients.

Here, we report the rates of clinical and immunologic relapses (22% and 34%, respectively) in our study population after a mean followup of almost 2 years. Interestingly, all of the clinical and immunologic relapses were associated with the absence of virologic clearance. These findings underline the importance of targeting the viral trigger (HCV) and not only the B cells. In accordance with the remarkable efficacy of rituximab and PEG–IFN alfa-2b plus ribavirin observed in our previous study (20), these results support the use of rituximab in combination with antiviral therapy in order to achieve viral eradication, except in cases in which antiviral therapy is contraindicated. The proposed therapeutic regimen consisting of weekly administration of 4 intravenous infusions of rituximab at a dose of 375 mg/m^2 followed by PEG–IFN alfa-2b plus ribavirin for at least 12 months appears very effective for achieving this objective.

B cell depletion and B cell recovery were correlated with the clinical and immunologic responses. B cell depletion was associated with a clinical response in 92% of patients and with an immunologic response in 96% of patients, while the absence of B cell depletion was associated with the absence of clinical and immunologic responses. In addition, among patients with B cell recovery of >50 cells/mm^3, 36% experienced a clinical relapse, and 70% experienced an immunologic relapse. Interestingly, among the patients with B cell recovery and an absence of virologic clearance, 57% experienced a clinical relapse, and 100% experienced an immunologic relapse, whereas none of those with B cell recovery and a sustained virologic response experienced a relapse.

We assessed the efficacy of repeated infusions of rituximab in patients who experienced a clinical and/or immunologic relapse. The clinical and immunologic efficacy of rituximab after repeated infusions was similar to that after induction therapy, with 100% of the re-treated patients exhibiting a complete clinical response, 50% exhibiting a complete immunologic response, and

50% exhibiting a partial immunologic response. These findings suggest that in patients with HCV-related vasculitis who were initial responders to rituximab, retreatment with rituximab is an effective therapeutic option. However, the findings also raise the issue of administering maintenance therapy with rituximab in patients who do not experience a virologic response.

Tolerability of rituximab was good in three-fourths of the patients. In the remaining one-fourth, the main side effect attributable to rituximab was the occurrence of serum sickness (19%) (15,26,27). Moreover, patients treated with rituximab without antiviral therapy showed stable levels of HCV RNA (6.1 log copies/ml at baseline, 5.4 log copies/ml at 12 months, and 6.0 log copies/ml at 24 months) and a slight increase in ALT levels (from 1.5-fold the upper limit of normal at baseline to 1.7-fold at the end of followup). These findings are reassuring regarding the use of rituximab in HCV-infected patients, even in the absence of antiviral therapy.

Last, our findings confirm the encouraging results of rituximab treatment demonstrated in previous studies, which were mainly limited by their small size and the short duration of followup (mean followup 9.7 months, versus 23 months in the current study). Indeed, a recent review of previous studies revealed great efficacy of this treatment for the main signs of vasculitis, with a clinical response observed in 80–93% patients, relapse of MC in 39% of patients, and successful retreatment with rituximab in 57% of patients. As in our study, rituximab was well tolerated (19).

In conclusion, rituximab is an effective treatment of severe HCV-related vasculitis, with the combination of rituximab and PEG–IFN alfa-2b plus ribavirin being particularly effective. Relapses are consistently associated with the absence of virologic control, and repeated rituximab infusions seem to have the same clinical and immunologic efficacy as the initial therapy.

AUTHOR CONTRIBUTIONS

All authors were involved in drafting the article or revising it critically for important intellectual content, and all authors approved the final version to be published. Dr. Cacoub had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study conception and design. Terrier, Cacoub.

Acquisition of data. Terrier, Saadoun, Sène, Sellam, Pérard, Coppéré, Karras, Blanc, Buchler, Plaisier, Ghillani, Rosenzwajg, Cacoub.

Analysis and interpretation of data. Terrier, Saadoun, Sène, Sellam, Pérard, Coppéré, Karras, Blanc, Buchler, Plaisier, Ghillani, Rosenzwajg, Cacoub.

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Errata

In the article by Tubach et al in the July 2009 issue of *Arthritis & Rheumatism* (pages 1884–1894), the name of the fourteenth author was spelled incorrectly. The author's correct name is O. Lortholary. Also, the term "anti-TNF therapy" was erroneously changed to "anti-TNF mAb therapy" at a late proof stage; "anti-TNF mAb therapy should read simply "anti-TNF therapy" in the following instances: page 1884 line 2 of left column and lines 1, 3, and 11 of right column, page 1885 line 5 of left column and lines 3, 6, 8, 12, 17, 22, and 28 of right column, page 1886 lines 16 and 26 of left column and last line of right column, page 1887 lines 1, 24, and 41 of left column, page 1888 lines 11, 13, 20, and 25 of left column and lines 4, 11, and 16 of right column, page 1889 lines 4 and 36 of right column, page 1891 line 15 of left column, page 1892 line 3 of right column ("anti-TNF mAb therapy" is correct in the other instances where it appears).

In the title of the article by Meinecke et al in the July 2009 issue of *Arthritis & Rheumatism* (pages 2065–2070), the term "The Small Ubiquitin-like Modifier" should have been "Small Ubiquitin-like Modifier 1"; thus, the correct title should be "Small Ubiquitin-like Modifier 1 Mediates the Resistance of Prosthesis-Loosening Fibroblast-like Synoviocytes Against Fas-Induced Apoptosis."

We regret the errors.