Hepatitis C: Cryoglobulinemia and non-Hodgkin lymphoma

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RESUMEN

La infección por el virus de la hepatitis C juega un papel principal en la patogénesis de la crioglobulinemia mixta, promoviendo la activación y expansión de las células B. Estos reajustes moleculares inducen la síntesis de crioglobulinas y la aparición de la vasculitis crioglobulinémica. El aclaramiento del virus provoca la resolución de las manifestaciones clínicas y de las alteraciones inmunológicas observadas en la crioglobulinemia mixta en un alto porcentaje de los pacientes, pero no en todos. En algunos casos, la crioglobulinemia puede aparecer tras la respuesta virológica sostenida. Muchos mecanismos de la patogénesis de la crioglobulinemia mixta están fuertemente relacionados con la infección por VHC y, cuando el virus es eliminado, hay una mejoría en el curso de la enfermedad. Aun así, los pasos independientes relacionados con otros factores no mejoran tras la erradicación del virus.

En algunos tipos de linfomas no-Hodgkin de bajo grado (linfoma linfoplasmocítico y linfoma de la zona marginal) la respuesta sostenida tras el aclaramiento viral induce una remisión de la neoplasia. El VHC tiene un papel secundario en los linfomas agresivos y el aclaramiento del virus puede no inducir la remisión, pero puede disminuir la hepatotoxicidad asociada a la quimioterapia.

Por tanto, en la hepatitis C crónica, la combinación de interferón pegilado y ribavirina es altamente recomendable en el tratamiento de la crioglobulinemia mixta sintomática y los linfomas no-Hodgkin asociados al VHC.

Palabras clave: Hepatitis C. Crioglobulinemia. Linfoma no-Hodgkin.

ABSTRACT

Hepatitis C virus infection plays a major role in the pathogenesis of mixed cryoglobulinemia, promoting activation and expansion of B cells. These molecular rearrangements induce synthesis of cryoglobulins and the appearance of cryoglobulinemic vasculitis. Clearance of the virus promotes resolution of the clinical manifestations and immunological disorders seen in mixed cryoglobulinemia in a large percentage of patients, but not in all. In some cases, cryoglobulinemia could appear after sustained response. Several steps in the pathogenesis of mixed cryoglobulinemia are strongly related to HCV infection and when the virus is eliminated, the disease course improves. However, independent steps related to other factors do not improve following viral clearance.

In some types of low-grade non-Hodgkin lymphoma (lymphoplasmocytic lymphoma, marginal zone lymphoma) sustained response following antiviral treatment induces remission of the neoplasm. HCV has a minor role in aggressive lymphomas and clearance of the virus may not induce remission, but could decrease the hepatotoxicity associated with the chemotherapy.

Therefore, in chronic hepatitis C, the combination of peginterferon + ribavirin is strongly recommended in treating symptomatic mixed cryoglobulinemia and HCV-related non-Hodgkin lymphomas.

Key words: Hepatitis C. Cryoglobulinemia. Non-Hodgkin lymphoma.

INTRODUCTION

Recent World Health Organization estimates are that 123 million individuals are infected with hepatitis C. This infection is responsible not only for chronic hepatitis that could lead to cirrhosis, end-stage liver disease and hepatocellular carcinoma, but also extrahepatic diseases. The wide range of extrahepatic manifestations suggests that chronic hepatitis C ought to be considered a systemic disease. The extrahepatic manifestations of hepatitis C have been classified into four different categories according to the implication of chronic hepatitis C infection in the pathogenesis. Mixed cryoglobulinemia is due mainly to hepatitis C and the virus is the major etiologic factor. Non-Hodgkin lymphoma has been found to be related to HCV infection, but a pathogenetic link has not been re-
Mixed cryoglobulinemia (MC) is a systemic vasculitis that mainly affects small and, less frequently, medium size vessels. MC is characterized by the proliferation of B-cell clones producing pathogenic IgM-rheumatoid factor. Three types of MC have been described based on the immunoglobulin complexes found in the precipitate: Type I is a monoclonal IgM in the Waldenstrom macroglobulinemia site; type II is a monoclonal IgM-rheumatoid factor plus polyclonal IgG; and type III is an oligoclonal IgM rheumatoid factor plus polyclonal IgG. Mixed cryoglobulinemia (type II and type III) has been found strongly related to hepatitis C. In a cohort of 168 patients with mixed cryoglobulinemia, anti-HCV was observed in 155 (92%) and HCV RNA in 152 out of 155 (98%) patients (1). The data supporting the role of HCV in the pathogenesis of MC include: Anti-HCV has been found in the polyclonal IgG precipitates; HCV RNA is usually present in the cryoprecipitate; and, lastly, cryoglobulins are more often seen in patients with hepatitis C than other liver diseases such as hepatitis B.

The pathogenesis of MC-related HCV is complex and is likely to involve several mechanisms (2). HCV seems to induce a polyclonal activation of B cells after interaction between protein E2 and CD81 receptor. The activation of STAT3 by core protein could promote expansion and modifications in B cell activation causing over-expression of CD69, CD71, CD86 and > 5-fold increase in CXCR3, together with a decrease in the expression of CXCR4. These molecular events lead to several immunological disorders responsible for monoclonal expansion of B-cell-producing cryoglobulins, and alterations in T-cell response.

Patients with hepatitis C and mixed cryoglobulinemia show a decrease in regulatory T cells such as CD25+ (3). Moreover, Th1 response is enhanced with higher levels of TNF, IFN-α and interleukin-12, interleukin-18, and decreased concentrations of Th2 interleukin such as interleukin-10 (4). The immunological disturbances are in accord with pathological data demonstrating that neutrophilic infiltration with leukocytoclastic changes. Typical immunocomplex-mediated vasculitis was rarely found, while the presence of lympho-histiocytic infiltrates suggests a T cell-mediated pathogenesis, with only a minor role for the humoral immunity. HCV RNA levels in circulating lymphocytes obtained from patients with cryoglobulinemia are higher than in non-cryoglobulinemic patients, despite there being no differences in serum HCV RNA concentrations. Several cells such as B lymphocytes harbor and sustain infection by HCV. The minus strand of HCV RNA has been detected in peripheral blood mononuclear cells from patients with cryoglobulinemia, but not in patients with chronic or acute hepatitis C without this extrahepatic manifestation (5). These data support the hypothesis that HCV infects B lymphocytes and induces changes in the maturation of B cells, and promotes the production of cryoglobulins.

Data analyzing the role of hepatitis C in the manifestation of cryoglobulinemia are scarce. The solubility of the proteins depends on the concentration, temperature, pH, ionic strength of the solution, and surface charge. Several changes have been proposed to explain the production of cryoglobulins in the presence of hepatitis C: a) Changes in the primary structure of the variable portion of the immunoglobulins; b) reduced concentration of sialic acid; and c) reduced amount of galactose in the Fc region of the immunoglobulins (6).

The main clinical manifestations are purpura, arthralgia, weakness, and renal or neurologic involvement. Cryoglobulinemia has been found in association with steatosis and fibrosis progression in hepatitis C (7), and cryoglobulins are more often seen in cirrhotic patients. In a meta-analysis of 19 studies and 2,323 patients, cryoglobulinemia was found associated with cirrhosis after adjustment for age, gender and estimated duration of disease (8). Specific cryoglobulins play a role in the development of membranous proliferative glomerulonephritis lesions and skin lesions (9), but involvement in the pro-
gression of liver fibrosis has not been demonstrated, as yet. Cryoglobulins may also be merely markers of advanced fibrosis, as has been reported for antiphospholipid antibodies (10).

The natural history of cryoglobulinemia has been addressed by Ferri et al. (1) and Sene et al. (11) in several large cohorts. The studies indicated that, from a clinical point of view, purpura occurred in 40-81%, arthralgias in 43-72% and weakness in 80%. Peripheral neuropathy was present in 47-58% and renal involvement in 10-20%. Despite no strong differences having been reported between hepatitis C patients with and without mixed cryoglobulinemia, the former were more often older females with a longer duration of infection, and with genotype 2 or 3 being slightly overrepresented. Epidemiological features of patients with HCV-related mixed cryoglobulinemia were similar to patients with hepatitis C without cryoglobulinemia with respect to genotype distribution and fibrosis stage. Low C4 and CH50 levels together with normal or raised C3 were seen in the majority of cases. Renal involvement in 33%, end-stage-liver disease 13%, B-cell lymphoma 13%, hepatocellular carcinoma 10% and miscellaneous 30% were the main causes of death. The cause of death was MC-related in 64%, “possibly related” in 13% and “non-related” in 23%. The independent factors influencing survival were age > 60 years, female gender, and renal involvement. Factors associated with symptomatic mixed cryoglobulinemia and poorer prognosis were age-at-diagnosis, levels of cryoglobulins in serum at diagnosis, duration of HCV infection and type of cryoglobulinemia (type II cryoglobulinemia having poorer prognosis) (41). However, during follow-up > 40% of cases had altered features of cryocrit and, thus, the type of mixed cryoglobulinemia. Fewer than 60% of patients diagnosed as type III mixed cryoglobulinemia changed to type II.

The effect of HCV clearance on cryoglobulinemia has been addressed from several points of view. Since mixed cryoglobulinemia is an immune-driven disease resulting from chronic HCV infection, the main aims in the treatment of this entity focus on viral eradication. The impact of sustained HCV clearance on cryoglobulinemia could be addressed by assessing several aspects such as ameliorating clinical manifestation of the disease and avoiding life-threatening complications and, as well, reversing the immunologic derangement that is producing the cryoglobulins. In the majority of patients, achieving sustained viral response is associated with an improvement in clinical manifestations of the disease such as purpura, arthralgia or weakness, and a decrease in the cryocrit level to below the detection threshold. The reappearance of HCV RNA in relapers is accompanied, in the majority of cases, by a relapse in the mixed cryoglobulinemic vasculitis. However, the development of cryoglobulinemia following the clearance of the virus has been reported in sustained responders without cryoglobulins at baseline (12). This event is not clearly understood. It could be explained as a consequence of the persistence of viral reservoirs that may support a continuous B-cell expansion; cryoglobulin production could become an autonomous process following HCV clearance, or it could be an immunological adverse event to peginterferon or of ribavirin administration.

First-line therapy in mixed cryoglobulinemia is antiviral therapy. However, in patients with renal involvement, severe neuropathy and life-threatening complications, several immunosuppressive drugs (corticosteroids, cyclophosphamide or azathioprine) or plasmapheresis need to be used before, or in conjunction with, the antiviral treatment. Sustained virological response was observed to increase in patients treated with peginterferon + ribavirin compared to patients receiving interferon alone or standard interferon + ribavirin (Fig. 1).

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Patients with mixed cryoglobulinemia treated with peginterferon + ribavirin demonstrated a wide range of sustained response rate between 44 and 58%, depending on the distribution of genotypes and the dose of peginterferon alfa 2b used. Peginterferon 1 µg/kg/week seems to lead to a higher relapse rate than full-dose 1.5 µg/kg/week (13). The cryocrit disappeared in 30-96%, and a clinical response was achieved in 62.5-89% of cases. In sustained responders, the cryocrit decreased from 1.1 ± 1.2 to 0.2 g/l; p < 0.001 and proteinuria from 4.4 ± 2.3 to 0.6 ± 0.9 g/d; p = 0.002 but, in non-responders, the cryocrit also fell from 1 ± 1.5 to 0.5 ± 0.9 g/l; p = 0.002 and proteinuria decreased from 4.1 ± 3.2 to 2 ± 3.1; p = ns. Sustained virological response was achieved in just under two thirds of patients (62.5%), disappearance of cryoglobulins in
just over a half of the patients (57.5%) and clinical remis-
sion was seen in 62.5% (Fig. 2) (14). Although, antiviral
therapy has been found to be useful in the management of
kidney diseases in patients with mix
cryoglobulinemia, no influence has been seen in serum creatinine
levels, glomerular filtration rate and glomerular lesions, even in
sustained responders. Viral clearance improves renal dis-
 ease, mainly due to suppression of B lymphocyte stimu-
lation and cryoglobulin production. However, persistent
immune complex deposition promotes damage and scler-
osis and, as such, patients should receive early treat-
ment to avoid non-reversible lesions in the kidney. In a cohort
of 18 patients with mixed cryoglobulinemic nephropathy
with standard (n = 14) or pegylated (n = 4) inter-
feron plus ribavirin, sustained response rate was 67%, de-
spite half the patients being infected by genotype. 1. Fur-
ther, cryoglobulins disappeared in 5 out of 12 responders,
but in none of the non-responders (15). Lastly, mild-to-
omodate peripheral neuropathy improves, or disappears,
following viral clearance in more than two thirds of cas-
es. However, severe cases show a very low response rate.

Tolerability and sustained response rate could be
quite similar in patients with cryoglobulinemic as in he-
patitis C patients without cryoglobulins (16). Indeed, as
reported in non-cryoglobulinemic patients, factors as-
associated with sustained response in mixed cryoglobu-
linemia were genotype 2 or 3, age < 50 years, low viral
load, non-Afro-American ethnicity, and female gender
(17). In the largest cohort studied, the independent
variable associated with sustained response was ab-
ence of renal insufficiency. When glomerular filtration
rate was < 70 ml/min complete clinical response rate
became a fifth in comparison with patients without re-
nal involvement (14).

NON-HODGKIN LYMPHOMA IN CHRONIC
HEPATITIS C

Hepatitis C appears to be closely associated with non-
Hodgkin lymphoma (NHL) in some countries, but not in
others. In Italy, the prevalence of hepatitis C in patients
with NHL was found to be between 8.9 and 37.1% with
an average of 19.8% in a cohort 2,668 cases from 18
studies. Conversely, in more than 300 cases of NHL in
Northern Europe, no patients showed antiHCV antibod-
ies. No association was seen in 2 studies from Canada,
but in USA the association depended on the individual
State. In Japan, the prevalence of hepatitis C ranged from
5.7-22.2% with an average of 11.3%. Matsuo et al. (18)
found the prevalence of hepatitis C was increase at least
5-fold in patients with non-Hodgkin lymphoma. A meta-
analysis that included 48 studies concluded that the
prevalence of HCV in patients with B-cell NHL was 15
percent; much higher than in the general population
(around 1.5%) and in patients with other hematologic
malignancies (2.9%) and suggested that HCV had an eti-
ologic role in NHL (19). Despite the variation in hepatitis
C prevalence, it seems that the relative risk for non-
Hodgkin lymphoma in patients with chronic hepatitis C
is 2- to 4-fold higher than in non-infected subjects; at
least in patients from Japan or the Mediterranean region,
but not in cases from North America or Northern Europe
(20). These data suggests that HCV-related induction of
non-Hodgkin lymphomas emerge from the interaction of
genetic and environmental factors. Interleukin-10 poly-
morphisms (such as 1082 GG) have been found to be as-
sociated with increased interleukin-10 production, and a
higher risk for the development of non-Hodgkin lymph-
oma in patients with chronic hepatitis C compared to
those with hepatitis C without lymphoma, or patients with
non-Hodgkin lymphoma but non-HCV infection (21).

The lymphomagenesis induced by HCV appears to be-
gin with HCV antigen-driven processes inducing a poly-
clonal activation of B cells following interaction between
protein E2 and CD81 receptor; the activation of STAT3
by core protein could promote expansion and modific-
ations in B cell activation. These molecular events lead to
B-cell monoclonal expansion, chromosomal transloca-
tion [t(14:18)], bcl-2 over-expression and, finally, to B-
cell lymphoma. Despite HCV RNA replicate without in-
termediate DNA not being integrated into the host
genome, the C virus could promote malignancies by oth-
er mechanisms such as oncogene activation, cellular
growth induction and apoptotic cell death inhibition (22).
There is increasing evidence supporting a possible role
for infectious agents in the development of several neo-
plasms such as Epstein Barr virus in Burkitt’s lymphoma,
parvovirus 19 in pure red cell aplasia, Helicobacter py-
lori in gastric B-cell lymphoma type MALT, and human
herpes virus Kaposi’s sarcoma. Non-Hodgkin lymphoma
in patients with chronic hepatitis C should be classified in
two groups: Lymphoma developing in the setting of

Fig. 2. Virological, clinical and immunological response to peginterfe-
ron + ribavirin in patients with chronic hepatitis C and mixed cryoglo-
bulinemia. Sustained virological response (SVR), clinical response (CR),
immunological response (IR). Data extracted from Ref. 14.
mixed cryoglobulinemia, and idiopathic forms. Non-Hodgkin lymphoma was observed in 5-15% of cases of mixed cryoglobulinemia associated with hepatitis C (23-25). Thus, the incidence of lymphoma in patients with mixed cryoglobulinemia is > 35-fold that expected in the general population. Histological types of lymphomas seen in mixed cryoglobulinemia are mainly non-aggressive lymphomas and include: Lymphoplasmocytic lymphoma, marginal zone lymphoma [nodal, splenic and extranodal (MALT form)], B-cell chronic lymphocytic leukemia and follicular lymphoma. In patients with hepatitis C without mixed cryoglobulinemia, the incidence of high-grade large-B-cell lymphomas and follicular lymphomas was higher than in patients with cryoglobulinemia, and similar to that in patients not infected by HCV (26,27) (Table I).

The prognosis of diffuse large B-cell lymphoma associated with hepatitis C is poorer than lymphomas non-related to hepatitis C. A cohort of 26 cases of non-Hodgkin lymphoma associated with hepatitis C was compared with matched cases of non-HCV-infected patients. Diffuse large B cell lymphomas were more often transformed from low grade lymphomas (32%) in HCV positive patients than in HCV-negative patients (6%). Moreover, splenic involvement was more often seen in HCV-positive patients. Patients with hepatitis C and diffuse large B cell lymphoma showed poorer overall survival and, at two years, the survival rate was 56% in HCV positive versus 80% in HCV-negative. Further, liver toxicity was more often seen in HCV patients, and which appeared in two thirds of cases (15 out of 23 patients) (28). Thus, hepatitis C has a negative influence on the natural history of non-Hodgkin lymphoma.

Lymphomas with the closest links to hepatitis C include: Lymphoplasmocytic lymphoma and marginal zone lymphomas. Splenic lymphoma with villous lymphocytes is a chronic B-cell lymphoproliferative disorder histologically indistinguishable, in spleen and bone marrow, from splenic marginal zone lymphoma (29). There is increasing evidence, in certain low-grade B-cell lymphomas, that treatment of the hepatitis C with combined antiviral therapy such as peginterferon + ribavirin can lead to remission of the lymphoma. Splenic lymphoma with villous lymphocytes associated with hepatitis C and mixed cryoglobulinemia regresses after effective antiviral therapy in > 80% of cases (30). In the largest cohort studied, 18 cases were treated with interferon (standard or pegylated) + ribavirin. Sustained viral response was seen in 14 out 18 (77.8%) and this was associated with hematological response. Of the 4 non-responder patients, hematologic response was complete in 2 and partial in the other 2 cases (31). However, despite complete hematological and viral response, monoclonal immunoglobulin gene rearrangement persisted. These findings contrast with previous reports in which the disappearance of B-cell clones was frequently seen following viral eradication, at least in chronic hepatitis C, with or without cryoglobulinemia. This discrepancy may reflect differences in tumor load or in survival requirements. In splenic lymphoma with villous lymphocytes, some clones may survive even after effective eradication of the C virus (32).

The hematological response has been reported stable for many years following the clearance of the C virus.

In low grade non-Hodgkin lymphoma strongly related to hepatitis C and mixed cryoglobulinemia such as lymphoplasmocytoid immunocytomas, complete remission of the lymphoma has been achieved following successful clearance of the virus (33). Further, marginal zone lymphomas including extranodal cases such as MALT type lymphoma from stomach, duodenum or ileum reached complete remission when the associated hepatitis C was successfully cleared. Thus, HCV infection plays a role in the development of MALT lymphoma similar to that of Helicobacter pylori and, under some circumstances, a synergy between the 2 agents could promote extranodal low-grade lymphomas (34).

Lastly, eradication of HCV has been implicated in the remission of natural killer cell lymphoma of the liver, despite this lymphoma belonging to an aggressive form (35). Also, regression of advanced non-splenic marginal lymphoma has been reported following the treatment of hepatitis C (36). In a cohort of 12 low-grade non-Hodgkin lymphomas associated with hepatitis C, antiviral treatment achieved complete remission in 7, partial response in 2, stable disease in 2 and progressive disease in 1 patient. Lymphoma remission correlated with viral clearance. All non-responders had not hematologic response. Tolerance to combined peginterferon + ribavirin was quite similar to that reported for patients with hepatitis C without malignancies (37).

Table I. Percentage of B-NHL subtypes among patients with hepatitis and mixed cryoglobulinemia and patients with hepatitis C without mixed cryoglobulinemia in comparison with International lymphoma study

<table>
<thead>
<tr>
<th>Lymphoma type</th>
<th>HCV-cryoglobulinemia</th>
<th>HCV-cryoglobulinemia</th>
<th>International lymphoma study</th>
</tr>
</thead>
<tbody>
<tr>
<td>Lymphoplasmacytic</td>
<td>33%</td>
<td>5%</td>
<td>1.2%</td>
</tr>
<tr>
<td>MZL: Nodal</td>
<td>13%</td>
<td>3%</td>
<td>1.8%</td>
</tr>
<tr>
<td>MZL: Splenic</td>
<td>5%</td>
<td>2%</td>
<td>&lt; 1%</td>
</tr>
<tr>
<td>MZL: Extranodal</td>
<td>9%</td>
<td>42%</td>
<td>7.6%</td>
</tr>
<tr>
<td>Follicular</td>
<td>12%</td>
<td>20%</td>
<td>22.1%</td>
</tr>
<tr>
<td>Diffuse large B-cell</td>
<td>25%</td>
<td>26%</td>
<td>30.6%</td>
</tr>
<tr>
<td>Patients with B-NHL (n)</td>
<td>137</td>
<td>85</td>
<td>1.069</td>
</tr>
</tbody>
</table>
Lymphomagenesis is a multi-step process and hepatitis C seems to play an important role in some types of lymphomas and, in which, early eradication promotes lymphoma regression (38). However, aggressive lymphomas are rarely associated with hepatitis C and antiviral treatment shows little or no effect. Thus, a multi-level approach is necessary in the clinical management of these patients. Increased rate of toxicity in the liver has been reported in association with chemotherapy in patients with aggressive lymphoma and hepatitis C (39). Thus, clearance of the virus if unable to induce remission of the lymphoma could, at least, decrease or pre-empt hepatotoxicity associated with the chemotherapy (40).

REFERENCES


