Editorial

When something goes wrong: B cell responses to hepatitis C virus

Vito Racanelli, Patrizia Leone

Department of Biomedical Sciences and Human Oncology, Section of Internal Medicine, University of Bari Medical School, Bari, Italy

Correspondence to: Vito Racanelli, MD. Department of Biomedical Sciences and Human Oncology, University of Bari Medical School, Policlinico 11, Piazza G. Cesare, 70124 Bari, Italy. Email: vito.racanelli1@uniba.it.

Comment on: Tucci FA, Kitanovski S, Johansson P, *et al.* Biased IGH VDJ gene repertoire and clonal expansions in B cells of chronically hepatitis C virus-infected individuals. Blood 2018;131:546-57.

Submitted Feb 27, 2018. Accepted for publication Mar 29, 2018.
doi: 10.21037/tcr.2018.04.05
View this article at: http://dx.doi.org/10.21037/tcr.2018.04.05

Understanding humoral responses to hepatitis C virus (HCV) has proven extremely difficult and B cells have generally been neglected because they do not seem to significantly influence the course and outcome of HCV infection. Despite almost all infected patients are positive for virus-specific antibodies, approximately 80% of these individuals develop chronic and often progressive disease, whose major long-term complications are cirrhosis, end-stage liver disease, and hepatocellular carcinoma (1,2). Mixed cryoglobulinemia (MC) and B cell non-Hodgkin lymphoma (B-NHL) may also occur and often dominate the clinical picture of chronic HCV infection (3). It is then assumed that B cells are basically inefficient in resolving HCV infection while they are responsible for its lymphoproliferative complications.

MC is a chronic immune complex mediated vasculitis with underlying B cell clonal proliferation that occurs in 10–15% of HCV-infected patients (3). MC immune complexes, which contain monoclonal IgM rheumatoid factor (RF), polyclonal IgG, and viral RNA, deposit in small-to-medium vessel walls causing an inflammatory reaction that can lead to skin lesions, peripheral neuropathy, and renal damage (3,4). IgM RF isolated from unrelated individuals typically display cross-reactive idiotypes, suggesting a high grade of conservation of their germline genes (4). Polyclonal IgG are directed to HCV antigens, binding predominantly core protein (3,4). B-NHL may develop in HCV-infected individuals with or without a history of MC (5) and may include three principal histologic types: lymphoplasmacytic, marginal zone, and diffuse large B cell lymphoma (6). The overall prevalence of HCV infection in patients with B-NHL is roughly 15%, higher than that reported in the general population and in patients with other hematologic malignancies (7). In our own experience, the rate of B-NHL was 1% in HCV-infected patients with and 6.2% in those without MC (P=0.003), after a 10-year follow-up (3). Successful antiviral therapy may not only prevent lymphoma development (8) but also result in its complete regression (5), thus strengthening the etiological link between HCV and lymphoproliferative disorders.

HCV-associated B cell proliferation most likely represents a continuum from the relatively benign clonal B cell expansion of MC to overt NHL. Clonal B cells are predominantly IgM RF-bearing cells with a stereotyped B cell receptor (BCR) commonly encoded by rearranged VH1-69 and Vκ3-20 variable region genes (9-14) in both MC and B-NHL, supporting the hypothesis that B cell clones are selected by a limited number of antigens. Unfortunately, these antigens have not been identified so far and the mechanisms by which HCV drives abnormal B cell expansion remain puzzling. A direct transforming role of the virus appears unlikely, considering that B cells are not direct targets for productive virus replication and that viral RNA sequences cannot be integrated in the host genome. Indeed, B cells do not express the full set of known factors that are essential for HCV entry into hepatocytes (15-17) and neither cell culture-produced genotype 2a HCV, nor pseudoparticles containing functional E1/E2 HCV envelope glycoprotein complexes of different genotypes, can infect primary B cells or B cell lines (17,18). Moreover, level of HCV RNA associated with lymphocytes from patients' blood samples is very small and far below one copy per cell (19-22), demonstrating that replication is inefficient in human lymphocytes. Activation of B cells via engagement of CD81

by HCV E2 protein has also been proposed (23), but it is in conflict with the observation that activated B cells in MC are not polyclonal (9-14) and that complete circulating lipoviral particles are not able to stimulate the activation described *in vitro* with high concentrations of recombinant E2 (16).

Tucci *et al.* (24) provide insight into the BCR gene repertoire and clonality of B cells in HCV-infected patients without MC. They found increased frequency of class-switched memory B cells and decreased frequency of transitional and naïve B cells in these patients compared to healthy subjects. By performing high throughput sequencing of Ig heavy chain (IGHV) VDJ rearrangements, the authors reveal a preferred usage of some IGHV genes, such as IGHV1-69 and IGHV4-59, in IgM^+CD27^+ non-class-switched memory B cells. Within this B cell compartment, they also found large expanded clones, many of whom displayed intra-clonal diversity. This characteristic is indeed an indicator of antibody diversification and affinity maturation occurred in germinal centers, where proliferating B cell clones undergo somatic hypermutation.

The findings add a new piece of information to B cell biology during HCV infection. They are reminiscent of those observed by our group in subjects who had spontaneously resolved an HCV infection (25,26). Few months after viral clearance, $CD27^+$ memory B cells from these subjects showed a preferential occurrence of specific VDJ elements, thus suggesting that B cell clones that were possibly implicated in the virus eradication were also those prone to aberrant proliferation. It is also interesting to note that in a small fraction of patients, clinical and immunological features of MC vasculitis persist in spite of direct-acting antiviral-induced HCV clearance. Lack of removal of RF from serum after HCV eradication has been found to be associated with a delay in the restoration of normal B cell subset representation, but the precise mechanisms underlying these virus-cleared vasculitides remains to be elucidated (27). Work is ongoing to test viral and self-antigens, along with host genetic factors, in experimental systems mimicking HCV-associated B cell activation.

Acknowledgments

Funding: None.

Footnote

Provenance and Peer Review: This article was commissioned and reviewed by the Section Editor Xuzhen Qin (Department of Laboratory Medicine, Peking Union Medical College Hospital, Beijing, China).

Conflicts of Interest: Both authors have completed the ICMJE uniform disclosure form (available at http://dx.doi.org/10.21037/tcr.2018.04.05). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved.

References

1. Hajarizadeh B, Grebely J, Dore GJ. Epidemiology and natural history of HCV infection. Nat Rev Gastroenterol Hepatol 2013;10:553-62.
2. Webster DP, Klenerman P, Dusheiko GM. Hepatitis C. Lancet 2015;385:1124-35.
3. Dammacco F, Racanelli V, Russi S, et al. The expanding spectrum of HCV-related cryoglobulinemic vasculitis: a narrative review. Clin Exp Med 2016;16:233-42.
4. Gorevic PD. Rheumatoid factor, complement, and mixed cryoglobulinemia. Clin Dev Immunol 2012;2012:439018.
5. Peveling-Oberhag J, Arcaini L, Hansmann ML, et al. Hepatitis C-associated B-cell non-Hodgkin lymphomas. Epidemiology, molecular signature and clinical management. J Hepatol 2013;59:169-77.
6. de Sanjose S, Benavente Y, Vajdic CM, et al. Hepatitis C and non-Hodgkin lymphoma among 4784 cases and 6269 controls from the International Lymphoma Epidemiology Consortium. Clin Gastroenterol Hepatol 2008;6:451-8.
7. Gisbert JP, Garcia-Buey L, Pajares JM, et al. Prevalence of hepatitis C virus infection in B-cell non-Hodgkin's lymphoma: systematic review and meta-analysis. Gastroenterology 2003;125:1723-32.

8. Kawamura Y, Ikeda K, Arase Y, et al. Viral elimination reduces incidence of malignant lymphoma in patients with hepatitis C. Am J Med 2007;120:1034-41.
9. Chan CH, Hadlock KG, Foung SK, et al. V(H)1-69 gene is preferentially used by hepatitis C virus-associated B cell lymphomas and by normal B cells responding to the E2 viral antigen. Blood 2001;97:1023-6.
10. Charles ED, Brunetti C, Marukian S, et al. Clonal B cells in patients with hepatitis C virus-associated mixed cryoglobulinemia contain an expanded anergic CD21low B-cell subset. Blood 2011;117:5425-37.
11. De Re V, De Vita S, Marzotto A, et al. Pre-malignant and malignant lymphoproliferations in an HCV-infected type II mixed cryoglobulinemic patient are sequential phases of an antigen-driven pathological process. Int J Cancer 2000;87:211-6.
12. Ivanovski M, Silvestri F, Pozzato G, et al. Somatic hypermutation, clonal diversity, and preferential expression of the VH 51p1/VL kv325 immunoglobulin gene combination in hepatitis C virus-associated immunocytomas. Blood 1998;91:2433-42.
13. Marasca R, Vaccari P, Luppi M, et al. Immunoglobulin gene mutations and frequent use of VH1-69 and VH4-34 segments in hepatitis C virus-positive and hepatitis C virus-negative nodal marginal zone B-cell lymphoma. Am J Pathol 2001;159:253-61.
14. Terrier B, Joly F, Vazquez T, et al. Expansion of functionally anergic CD21-/low marginal zone-like B cell clones in hepatitis C virus infection-related autoimmunity. J Immunol 2011;187:6550-63.
15. Dustin LB, Charles ED. Primary, post-primary and non-specific immunoglobulin M responses in HCV infection. Antivir Ther 2012;17:1449-52.
16. Lindenbach BD, Rice CM. The ins and outs of hepatitis C virus entry and assembly. Nat Rev Microbiol 2013;11:688-700.
17. Marukian S, Jones CT, Andrus L, et al. Cell culture-produced hepatitis C virus does not infect peripheral blood mononuclear cells. Hepatology 2008;48:1843-50.
18. McKeating JA, Zhang LQ, Logvinoff C, et al. Diverse hepatitis C virus glycoproteins mediate viral infection in a CD81-dependent manner. J Virol 2004;78:8496-505.
19. Mellor J, Haydon G, Blair C, et al. Low level or absent in vivo replication of hepatitis C virus and hepatitis G virus/GB virus C in peripheral blood mononuclear cells. J Gen Virol 1998;79:705-14.
20. Pham TN, King D, Macparland SA, et al. Hepatitis C virus replicates in the same immune cell subsets in chronic hepatitis C and occult infection. Gastroenterology 2008;134:812-22.
21. Veerapu NS, Raghuraman S, Liang TJ, et al. Sporadic reappearance of minute amounts of hepatitis C virus RNA after successful therapy stimulates cellular immune responses. Gastroenterology 2011;140:676-85.e1.
22. Zehender G, Meroni L, De Maddalena C, et al. Detection of hepatitis C virus RNA in CD19 peripheral blood mononuclear cells of chronically infected patients. J Infect Dis 1997;176:1209-14.
23. Rosa D, Saletti G, De Gregorio E, et al. Activation of naive B lymphocytes via CD81, a pathogenetic mechanism for hepatitis C virus-associated B lymphocyte disorders. Proc Natl Acad Sci U S A 2005;102:18544-9.
24. Tucci FA, Kitanovski S, Johansson P, et al. Biased IGH VDJ gene repertoire and clonal expansions in B cells of chronically hepatitis C virus-infected individuals. Blood 2018;131:546-57.
25. Racanelli V, Brunetti C, De Re V, et al. Antibody V(h) repertoire differences between resolving and chronically evolving hepatitis C virus infections. PLoS One 2011;6:e25606.
26. Racanelli V, Frassanito MA, Leone P, et al. Antibody production and in vitro behavior of CD27-defined B-cell subsets: persistent hepatitis C virus infection changes the rules. J Virol 2006;80:3923-34.
27. Reyes-Avilés E, Kostadinova L, Rusterholtz A, et al. Presence of rheumatoid factor during chronic HCV infection is associated with expansion of mature activated memory B-cells that are hypo-responsive to B-cell receptor stimulation and persist during the early stage of IFN free therapy. PLoS One 2015;10:e0144629.

Cite this article as: Racanelli V, Leone P. When something goes wrong: B cell responses to hepatitis C virus. Transl Cancer Res 2018;7(Suppl 5):S538-S540. doi: 10.21037/tcr.2018.04.05

