“Dangerous Liaisons”- An invited commentary

“ High Risk of Hepatocellular Carcinoma and Death in Patients with Immune-Tolerant Phase Chronic Hepatitis B”

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Long-term relationships are somehow unpredictable. Periods of harmony are often followed by times of conflict with outcomes often difficult to predict. This precept can apply to the relationship between HBV and human species. HBV acquired at birth or in early childhood establishes life-long persistent infection in the majority of subjects, which is evolving, and characterized by fluctuations of virological and clinical parameters. The overall impact of these fluctuations in the development of liver fibrosis and hepatocellularcarcinoma has often puzzled clinicians and researchers studying this complex and somewhat fascinating interaction between HBV and our species. An interesting new piece of information, related to this interaction, has now been added, thanks to the work of the group of GA Kim and YS Lim, published in this issue of Gut[1].

Let's try to first summarize the main points of this puzzle.

The early phase of this virus-host relationship is characterized by normal serum alanine aminotransferase (ALT) and high-titer viremia. This phase has been historically considered as "immune tolerant" and "disease-free". Based on this clinical categorisation; excluding both immunological and inter-individual virus spreading considerations; patients in this disease phase have been excluded from treatment recommendations. This early phase in natural history is then followed by a period of ALT perturbation and fluctuations of viremia (i.e., defined as immune active or HBeAg+/anti-HBe + hepatitis by new EASL nomenclature)[2], in which immunological and pathological events within the liver are considered more active and where treatment is recommended [2] .

This schematic interpretation has, however, been disputed in recent years initially by work pointing out that the initial phase of HBV disease is not immunologically inert [3].and that serum ALT values are a poor surrogate measurement of the strength of anti-HBV immunity [4].
More recently, the belief that events potentially leading to cumulative and measurable liver damage can only occur in the phase of active liver inflammation (i.e., with high serum ALT levels) has been challenged by evidence of high levels of HBV-DNA integration and clonal hepatocyte

expansion in patients considered in the IT phase of disease [5]. Since these modifications of the hepatocyte population can predispose to HCC development, the concept that IT patients are completely "disease-free" has been *de facto* dismissed.

In this work, GA Kim and YS Lim directly measured the risk of HCC development in a cohort 413 CHB patients considered immune tolerant and followed for more than 10 years[1]. The study not only shows that IT patients can develop HCC (~12% in 10 years), but also provides evidence that the incidence of HCC in this group of IT patients is higher than that detected in treated "immune active" patients (~ 6% in 10 years).
Several important conclusions can be drawn from these data. The IT phase of HBV infection cannot be considered fully benign. The clinical significance of the historical division of immune tolerant and immune active phases of HBV infection based mainly on serum ALT values is further challenged. Serum HBV-DNA levels more than ALT values are risk factors for HCC development [6] . Overall therapeutic reduction of HBV-DNA by nucleoside analogue (NA) therapy might therefore be indicated in all CHB patients, including whose in the IT phase, to reduce their risk of HCC development. It is worth noting that a recent study performed by another South Korean group, elegantly shows that treatment of IT patients has clinical benefits [7].

Nevertheless, a more detailed analysis of the large data set provided in this work poses additional questions, which emphasize the complexity of HBV-host interactions and the difficulty to predict in whom the pathological consequences of HBV infection will develop.

The first point to observe is that Kim and Lee defined the IT patients as those with HBV-DNA levels > 20,000 IU/Ml (~ > 10^5 copies x mL) and ALT values lower than 2 x ULN. Purists will argue, with some reason, that classical IT patients have much higher level of HBV viremia. However, in our opinion, the most important information is that deconvolution of IT patients in groups with high (>10^8), intermediate (10^7 -$10^{7.9}$) and low (10^4 -$10^{6.9}$) levels of HBV-DNA shows that the risk of HCC development is higher among the patients category

with lower HBV-DNA levels (Figure S3 of the paper). Another interesting observation is that patients with minimally elevated ALT values (> ULN but < 2 x ULN) are at higher risk of developing HCC as compared to subjects with genuinely normal serum ALT. These data would suggest that any reduction in HBV DNA level or minimal perturbation in serum ALT in the highly replicative eAg positive patient cohort are warning signs of disease progression and increased risk for HCC development.

One significant caveat of the study, however, is that we do not have information on HBsAg levels. It would be interesting to know whether the IT patients more likely to develop HCC are also those with higher HBsAg levels, a feature that would be in accordance with other studies showing that HBsAg level is a risk factor for HCC[8]. Moreover, high levels of HBsAg associated with intermediate levels of HBV-replication might be indicative of higher frequencies of HBV-DNA integration[9,10].

How can we reconcile all these information strands in a rational scenario of HCC development in chronic HBV infection?

One possible interpretation provided by the authors is that this category of patients (minimally altered ALT and HBV-DNA just above 20,000 IU/ml (10^4 - $10^{6.9}$ copies x mL)) represents the patients with higher cumulative immune damage that would predispose them more to HCC development. This is certainly a possibility since patients with this level of HBV-DNA have a residual HBV-specific T cell immunity[11]. Persistent activation of partially functional T cells that are unable to control HBV might just be good enough to sustain liver inflammatory events that could predispose to HCC development.

An additional possibility is that the overall uncontrolled but low level of HBV-DNA, detected in the IT patients at higher risk of HCC development, is a reflection of the patient age (i.e., advancing age being a risk factor for HCC development), but also reflects the quantity of HBV-DNA integration present in this HBV patient category[10].

These two different interpretations derive two different scenarios regarding the extremely important question raised by this work of when treatment of IT patients should start.

If HCC development is caused more from inflammatory events triggered by a crippled but still active immunity, we might speculate that NA treatment started only at the age of 30, that corresponds to the age from which HCC incidence starts to rise, might be sufficient to reduce HCC development since liver inflammatory events are suppressed by NA treatment[2]. However, if HCC development is mainly dependent on the quantity of precancerous events (e.g., HBV-DNA integration being a major one) that start to occur immediately after infection[5], then we could argue that treatment should be initiated as close as possible to the point of infection or as soon as persistent infection is diagnosed[12].

Of course, we accept that not all patients will eventually develop HCC and agree that universal treatment could be excessive. However, we should be capable of better identifying patients who are at risk of developing HCC. In this respect, we would like to comment on the fact that, in addition to the important question related to the therapeutic clinical management of patients with chronic HBV infection, this paper also clearly highlights the deficiency of the present methods to evaluate HBV infection and to better stratify patients based on immunologic, pathologic and also comprehensive virologic criteria. Surely HBV-DNA values and serum ALT levels alone are inadequate for any meaningful interpretation of this dynamic and complex persistent infection. Moreover, while HBsAg levels and HBeAg status are valuable additional parameters, they seem insufficient to differentiate patients at risk of pathological consequences. In this respect, better immunological and virological characterizations are needed. Quantity and function of HBV-specific immunity (T and B) are still too complex to be evaluated routinely and new more easy methods to quantity correctly such parameters should be developed. On the other hand, novel virologic entities have been recently described, including RNA-containing and genome-free enveloped capsid, which compose together with infectious particles the so-called HBcr antigen [13,14] . This composite antigen could play a role, as HBsAg and HBeAg, in HBV pathogenesis, and might represent a novel biomarker for patient categorization and/or for predicting therapeutic responses. In addition, circulating viral RNAs, which can be either contained in enveloped capsid, exosomes, or any other kind of host-derived serum particles, are also recently described entities that could be of interest to further stratify patients .[13,14]

In conclusion, understanding complex relationships requires a considerate approach to comprehend the multiple points of views.

Deconstructing the complex relationship between virus and host will be critical to improving outcomes and reducing the complications of life-long chronic infection. Kim and Lim's work shows clearly that the so called IT phase of HBV infection cannot be considered fully benign but it also exposes the inadequacy of current parameters for accurate disease assessment.

The need for new virological and immunological tools to better stratify disease and dictate the optimum timing of therapeutic intervention to prevent the sequelae of CHB infection seems obvious.

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