Two decades have passed since the discovery of hepatitis C virus (HCV) in 1989 [1]. Despite a massively expanded understanding of the virus and rapidly improving treatments, there is still no effective vaccine to prevent new HCV infections. A central question in HCV vaccine development is whether long-lasting, sustained protective immunity against HCV can be achieved. Multiple chimpanzee and human studies have provided valuable insights into this question. However, depending on the interpretation of these studies, the door to vaccine development has variably been perceived as open or closed.

Approximately one-third of persons will spontaneously clear a primary HCV infection, while others will have a persistent infection marked by ongoing viremia. This outcome is determined by a partially understood complex set of virus–host interactions. The available evidence indicates that the timing, magnitude, and breadth of adaptive immune responses are important [2–8]. CD4+ T cell responses appear to be critical to coordinate effective immunity, which probably explains the higher rate of HCV persistence in HIV-infected persons with CD4+ lymphocyte depletion.

Spontaneous recovery from HCV infection indicates that successful immunity is possible. However, reinfection has been demonstrated in chimpanzee and human studies. In chimpanzees, reinfection with both homologous and heterologous viruses has been documented [9,10]. Compared to primary infection, reinfection episodes have been associated with improved control of viral replication, an attenuated course of infection, and an increased likelihood of clearance [11–16]. Following both homologous and heterologous viral rechallenge, reductions in the duration and magnitude of viremia when compared to the initial infection have also been observed [14,15].

Chimpanzee studies have also provided unique insights into the nature of successful adaptive immunity. Rapid virological control upon reinfection is associated with HCV-specific T cell responses [13,15,16]. When CD4+ T cells were depleted in vivo prior to reinfection, persistent HCV infection ensued [17]. Similarly, CD8+ T cell depletion led to prolonged HCV viremia, which was only controlled once CD8+ T cells reappeared in the liver [16]. In this context, cross-genotype immunity has been documented [14], but persistence appears more likely in the setting of heterologous re-challenge [15]. Collectively, these data in chimpanzees suggest that immunity against HCV can be generated by initial infection.

In humans, HCV reinfection has also been documented in a variety of settings [18–23]. Similar to what has been observed in chimpanzees, levels of HCV viremia following reinfection are lower, generally transient, and shorter in duration than compared to the initial infection [24,25]. Participants followed through episodes of primary HCV infection, spontaneous clearance and subsequent reinfection have been described [25–27]. Aberle et al. reported two cases of reinfection following spontaneous clearance, both of whom also cleared reinfection [27]. In one of these cases, a strong antiviral Th1 response to NS3-4 was maintained at high levels [27].
a study by Osburn et al., HCV reinfection was observed in 11 of 22 (50%) injection drug users (IDUs) with spontaneous HCV clearance [25]. Subsequent clearance of reinfection was observed in nine of 11 (83%). Both the duration and peak HCV viremia in the setting of reinfection were decreased when compared to initial infection. Further, among reinfected participants, an increased breadth in HCV-specific T cell and broadened neutralizing antibody responses were observed. Data from these human studies are consistent with chimpanzee studies and suggest that protective HCV immunity, albeit not sterilizing, does exist in humans.

There are also a growing number of epidemiologic cohort studies which have sought to evaluate HCV reinfection rates and implications for protective immunity [28–31]. In this issue of the Journal, van de Laar et al. evaluated HCV reinfection and superinfection among participants with HCV seroconversion enrolled in a large, prospective cohort of drug users in Amsterdam, the Netherlands followed up at six monthly intervals during the period 1985–2005 [32]. By nature of their initial acquisition of HCV infection, the study participants represented persons at substantial risk for subsequent exposure to HCV. Using a standardized methodology for the virological testing of samples collected over the 20-year period, the investigators performed a detailed clinical and virological characterization of acute HCV infection and viremia that was detected after the acute phase, which they classified as superinfection when the virus detected in the blood changed (in someone with persistent primary infection) or as reinfection when viremia first appeared to be cleared before another new virus was detected in the blood.

Reinfection and superinfection were frequent among the 59 participants with HCV seroconversion, with 24 participants (39%) having multiple HCV infections during study follow-up. Among 24 participants with spontaneous HCV clearance of primary infection, HCV reinfection was subsequently observed in nine, while viremia remained negative over long-term follow-up in 14 participants, despite ongoing drug use in 50%. Reinfection could be confirmed in two of nine participants with HCV reinfection and the remaining seven cases were categorized as having probable reinfection, given that HCV RNA could not be sequenced from samples at either the initial (n = 6) or reinfection (n = 1) time points, likely due to low-level viremia at the time of sequencing. Following reinfection, viral persistence was observed in six of nine participants (two of which were co-infected with HIV infection). However, three of seven participants with HCV mono-infection demonstrated prolonged viral clearance, despite ongoing risk behaviors for HCV acquisition over long-term follow-up.

This important study by van de Laar and colleagues clearly demonstrates that HCV superinfection and HCV reinfection occur frequently among IDUs and that, with ongoing exposure, some who initially cleared develop persistent infection while others have prolonged viral clearance. This heterogeneity may be partially explained by early immune responses [2–8] and/or host genetic factors [33]. The observation that some individuals with the immunological capacity to clear initial infection develop viral persistence following reinfection provides further evidence that protective immunity against HCV is incomplete. Further, they observed a similar rate of reinfection among those with HCV clearance compared to the rate of infection in those previously uninfected. As the authors state in this paper, based on the incidence rate ratio in this study, it is tempting to imply that HCV protective immunity does not exist at all—even though the door to HCV vaccine development is closed. However, is that conclusion justified?

Neither HCV infection of chimpanzees or humans confers sterilizing immunity, defined as absolute protection against reinfection. However, the primary goal of vaccination is to prevent chronic hepatitis C, which causes cirrhosis and hepatocellular cancer. Thus, the question is whether immunity can be achieved to protect against viral persistence.

The human studies that have attempted to answer this question all have limitations that explain differences in their outcomes. Notably, most limitations underestimate the degree of protective immunity. For example, differences in subject characteristics including age, ethnicity, risk behaviors and HIV status, may affect observed rates of primary and reinfection clearance. Especially crucial, the actual number of HCV exposures is unknown and proxies such as drug use practices are incomplete at best. While chimpanzee studies count exposures, human studies only count incidents of viremia. An implication of this limitation is that long HCV RNA testing intervals underestimate exposure and protective immunity because reinfection events are overlooked, while viral persistence (by definition) is not (Fig. 1). In a study with a relatively broad testing interval, such as in the study by van de Laar et al., cases of reinfection and clearance would have been missed (Fig. 1, panel A) leading to an apparent predominance of viral persistence (Fig. 1, panel C) and the interpretation that there was a lack of protective immunity. Further, the outcome of reinfection (clearance vs. persistence) is often not adequately characterized because the person was not studied again after reinfection was first noted, again creating overestimation of viral persistence (and lack of protective immunity) bias.

There are also limitations in the classification of persons as “reinfected” or “superinfected”. Studies of persons infected by transfusion have shown that viremia can be cleared for months and then return without another exposure [34]. In an IDU, this pattern would be called reinfection. If the virus evaluated at the second point was genetically distinct from that detected at the
initial viremia, one might more confidently conclude there was reinfection. However, in careful studies of persons who received more than one HCV-contaminated blood transfusion the same pattern has been reported (donor A virus, an extended period with no detectable viremia, then donor B virus) [35].

So, which way is it? We believe the door to development of an HCV vaccine that protects against chronic infection remains open. Available data suggest that the goal is both possible yet difficult. Further studies are required to determine just how open that door is. Ideally, these studies should carefully capture primary infection and repeat events with frequent regular sampling and detailed risk assessments and detailed virologic and immunologic studies.

Perhaps more than any other lesson, the data from van de Laar et al. reveal the inordinately high risk of HCV infection among IDUs and the urgency of opening all available doors that might more effectively contribute to prevention of infection in this setting.

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References


