VIRAL HEPATITIS

A new HCV cell culture model for the next clinical challenges

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Refers to Saeed, M. *et al.* SEC14L2 enables pan-genotype HCV replication in cell culture. *Nature* **524**, 471–475 (2015)

Despite advances in hepatitis C treatment, substantial clinical hurdles remain to achieve universal cure and global control of infection. Saeed *et al.* identified SEC14L2 as a host factor permitting replication of clinical HCV isolates in cell culture, providing a novel system to model infection of patient-derived viruses.

Worldwide ~160 million individuals are chronically infected with HCV, which increases their risk of progressive liver disease, including cirrhosis and hepatocellular carcinoma.[1] Although direct-acting antiviral agents (DAAs) have markedly improved the standard of care for chronic HCV infection, important clinical challenges remain, including treatment failure in some patient groups, limited access to therapy and the development of a protective vaccine. Overcoming these hurdles will require robust model systems enabling infection by viral isolates from patients. Recently, Saeed *et al.*[2] identified SEC14L2—SEC14-like protein 2, also known as tocopherol-associated protein 1, a lipid-binding protein ubiquitously expressed in human tissues, but not detected in hepatoma cell lines—as a novel host factor for HCV infection. Remarkably, SEC14L2 expression enabled replication of several different clinical HCV isolates in cell culture, thus providing the first cell culture model for the study of non-cell-culture-adapted HCV strains.

Such a cell culture model is critical to tackle the remaining clinical challenges for HCV infection. Not all patient groups respond to currently available therapies and the emergence of viral variants resistant to DAAs leads to treatment failure in some patients.[3] The very high costs of DAAs preclude access to treatment in the large majority of HCV-infected patients. For example, the recently FDA-approved sofosbuvir costs up to US$84,000 per person for a 12-week treatment course in the USA. Thus, access to DAA-based treatment is limited even in high-resource countries. Furthermore, viral cure does not eliminate the risk of development of hepatocellular carcinoma in patients with fibrosis.[4] Finally, an HCV vaccine to eradicate viral infection and prevent virus-induced liver disease and cancer on a global level remains an urgent unmet medical need.[5] In the absence of robust cell culture systems enabling the study of diverse clinical isolates, the sequence heterogeneity of HCV and its ability to evade host immune responses have so far precluded the development of a broadly protective vaccine.

Current model systems have contributed greatly to our understanding of HCV–host interactions and the development of antiviral therapies, but are limited in their ability to address many of the remaining clinical challenges. So far, the model systems allowing robust study of the complete viral life cycle are based largely on a unique cell-culture-adapted strain of HCV JFH-1, which does not reflect the variability and diversity of HCV isolates from patients. Most attempts to infect cultured Huh7-derived human hepatoma cells with clinical isolates have failed, although patient sera are infectious to chimpanzees or human liver-chimeric mice. Studies in cell culture with patient-derived viruses, which would yield considerable insight into disease pathogenesis and mechanisms of treatment failure, have not been possible until now.

The discovery of SEC14L2 by Saeed *et al.*[2] helps to address this challenge. SEC14L2 is a novel host factor permitting replication of nonadapted HCV in cell culture. Most HCV strains require cell-culture-adaptive mutations to replicate, but the factors that limit replication of nonadapted strains are not clearly defined. By using a gain-of-function approach, Saeed *et al.*[2] found that SEC14L2 enabled replication of all replicons in cell lines. Mechanistic studies demonstrated that SEC14L2, a lipid-binding protein, protects against lipid peroxidation through vitamin E (tocopherol)-dependent mechanisms (Figure 1). Given that lipid peroxidation has been shown to severely restrict HCV replication in Huh-7 cells,[6] it is probable that SEC14L2-mediated inhibition of lipid peroxidation creates a more favourable cellular environment for HCV infection. Strikingly, SEC14L2 enabled infection of cultured cells by several clinical HCV isolates (including genotypes 1a, 1b and 3a from infected sera) (Figure 1). The discovery of SEC14L2 as a host factor is a critical advance, enabling the study of patient-derived HCV infection in cell culture for the first time.

A possible limitation of the model is the use of transformed Huh7.5 hepatoma cells, which might not reflect all features of primary human hepatocytes. Moreover, the mechanism of action of SEC14L2 is still

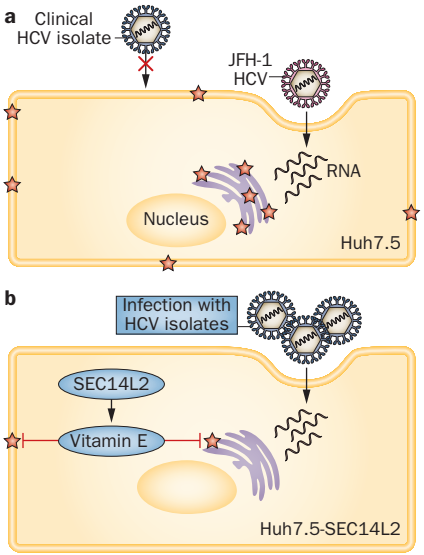

Figure 1 | SEC14L2 enables infection of cultured cells by clinical HCV isolates. SEC14L2 seems to facilitate infection by protecting against lipid peroxidation to create a more favourable cellular environment, enabling robust HCV replication. Whereas HCV JFH1-based recombinant viruses can infect liver cell lines in the absence of SEC14L2 (**a**), robust infection of cells with clinical isolates requires expression of SEC14L2 (**b**). This finding paves the way to study infection by patient-derived HCV isolates in cell culture. Peroxidized lipids are represented as stars on the figure.

only partially understood. Further mechanistic insights could improve the robustness of the SEC14L2 cell culture model, and might also enable improved understanding of viral pathogenesis and novel therapeutic approaches.

What are the implications of this new model system for future clinical challenges and research? In addition to enabling the study of nonadapted clinical variants in cell culture, the SEC14L2 model opens the possibility of resistance-testing of patient-derived HCV isolates for antivirals in preclinical stages of drug development. A better understanding of the underlying mechanisms of drug resistance in the context of patient viral isolates will help to understand resistance to licenced DAAs and could be incorporated into future drug development to limit emergence of resistance. Additionally, some patient groups fail to respond to DAA therapy even in the absence of resistance.[3] HCV isolates from these patients could be used in the SEC14L2 system to model and understand treatment failure in cell culture. An important example for this challenge is infection with HCV genotype 3, as DAAs are less effective against this genotype, particularly in patients with advanced liver disease.[7]

Infection of SEC14L2-expressing cells with clinical HCV isolates will also be highly useful in the context of vaccine development. The ability to identify and characterize neutralizing antibodies and immune responses against the challenge of highly diverse patient-derived viruses in cell culture will guide vaccine design and accelerate preclinical and clinical development. Furthermore, by modelling immune responses against clinical isolates in cell culture, the system could also contribute to our still limited understanding of immune-mediated protection and clearance of viral infection, another hurdle for efficient vaccine design.[8]

Finally, the SEC14L2 cell culture model might also open avenues to investigate viral pathogenesis using patient viruses associated with specific disease outcomes. Such studies could contribute greatly to our overall understanding of HCV biology and disease pathogenesis. For example, clinical and epidemiological evidence is accumulating that HCV genotype 3 seems to have a distinct pathology that is associated with steatosis and more rapid progression of liver fibrosis to cirrhosis and hepatocellular carcinoma.[9,10] Currently, robust model systems needed to conduct these studies are still in development and pathogenic mechanisms of liver disease associated with HCV genotype 3 are still poorly understood. In this context, the discovery that SEC14L2 allows patient-derived genotype 3 infection of cultured cells might help to uncover mechanisms of disease pathogenesis and identify novel therapeutic targets not only for cure of infection, but also for prevention and treatment of advanced liver disease.

In summary, the identification of SEC14L2 provides a long-awaited and much-needed model system to study infection, virus–host interactions and pathogenesis of clinical HCV isolates. Thus, this new discovery could contribute to overcome the remaining hurdles for universal cure and prevention of chronic hepatitis C and liver disease.

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Competing interests
The authors declare no competing interests.

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