In Vitro Model for Hepatitis C Virion Production

Description of Technology:

This invention provides an *in vitro* hepatitis C virus (HCV) replication system that is capable of producing viral particles in a culture medium. Hepatitis C is a major public health problem, the development of therapeutics for which has been hampered by a lack of a robust model system to study the complete viral life cycle. This invention provides a new model system for the complete replication cycle of hepatitis C virus and virion production, assembly and release. The model is useful for screening antiviral agents against HCV.

A full length HCV construct, CG1b of genotype 1b which is known to be infectious, was placed between two ribozymes designed to generate the exact 5' and 3' ends of HCV when cleaved. Using this system, HCV proteins and positive and negative RNA strands have been shown to reproduce intracellularly, and viral particles that resemble authentic HCV virions are produced and secreted into the culture medium.

The patent application includes claims directed toward the following:

- a construct comprising specific nucleic acid sequences including HCV genotype 1b, genotype 1a, genotype 2a or potentially other genotypes
- a method for identifying a cell line that is permissive for infection with HCV
- a method for propagating HCV in vitro
- a method for screening agents capable of modulating HCV replication or activity
- a method for testing the level of HCV replication or activity
- an HCV vaccine comprising HCV virus particles

Applications:

The model offers a novel method for investigating the entire HCV life cycle including replication and pathogenesis and is useful for high-throughput antiviral screening. This technique may also be useful for making infectious particles that are useful in the production of HCV vaccines.

Advantages:

This system provides a new, stable and efficient cell culture model to further study the life cycle and biology of HCV, and to test potential therapeutic targets for hepatitis C.

Market:

Hepatitis C virus (HCV) chronically infects approximately 200 million people worldwide and increases the risk of developing cirrhosis and hepatocellular carcinoma. This technology would be useful for studying the HCV life cycle, screening for therapeutic agents against multiple HCV strains, including Genotype 1a, 1b and 2a, and the development of HCV vaccines. HCV genotypes 1 and 2 are the major genotypes with worldwide distribution; they are known to be associated with different clinical profiles and therapeutic responses. Hence, the model may be used to screen for varying levels of effectiveness of therapeutics against the major HCV genotypes.

Development Status:

This technology is available for use in diagnostics, drug/vaccine discovery, production and development. Current work is directed toward studies into the HCV life cycle and replication and the pathogenesis of HCV screening for antiviral agents against multiple HCV strains. This model has been used to generate in cell culture HCV strains infectious for chimpanzees, the only experimental animal susceptible to infection with the hepatitis C virus, a critical step in the development of new vaccines for Hepatitis C. Future work may be directed toward the use of this system for development of vaccine candidates against HCV.

Inventors:

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Publications:

Hu Z, Zhang Z, Kim JW, Huang Y, Liang TJ Altered proteolysis and global gene expression in hepatitis B virus X transgenic mouse liver. *J Virol* (80): 1405-13, 2006. Pubmed References]

Heller T, Saito S, Auerbach J, Williams T, Moreen TR, Jazwinski A, Cruz B, Jeurkar N, Sapp R, Luo G, Liang TJ An in vitro model of hepatitis C virion production. *Proc Natl Acad Sci U S A* (102): 2579-83, 2005. [Pubmed References]

Patent Status:

DHHS Reference No. E-324-2004, PCT Application Serial No. PCT/US2005/035487

Licensing Status:

Available for exclusive or non-exclusive licensing

Licensing Contact:

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Collaborative Research Opportunity:

The National Institute of Diabetes and Digestive and Kidney Diseases, Liver Diseases Branch is seeking parties interested in collaborative research directed toward molecular strategies for vaccine and antiviral development, and animal models of viral hepatitis C. Please contact Dr. T. Jake Liang at 301-496-1721, jliang@nih.gov or Rochelle S. Blaustein at Rochelle.Blaustein@nih.gov for more information.

Research Focus and Selected Publications for Principal Investigator

Liver Diseases Branch

Hepatitis B (HBV) and C (HCV) viruses are the leading causes of chronic liver diseases in the world. Chronic infection with these viruses has been linked to the development of hepatocellular carcinoma, a major leading cause of death from cancer worldwide. The overall goals of the laboratory are to understand the basic mechanisms of hepatocellular injury, recovery from hepatitis viral infection, persistent infection, progression of disease, and development of hepatocellular carcinoma. Other research efforts are directed at molecular strategies for vaccine and antiviral development, and animal models of viral hepatitis.

1. Baumert TF, Yang C, Schurmann P, Kock J, Ziegler C, Grullich C, Nassal M, Liang TJ, Blum HE, von Weizsacker F Hepatitis B virus mutations associated with fulminant hepatitis induce apoptosis in primary Tupaia hepatocytes. *Hepatology* (41): 247-56, 2005. [Pubmed References]

2. Wakita T, Pietschmann T, Kato T, Date T, Miyamoto M, Zhao Z, Murthy K, Habermann A, Krausslich HG, Mizokami M, Bartenschlager R, Liang TJ Production of infectious hepatitis C virus in tissue culture from a cloned viral genome. *Nat Med* (11): 791-6, 2005. [Pubmed References]

3. Cai Z, Zhang C, Chang KS, Jiang J, Ahn BC, Wakita T, Liang TJ, Luo G Robust production of infectious hepatitis C virus (HCV) from stably HCV cDNA-transfected human hepatoma cells. *J Virol* (79): 13963-73, 2005. [Pubmed References]

4. Huang Y, Yang H, Borg B, Su X, Rhodes SL, Yang K, Tong X, Tang G, Howell CD, Rosen HR, Thio CL, Thomas DL, Alter H, Sapp RK, Liang TJ. A novel functional single nucleotide polymorphism of interferon-γ gene is important for interferon-α-induced and spontaneous recovery from HCV infection. Proc Natl Acad Sci USA 2007; 104: 985-990. [Pubmed References]

5. Huang Y, Feld JJ, Sapp RK, Nanda S, Lin J-H, Blatt LM, Fried MW, Murthy K, Liang TJ. Defective hepatic response to interferon and activation of suppressor of cytokine signaling 3 in chronic hepatitis C. Gastroenterology 2007; 132: 554-563. [Pubmed References]

6. Kato T, Matsumura, Heller T, Saito S, Sapp RK, Murthy K, Wakita T, Liang TJ. Production of infectious hepatitis C virus of various genotypes in cell culture. J Virol 2007; 81:4405-4411. [PubMed Abs]

7. Cai Z et al. Robust production of infectious hepatitis C virus (HCV) from stably HCV cDNA-transfected human hepatoma cells. J Virol. 2005 Nov;79(22):13963-13973. [PubMed abs]

8. T Wakita et al. Production of infectious hepatitis C virus in tissue culture from a cloned viral genome. Nat Med. 2005 Jul;11(7):791-796. Erratum in Nat Med. 2005 Aug;11(8):905. [PubMed abs]

9. T Heller et al. An in vitro model of hepatitis C virion production. Proc Natl Acad Sci USA. 2005 Feb 15;102(7):2579-2583. [PubMed abs]