Hepatitis C Virus and Cytokine Responses

Eui-Cheol Shin, M.D., Ph.D.
Laboratory of Immunology & Infectious Diseases (LIID), Graduate School of Medical Science & Engineering (GSMSE), KAIST Daejeon, Korea
ecshin@kaist.ac.kr
Hepatitis C Virus (HCV) Infection

1. Worldwide, 170 million people are infected.

2. After acute infection, 60-80% of patients develop persistent infection.

3. Chronic persistent infection progresses to liver cirrhosis and hepatocellular carcinoma.

4. HCV modulates cytokine responses of host cells.
The HCV cell culture (HCVcc) system

pJFH-1 plasmid

Transfection

Transcription

JFH-1 RNA

Huh-7.5 cell

Membranous web

Nucleus

ER

Translation

Membrane-assoc. RNA replication

Maturation

Release

JFH-1-transfected Huh-7.5 cells

Supernatant collection at peak HCV release

Infection of Huh7.5 cells with HCV supernatant

JFH-1-infected Huh-7.5 cells

Supernatant collection at peak HCV release

HCVcc Stock
1. TNF response

2. IFN response
HCV Infection Sensitizes Cells to TNF-α-Induced Cell Death

HCV Infection Suppresses TNF-\(\alpha\)-Induced NF-\(\kappa\)B Activation

**A**

- **Graph:**
  - Cell death (% compared to control)
  - Conditions: No inhibitor, SP600125, SN50, SP600125+SN50
  - Comparison: Huh-7 vs. Huh-7.5
  - Statistical significance: *P < 0.05*, **P < 0.001**

**B**

- **Table:**
  - HCV infection (Huh-7.5)
  - Time points: 0, 2, 5, 10, 30, 60 (min)
  - Proteins: P-IKK-\(\alpha/\beta\), T-IKK-\(\beta\), P-I\(\kappa\)B-\(\alpha\), P-JNK, T-JNK, GAPDH

**C**

- **Western Blot:**
  - Conditions: HCV, TNF-\(\alpha\)
  - **Supershift**:
    - NF-\(\kappa\)B

---

SP600125: JNK inhibitor
SN50: NF-\(\kappa\)B inhibitor

HCV Infection Suppresses TNF-α-Induced Expression of Anti-Apoptotic Proteins

HCV Core, NS4B, and NS5B Inhibit TNF-α-Induced NF-κB Activation

HCV infection

Core, NS4B, and NS5B

TNF-α

NF-κB

Anti-apoptosis (by Bcl-xL, XIAP, and c-FLIP_L)

Cell death of hepatocyte

Liver injury

JNK
1. TNF response

2. IFN response
CD8+ T cells and MHC class I

- CD8+ T cells often fail to eradicate HCV infection.
- CD8+ T cells recognize virus-infected cells by detecting viral peptides presented on MHC class I molecules.
- Many viruses evade CD8+ T cell responses by downregulating MHC class I expression on virus-infected cells.
HCV infection attenuates IFN-induced MHC class I upregulation

Kang W et al. *Gastroenterology* 2014, 146:1351
HCV infection attenuates IFN-induced MHC class I upregulation

Kang W et al. Gastroenterology 2014, 146:1351
HCV protein expression does not lead to attenuation of IFN-induced MHC-I upregulation

Kang W et al. *Gastroenterology* 2014, 146:1351
HCV infection reduces synthesis rate of MHC class I protein

Protein synthesis rate

Kang W et al. *Gastroenterology* 2014, 146:1351
HCV infection phosphorylates PKR and eIF2α

Garaigorta U & Chisari FV. *Cell Host & Microbe* 6:513, 2009

HCV blocks IFN-α/β-induced ISG production by inducing PKR phosphorylation.

Kang W et al. *Gastroenterology* 2014, 146:1351
PKR silencing restores IFN-induced MHC class I expression.

Kang W et al. *Gastroenterology* 2014, 146:1351
• HCV infection attenuates IFN-induced MHC class I expression at translational level through activation of the PKR-eIF2α pathway; not by blocking IFN signaling or mRNA transcription.

• This leads to a reduction of the antiviral effector functions of HCV-specific CD8+ T cells.

• Herein, we suggest a novel mechanism by which HCV circumvents antiviral adaptive immune responses.
Antigen-Processing & Presentation for CD8 T Cells

Induction of Immunoproteasome by IFNs

Constitutive proteasome

Viral infection

↓

T cells / NK cells

↓

IFN-γ

Type I IFN

Immunoproteasome

LMP2

LMP7

MECL1

1. Immunoproteasome generates peptides more efficiently.
2. Immunoproteasome generates peptides which cannot be generated by constitutive proteasome.
   → immunoproteasome-dependent peptides
HCV infection attenuates IFN-stimulated immunoproteasome induction

Oh IS et al. FP-22 (6/12, 16:30)
Acknowledgments

Laboratory of Immunology & Infectious Diseases (LIID), Graduate School of Medical Science & Engineering (GSMSE), KAIST Daejeon, Korea
Wonseok Kang, Pil Soo Sung, In Soo Oh, Dong-Yeop Chang

Department of Microbiology, Ajou University School of Medicine, Suwon, Korea
Sarah Yoon, Yong-Joon Chwae

Liver Diseases Branch, NIDDK, National Institutes of Health, Bethesda, MD, USA
Su-Hyung Park, Barbara Rehermann

Department of Internal Medicine, Yonsei University School of Medicine, Seoul, Korea
Seungtaek Kim, Ja Kyung Kim, Kwang Hyub Han

Department of Bio & Brain Engineering, KAIST, Daejeon, Korea
Junseong Park, Seung-Wook Ryu, Kyungsun Choi, Chulhee Choi

Department of Pathology, Pusan National University Hospital, Busan, Korea
Do Youn Park

Department of Physiology, Ewha Womans University School of Medicine, Seoul, Korea
Youn-Hee Choi
IFN Signaling
ISGF3 components are overexpressed without phosphorylation in HCV-infected livers

Sung PS et al. FP-61 (6/13,16:30)