Role of HIV-1 Nef in Acceleration of HCV-Mediated Liver Disease

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HIV-1/HCV co-infection

* Shared routes of infection:
  - sexual contact
  - blood stream
  - IDU

* Common with ~ 30% of all HIV-1-infected persons
Co-infection has profound, adverse consequences.

- elevate HCV viral load.
- expedite HCV-mediated liver disease progression.

- two-fold acceleration of fibrosis
- five-fold higher risk of cirrhosis-related liver complications, etc.

• Cirrhosis and end-stage liver disease – 50% of all deaths in co-infected patients - leading cause of morbidity and mortality in Western countries.
Genomic and virion structure of HIV-1

Structural: Gag, Pol, and Env
Regulatory:
  Early: Tat, Rev, and Nef
  Late: Vpr, Vpu, Vif
Distinct target cells for infection

1. Receptor/co-receptors
   A. HIV-1
      CD4 \[\rightarrow\] CCR5 \[\rightarrow\] T helper cells
      \[\rightarrow\] Monocytes/microphages
      \[\rightarrow\] Dendritic cells, etc
      CD4 \[\rightarrow\] CXCR4

   B. HCV
      LDLR
      CD81
      SR-B1
      Claudin-1
      Occludin \[\rightarrow\] Hepatocytes

2. Fundamentally different life cycles
Possible mechanisms

1. Direct infection of HIV-1 into HCV-infected hepatocytes/HSC

2. Indirect effect
   A. Viral proteins, such as Env, Tat, and Nef
   B. Dysfunction immune systems by HIV-1 and/or viral proteins
Replication of HIV-1 in human hepatocytes

A. 

B. 

C. 

RT (10\(^{-4}\) cpm/ml)

Days Infection

Mock

Jurkat

Huh7.5.1

FLuc (10\(^{-4}\))

Mock

VSV-G

HXBc2

89.6

RT (10\(^{-4}\) cpm/ml)

Mock

HXBc2

89.6

YU2
Viral protein candidates

1. Env - interact with CXCR4 or CCR5 co-receptor
   → enhance HCV replication in the replicon
   → induce apoptosis

2. Tat - diffusable protein
   → enhance hepatocarcinogenesis in transgenic mice
Relevant Nef functions for up-regulation of HCV replication

1. Induces formation of conduits (filopodia) and secretion of exosomes.

2. Regulates the amount of intracellular lipids by modulating expression of lipid molecules.

3. Forms complexes with and thereby activates several cellular kinases, such as the Src family of tyrosine kinases.

4. Alters host immune responses.
Exosome-mediated Nef transfer?

A.

Cells → Pellets

Sup (C) → Filter (0.22μ) (F) → Sup (100g 70 min) (S*) → Suc 20%

Pellets (S)

B.

AChEase activity

Mock F S S* F S S*

CMsp HIVsp

Virol. J. 2011
Transfer of Nef protein from Jurkat T cells into hepatocytes.
Nef is transferred from HIV-1-infected cells to hepatocytes

Huh7.5.1

\(\triangle\text{Nef-HIV-1} \quad \text{Nef} \quad \text{wt-HIV-1} \quad \text{Nef} \)

\(\text{SP-DiIC} \quad \text{Merged} \quad \text{SP-DiIC} \quad \text{Merged} \)
Biological significance of Nef transfer

1. Up-regulation of HCV replication
2. Generation of ROS
3. Effect on alcohol-mediated up-regulation of HCV replication
4. Others
Nef up-regulates HCV subgenomic replicon expression

A. 

B. 

C. 

Huh7.5.1

RLuc

Nef.GFP

ER

Merged

Nef.GFP

ER

Merged
Nef-mediated induction of ROS

A. 

Huh7.5.1

Mock  |  Nef

RLuc

Mock  |  Nef

B. 

![Graph of ROS vs Nef concentration](image)

- ROS (%) vs Nef (μg)
  - Huh7.5.1
  - RLuc

**Notes:**

- *p < 0.05
- **p < 0.01
Effect of Nef on ethanol-mediated up-regulation of HCV replication

- GFP
- Nef.GFP

RLuc (10^-3)

EtOH [mM]

0 12.5 25 50
Summary

HIV-1 → Nef → p53
Hepatocytes

EtOH → LD → HCV

HCC

CD4+ cells

MAPK
NFκB
TGFβ1
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