

## **Online Supplementary Material**

This supplementary material has been provided by the authors to give readers additional information regarding their study.

Supplement to: Yadong Wang et al. IP-10 Interfere with the Antiviral Response of Direct-Acting Antiviral Agents for Hepatitis C Virus Infection

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# **IP-10 Interfere with the Antiviral Response of Direct-Acting Antiviral Agents for Hepatitis C Virus Infection**

## **Online Supplemental Materials**

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## Supplementary 1

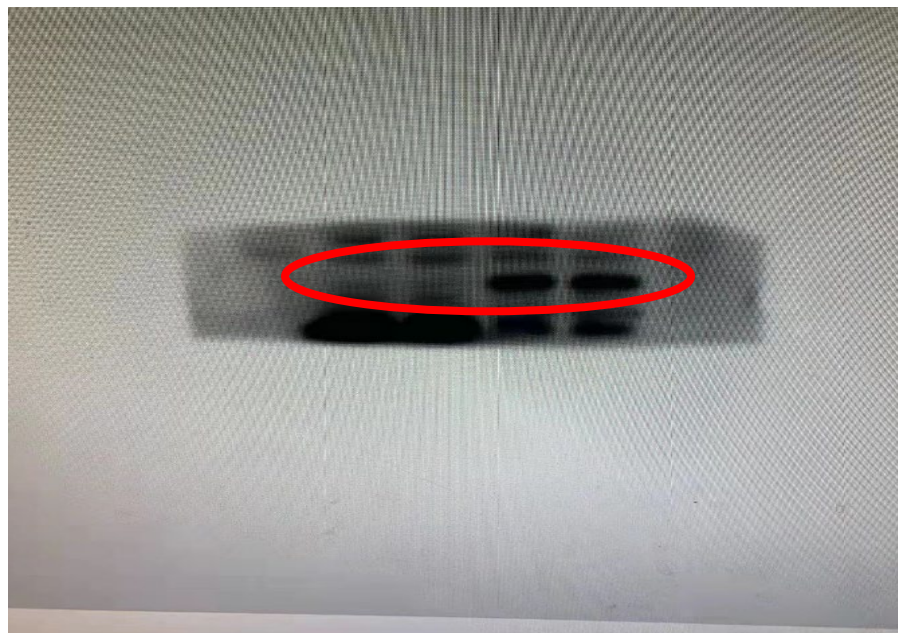
**Table S1. Genes detected in hepatocytes of HCV infected patients**

Gene	Location	Function
IP-10/CXCL10	4q21	Chemotactic for monocytes and T-lymphocytes. Binds to CXCR3
CXCR3	Xq13	A member of the receptor family of Th1 cells. The common receptor for the CXCL9, CXCL10, and CXCL11.

The genes detected in hepatocytes of HCV infected patients with PR or DAAs treatment and in Huh7 cells transfected with HCV replicons (named pHCV-rep-NeoR-hRluc) based on PCR Array Analysis.

## Supplementary 2

**Figure S1. The first experiment of HCV-core protein expression bands**



Huh7 cells were transfected with HCV core plasmids and inhibited by NS5A inhibitors. 48h after transfection, cells were lysed and analyzed by immunoblotting using the indicated antibodies. The Immunoblotting bands of HCV core protein expression in Huh7 cell detected by WB. In the red circle, The right two bands from the cells of HCV core plasmids transfection, the left two bands from the cells of HCV core transfection add NS5A inhibitors. The HCV-core protein expression bands were highlighted with the red circle in the figure.

### Supplementary 3

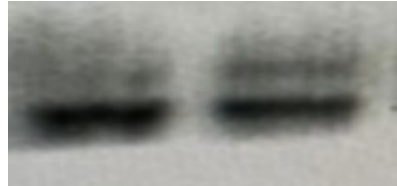
**Figure S2. The second experiment of HCV-core protein expression bands in Huh7 cells**



Huh7 cells were transfected with HCV core plasmids and inhibited by NS5A inhibitors. 48h after transfection, cells were lysed and analyzed by immunoblotting using the indicated antibodies. The Immunoblotting bands of HCV core protein expression in Huh7 cell detected by WB. In the red circle, the left two bands from the cells of HCV core plasmids transfection, the right two bands from the cells of HCV core transfection add NS5A inhibitors. The HCV-core protein expression bands were indicated with the red circle in the figure.

## Supplementary 4

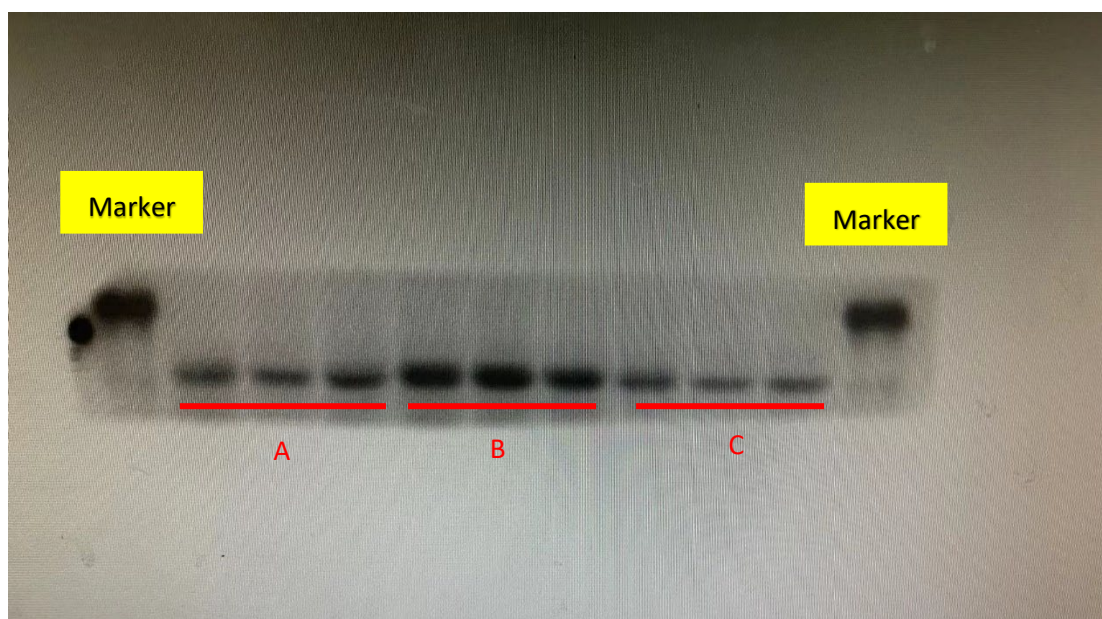
**Figure S3. The enhanced image of Figure S2**



The enhanced image was a cropped picture from Figure S2. The left immunoblotting band is the HCV-core protein expression in cells of HCV core plasmids transfection; the right immunoblotting band is the HCV-core protein expression in cells of HCV core plasmids transfection adds NS5A inhibitors.

## Supplementary 5

Figure S4. IP-10 protein expression bands



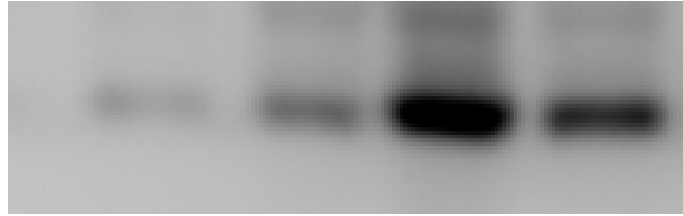
The Wild-type and HCV replicating Huh7 cells were treated with NS5A inhibitors for 48h, hepatocytes were collected followed by the analysis of IP-10 protein expression. The immunoblotting bands of IP-10 protein expression in Huh7 cells were detected by WB. IP-10 protein expression bands were indicated with markers on each side. The left three bands from the wild-type Huh7 cells added NS5A inhibitors (**A**), the middle three bands from the Huh7 cells of HCV core transfection (**B**), and the right three bands from the cells of HCV core transfection add NS5A inhibitors (**C**).



## Supplementary 6

Figure S5. IP-10 protein expression bands

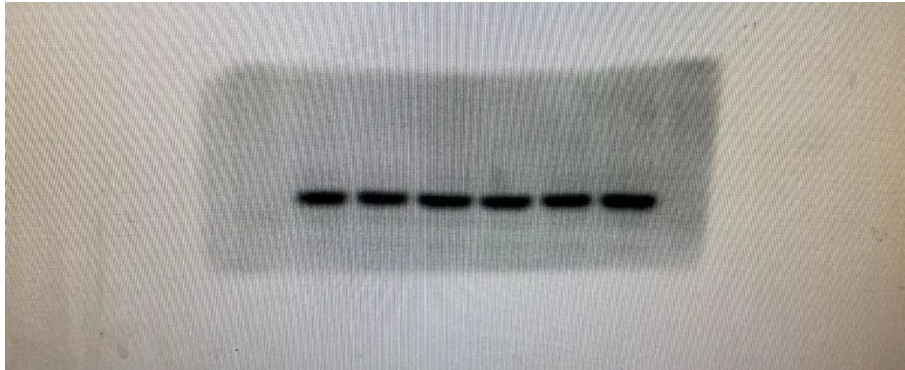
HCV-core	-	-	+	+
NS5A inhibitors	-	+	-	+



The above image was the cropped picture of IP-10 protein expression bands.

## Supplementary 7

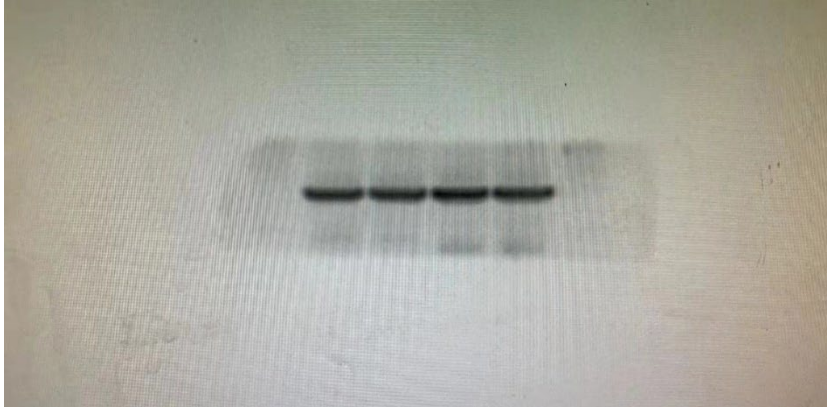
**Figure S6. GAPDH for HCV core and IP-10 protein expression**



The left three bands represented the GAPDH for HCV core, and the right three bands represented for IP-10.

## Supplementary 8

**Figure S7. GAPDH bands**



The above image was the cropped picture of GAPDH bands from figure S6.

## Supplementary 9

**Figure S8 HE staining for liver tissue section**

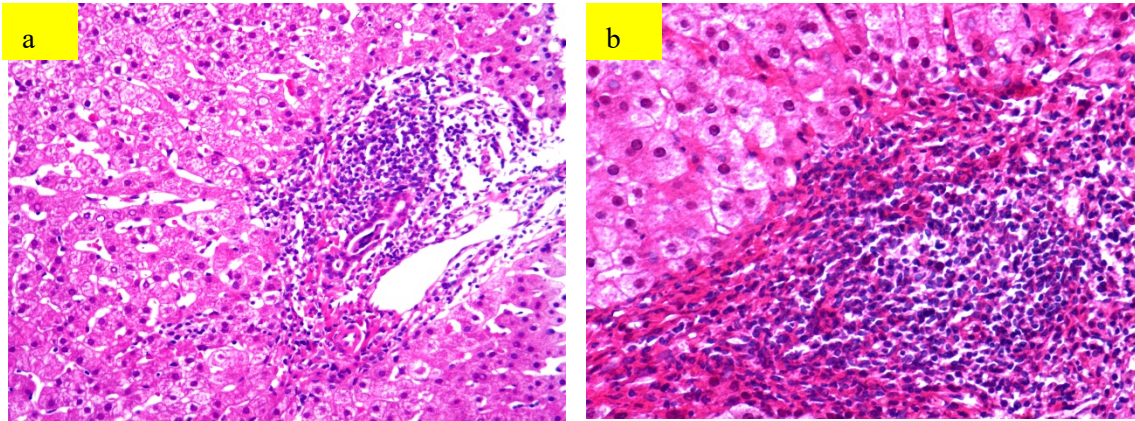


Fig a (100X) & b (200X). The HE staining for paraffin embedded sections of liver tissue. It was used to observe the characteristics of liver inflammation and evaluate HAI score in patients with CHC.

## Supplementary 10

**Figure S9 Intrahepatic IP-10 expression stained by IHC**

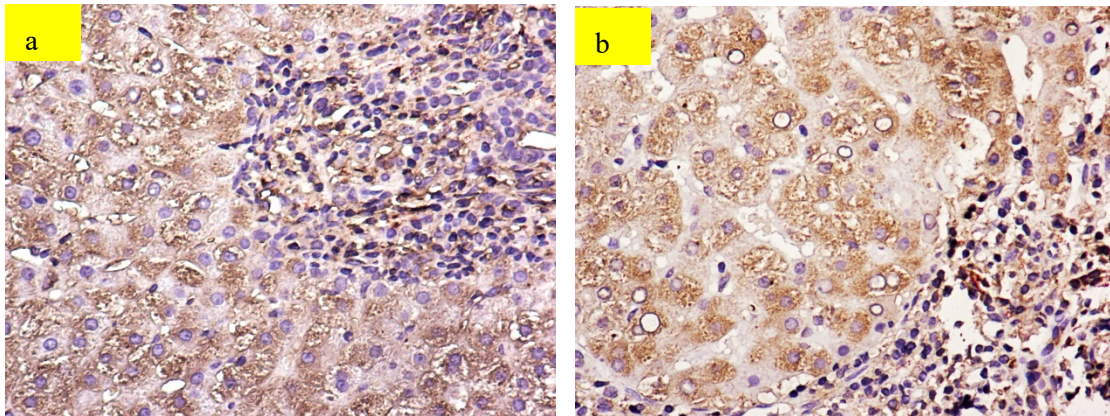


Fig a & b (200X). The expression of IP-10 in liver tissue of CHC patient stained by immunohistochemical staining. As shown in pathological figures, IP-10 is mainly expressed in hepatocytes and in a small number of inflammatory cells in the portal area

## Supplementary 11

**Figure S10** Intrahepatic CXCR3 expression stained by IHC

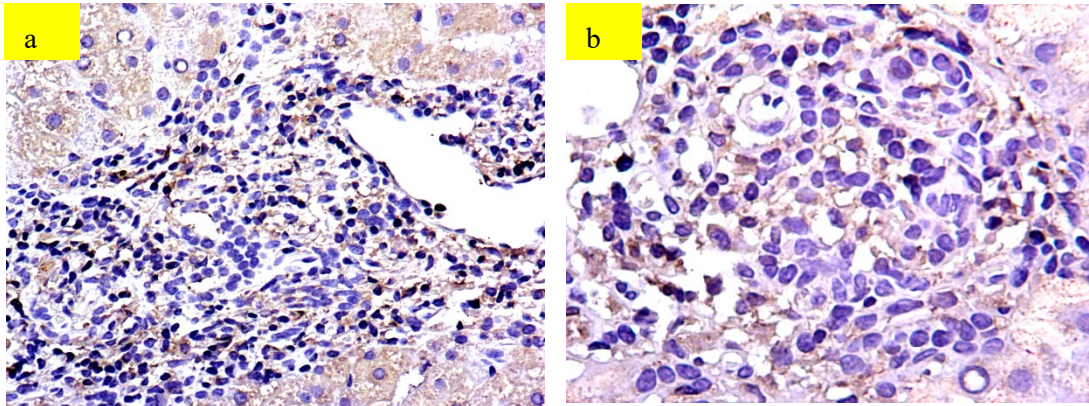


Fig a (200X) & b (400X). The expression of CXCR3 in liver tissue of CHC patient stained by immunohistochemical staining. CXCR3 is mainly expressed in lymphocytes in the portal area and inflammatory necrotic area, and also in hepatocytes.

## Supplementary 12

### References

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4. Zeremski M, Petrovic LM, Chiriboga L, Brown QB, Yee HT, Kinkhabwala M, Jacobson IM, Dimova R, Markatou M, Talal AH. Intrahepatic levels of CXCR3-associated chemokines correlate with liver inflammation and fibrosis in chronic hepatitis C. *Hepatology* (2008) 48(5): 1440-50. doi: 10.1002/hep.22500.