Author's response to reviews

Title: Hepatitis C virus E2 protein involve in insulin resistance through an impairment of Akt/PKB and GSK3beta signaling in hepatocytes

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Version: 5 Date: 21 May 2012

Author's response to reviews: see over
Reviewer #1 has no suggested changes for the manuscript
Reviewer #2

1. Ikuo Shoji published a very nice paper in 2011 (Shoji et al, 2011) showing that HCV-induced down-regulation of GLUT2 expression and up-regulation of gluconeogenesis. This suggests that high concentration of glucose in HCV-infected hepatocytes, although interpretation of their data should be cautioned because they used a HCV J6/JFH1-infected cells. It seems that this too could be part of the discussion in this manuscript.

   Answer:
   Thanks for this valuable suggestion and we have added the following sentences in the revised Discussion section. The following sentences are added in revised Discussion section at Line 5, Page 11:
   “In a recent study, it was shown that HCV-induced down-regulation of GLUT2 expression and up-regulation of gluconeogenesis [39]. This suggests that high concentration of glucose in HCV-infected hepatocytes, although interpretation of their data should be cautioned because they used a HCV J6/JFH1-infected cells.”

2. Figure 2 does not show much significant difference in IRS-1 status. Moreover, the actin bands too strong to make it difficult for loading comparison. The immunoblots should be done more clear, and quantification of each bands need to be done more precisely, with SD.

   Answer:
   To make this figure more clearly by following the reviewer’s suggestion, the expression of IRS-1 in HCV E2-transfected Huh7 have been added in Figure 2A.
3. Figure 4: The effect of MG132 was not very significant. The immunoblots should be done more clear, it would help to include scanned data with lower actin level for proper comparison.

Answer:
To make this figure more clearly by following the reviewer’s suggestion, the immunoblots of expression of IRS-1 in HCV E2-transfected Huh7 treated MG132 have been added in Figure 4A.

4. In Figure 5, the mark of phosphorylation is confusing. Please use uniform style for phosphorylation of Akt and GSK3beta in all figures and quantification of each band.

Answer:
The uniform style for phosphorylation of Akt and GSK3beta in all figures and quantification of each band was then corrected accordingly.
Minor criticisms:
In Figure 2C and 6B, the statistical analysis should be done between FLAG and E2.

Answer:
The statistical analysis should be done between FLAG and E2 in Figure 2C and 6B was then corrected accordingly.

Figure 2

Figure 6