Format for ANSWERING REVIEWERS

December 11, 2013

Dear Editor,

Please find enclosed the edited manuscript in Word format (##6436(ID00012386)-review-topic highlight.doc).

Title: Regulation of miRNA by HBV infection and their possible association with control of innate immunity

Author: Xia Jiang, Tatsuo Kanda, Shuang Wu, Masato Nakamura, Tatsuo Miyamura, Shingo Nakamoto, Arup Banerjee, Osamu Yokosuka

Name of Journal: *World Journal of Gastroenterology*

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The manuscript has been improved according to the suggestions of the reviewers:
1 Format has been updated

2 Revision has been made according to the suggestions of the reviewer

(1) The comment from reviewer ***00502982***

Response to your comment: Thank you for your encouraging comments.

(2) The comment from reviewer ***00006976***

Response to your comment: "Please consider making the following changes to the text. Abstract, line 2 '...hepatitis and is one...' Abstract, line 11 '... through the interaction with...' Abstract, line 13 'such as the innate...' Core tip, lines 4-6 '...Toll-like Receptors (TLRs)...', '... host immune responses...', '... and reviews the...' Page 5, paragraph 2, lines 4-6. 'TLR3, TLR7/8 and TLR9 are .....such as double-stranded RNA, single-stranded RNA and DNA.' Page 5, paragraph 2, lines 10-12. '...that mediate sensing.....also involved in recognition of pathogen-associated molecular patterns (PAMPs).' Page 5, paragraph 3, lines 3-5 and page 6, line 1. Please rephrase this sentence. Page 6, line 2 '...and IRF7 via TRIF inducing...' Page 6, line 4 '...MAPKs initiate...' Page 6, paragraph 2, line 2 '..NF-kB is activated by three TLR adaptors...' Page 6, paragraph 2, lines 5-7. Please rephrase the sentence beginning 'There...' Page 6, paragraph 2, line 7 '..were also downregulated...' Page 6, paragraph 2, line 8 ' after challenge with.....was impaired...' Page 6, paragraph 2, line 13. 'It has also been reported that HBV could target...' Page 6 last paragraph is not clear, please rephrase the last 4 lines on this page. Page 6, line 3. '..core promoter mutations...' Page 6, line 6. '...in persons who...' Page 8, line 1. '..endogenous controls...' Page 8, line 2. '..between the test sample...' Page 8, paragraph 3, line 2 '...HepG2.2.15 compared to...' Page 9, line 4. '...to TLR pathways.' Page 9, line 6. '...through MyD88 in HEK293 cells.' page 9, line 9. 'Thus miR-148/152 can act as fine-tuners...' Page 9, line 13. '...induction by stimulation

with....' Page 9, paragraph 2, line 3. '...TLR pathways...' Page 9, paragraph 2, line 3. '...is activated by the...' Page 10, line 2.'...HepG2.2.15 compared to...' Page 10, paragraph 2, line 1-2. '..involved in TLR signalling pathways...' Page 10, paragraph 3. Please rephrase this section.”

Thank you for your valuable suggestions. We agree with you. We revised our manuscript accordingly.

(3) The comment from reviewer ***01566092***

Response to your comment: **“**They, however, discussed only preliminary relationship between the TLR pathways and cytokines/IFNs output (Fig. 1). The manuscript suffers severely from the lack of a clear picture of how HBV triggers innate responses. It was difficult for readers to understand why the miRNA expression profiles link the outcome of the innate immune system. The points in the text were frequently out of the scope.”

Thank you for your comments. We agree with you. We deleted Figure 1 and revised our manuscript accordingly.

Response to your comment: **“**1. The focus of this review and the title are diverged. “Regulation of miRNA by HBV infection and their possible association with control of innate immunity” would be more suitable as a title; In that case, however, the references do not always reflect HBV and innate immunity.”

Thank you for your comments. We agree with you. We changed the title of our manuscript as follows. “Regulation of miRNA by HBV infection and their possible association with control of innate immunity” accordingly.

Response to your comment: **“**2. Their result “showed that HBV persistently infects hepatocytes through the regulation of miRNAs” (page 2 line18; page 10 line6). This is an over-interpretation.”

Thank you for your comments. We agree with you. We revised our manuscript as follows.

In abstract section, page 2, line 18,

We deleted this description.

In “**ROLE OF MIRNAS IN REGULATION OF INNATE IMMUNE RESPONSE IN HBV INFECTION”** section, page 10, lines 10-11,

…..through promoting JAK/STAT signaling pathway by targeting SOCS1, inhibiting HBV replication. The possibility cannot be ruled out that HBV persistently infects hepatocytes through the regulation of miRNAs.

Response to your comment: **“**3. In addition, it is inappropriate trying to include material/result in the writing of a review.”

Thank you for your comments. We revised our manuscript for Topic Highlights accordingly.

Response to your comment: **“**4. The explanation on the cytokines are scattered with very little solid facts on how these cytokines suppress HBV. Similar tendency is found in the elaboration of association between HBV and TLRs. Authors have to screen appropriate literatures.”

Thank you for your comments. We revised our manuscript as follows.

In "**INNATE IMMUNITY IS IMPORTANT FOR THE ERADICATION OF HBV"** section, page 5, lines 7-14,

….. viral mRNA[21-23]. It has been widely believed that the cytotoxic T lymphocyte response clears viral infections by killing infected cells. However, Chisari's group[21-24] reported that noncytopathic clearance of HBV from hepatocytes by cytokines, which abolish viral replication and HBV gene expression, is another important mechanism. Isogawa et al.[24] reported that TLR3, TLR4, TLR5, TLR7 and TLR9 ligands could induce antiviral cytokines and inhibit HBV replication in HBV transgenic mice, thereby indicating TLR activation as a powerful strategy for the treatment of chronic HBV infection. HBV replication can be controlled by innate immune response, involving TLRs, if it is activated in hepatocytes.[24] Together,…..

Response to your comment: "5. Although they mentioned the regulation of type I IFN, they poorly depicted the TRIF/TICAM-1 and MAVS/IPS-1 pathways, which are major pathways for type I IFN induction, rather than the MyD88 pathway."

In "**TOLL-LIKE RECEPTORS AND ANTI-VIRAL DEFENSES"** section, page 6, lines 5-12,

…. and MAPKs through RIPK. TLR3 also activates IFN regulatory factor 3 (IRF3) and IRF7 via TRIF/TICAM-1, inducing the production of type I IFN. The activated NF-κB and IRFs are translocated to the nucleus. NF-κB and MAPKs initiate the transcription of inflammatory cytokine genes, whereas IRFs initiate the transcription of type I IFN[2]. RIG-I and MDA5 pathways can also activate IRF3 to produce type I IFNs. RNA helicases RIG-I and MDA5, specific receptors for double-stranded RNA, and the downstream mitochondrial effector known as CARDIF/MAVS/VISA/IPS-1, are also major pathways for type I IFN induction.…

(4) The comment from reviewer ***00502943***
Response to your comment: Thank you for your encouraging comments.

3 References and typesetting were corrected

Thank you again for publishing our manuscript in the *World Journal of Gastroenterology.*

Sincerely yours,

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