

## Format for ANSWERING REVIEWERS



September 4, 2014

Dear Editor,

Please find enclosed the edited manuscript in Word format (file name: 0051238\_WJH\_revision).

**Title:** Occult Hepatitis B Virus Infection

**Author:** Min-Sun Kwak, Yoon Jun Kim

**Name of Journal:** *World Journal of Hepatology*

**ESPS Manuscript NO:** 13242

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

1 Format has been updated

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

**(1) Reviewer 1:**

1) Kwak et al review occult hepatitis B infection. This article is very interested. However, some revisions are necessary. The authors defined OBI as the presence of HBV DNA in the liver in the absence of detectable HBsAg. However, it is the definition which was determined in 2008 (Journal of Hepatology). Because the definition of OBI was not clear before 2008, the international workshop was hold in Italy I think that several definitions have been used in the OBI articles which the authors cited in this review. If the definition is different among articles, the authors should show us the difference of the definition in each article. Readers will be confused if the authors compare the articles which have different definitions.

**Answer:**

According to the reviewer's comment, we added the difference of the definition of OBI in the "definition and classification section" as follows;

"Several definitions for OBI have been suggested. In 2008 international workshop held in Italy, OBI was defined as the presence of HBV DNA in the liver (with or without HBV DNA in the serum) with

negative HBsAg by currently available assays <sup>[8]</sup>. Serum HBV DNA can be either detectable or undetectable, and when detectable, the level of HBV DNA is usually very low (<200 IU/mL). When serum HBV DNA levels are comparable to those usually detected in cases of overt HBV infection, "false OBI" should be considered. False OBI is usually due to rare infection with S gene escape mutants, which produce a modified HBsAg that is not recognized by routinely used detection assays <sup>[9, 10]</sup>. Defining OBI by liver HBV DNA is the most stringent way as HBV DNA from the liver can be detected even when HBV cannot be detected in serum <sup>[11]</sup>. However, obtaining hepatic HBV DNA is difficult in clinical practice, and assays for detection of HBV DNA in liver tissue are not well-standardized <sup>[7]</sup>.

HBV DNA detection assay in serum have been used with sufficient sensitivity, therefore, OBI is often defined as the presence of serum HBV DNA without detectable HBsAg in clinical practice and in many studies <sup>[7, 8, 12]</sup>. Brechot *et al.* proposed that occult HBV infection be defined as the detection of HBV DNA by PCR or other amplification assays in HBsAg-negative individuals <sup>[9]</sup>. Allain also defined OBI as the presence of HBV DNA without detectable HBsAg with or without anti-HBc or anti-HBs outside the pre-seroconversion window period <sup>[13]</sup>. This definition of OBI by serum HBV DNA is most commonly used in clinical practice.

OBI has also been defined as a serological condition characterized of isolated hepatitis B core antigen (anti-HBc) in the absence of HBsAg and anti-HBs <sup>[3, 14]</sup>. Anti-HBc, a surrogate marker of OBI, is useful when an HBV DNA test is not available or when intermittent viremia is suspected <sup>[8, 15]</sup>. However, when OBI is defined by anti-HBc alone, false positive and false negative of anti-HBc for the detection of OBI should be considered. Not all anti-HBc positive subjects are positive for HBV DNA. In addition, the absence of anti-HBc does not exclude seronegative OBI. As above, definition of OBI slightly differs according to the study, thus cautious interpretation should be exercised when comparing study results about OBI."

2) In the introduction, the authors should describe the history of occult hepatitis B infection (OBI). The description of the history of OBI will be helpful for reader to understand the pathogenesis of OBI. If possible, the authors should show new information as much as possible. Several reviews of OBI has recently been published (Gerlich WH: Dig Dis 2010, Hollinger FB: J viral Hepat 2010, Said ZNA: World J Gastroenterol 2011).

**Answer:**

According to the reviewer's kind comment, we added the description of the history of OBI in the introduction as follows;

"Chronic HBV infection is characterized by persistent HBV surface antigen (HBsAg) and viremia. In the past, clearance of HBsAg in patients with chronic hepatitis B was considered as remission of the disease and disappearance of viral DNA <sup>[1]</sup>. However, 'occult' or 'silent' form of HBV infection was first reported in late 1970s in blood donors with anti-HBc Ab without HBsAg who transmitted hepatitis B <sup>[1, 2]</sup>. The meaning of this clinical entity has been reviewed in 1998 by a panel of Europe and USA scientists as a proportion of serological pattern 'anti-HBc alone', although the term 'occult' was not used at that time <sup>[3]</sup>. And more and more data showed persistent low level of HBV DNA in serum and liver tissue after HBsAg clearance after acute self-limited or chronic HBV infection. Demonstration of this clinical entity has brought about the concept of 'occult or silent' HBV infection indicating presence of HBV DNA in the absence of detectable hepatitis B surface antigen (HBsAg) <sup>[3, 4]</sup>."

**(2) Reviewer 2:**

1) This article reviews our understanding of occult hepatitis B infection. Unfortunately it comes soon after an overlapping article has been published in the same journal (June 27, 2014 issue), that one dealing with the outcome of OBI in the immune compromised. The authors have defined OBI as positive HBV DNA in the liver in the presence ('seropositive') or absence ('seronegative') of serum markers (anti-HBc and / or anti-HBs). Although this is a stringent definition, the authors have clarified that it is also clinically difficult, and most studies use positive HBV DNA in serum (in the absence of HBsAg) as a surrogate marker. Their subsequent review also refers to such studies. In fact, the recent WJG article defines OBI as HBV DNA detectable in serum (low levels) and / or liver, in the absence of HBsAg/anti-HBs, and has even accepted anti-HBc positivity as a surrogate marker. The authors should therefore 1. Clarify this difference of opinion.

**Answer:**

According to the reviewer's comment, we added the difference of the definition of OBI in the "definition and classification section" as follows;

"Several definitions for OBI have been suggested. In 2008 international workshop held in Italy, OBI

was defined as the presence of HBV DNA in the liver (with or without HBV DNA in the serum) with negative HBsAg by currently available assays <sup>[8]</sup>. Serum HBV DNA can be either detectable or undetectable, and when detectable, the level of HBV DNA is usually very low (<200 IU/mL). When serum HBV DNA levels are comparable to those usually detected in cases of overt HBV infection, “false OBI” should be considered. False OBI is usually due to rare infection with S gene escape mutants, which produce a modified HBsAg that is not recognized by routinely used detection assays <sup>[9, 10]</sup>. Defining OBI by liver HBV DNA is the most stringent way as HBV DNA from the liver can be detected even when HBV cannot be detected in serum <sup>[11]</sup>. However, obtaining hepatic HBV DNA is difficult in clinical practice, and assays for detection of HBV DNA in liver tissue are not well-standardized <sup>[7]</sup>.

HBV DNA detection assay in serum have been used with sufficient sensitivity, therefore, OBI is often defined as the presence of serum HBV DNA without detectable HBsAg in clinical practice and in many studies <sup>[7, 8, 12]</sup>. Brechot *et al.* proposed that occult HBV infection be defined as the detection of HBV DNA by PCR or other amplification assays in HBsAg-negative individuals <sup>[9]</sup>. Allain also defined OBI as the presence of HBV DNA without detectable HBsAg with or without anti-HBc or anti-HBs outside the pre-seroconversion window period <sup>[13]</sup>. This definition of OBI by serum HBV DNA is most commonly used in clinical practice.

OBI has also been defined as a serological condition characterized of isolated hepatitis B core antigen (anti-HBc) in the absence of HBsAg and anti-HBs <sup>[3, 14]</sup>. Anti-HBc, a surrogate marker of OBI, is useful when an HBV DNA test is not available or when intermittent viremia is suspected <sup>[8, 15]</sup>. However, when OBI is defined by anti-HBc alone, false positive and false negative of anti-HBc for the detection of OBI should be considered. Not all anti-HBc positive subjects are positive for HBV DNA. In addition, the absence of anti-HBc does not exclude seronegative OBI. As above, definition of OBI slightly differs according to the study, thus cautious interpretation should be exercised when comparing study results about OBI."

2) Cover mainly the relevance of OBI in immunocompetent individuals

**Answer:**

The reviewers' comments are correct, and we thank them for the accurate comments. However, we regret that we were not noticed the review article entitled “Clinical impact of OBI in

immunosuppressed patients” (in *World J Hepatol* 2014; 27: 384) before the decision of this topic has been made. And, this article is mainly about OBI in immunocompetent hosts, except for the section “Reactivation.”

3. English editing was done by native speaker (file attached).

Thank you again for inviting me as a reviewer of *World Journal of Hepatology*.

Sincerely yours, Yoon Jun Kim, MD, PhD

Department of Internal Medicine and Liver Research Institute

Seoul National University College of Medicine

101 Daehang-no, Jongno-gu, Seoul 110-744, South Korea.

E-mail: yoonjun@snu.ac.kr

Telephone: +82-2-20723081 Fax: +82-2-7436701