SUMMARY

THE PREVALENCE OF OCCULT HEPATITIS B INFECTION AND MOLECULAR ANALYSIS ON SURFACE REGION OF HEPATITIS B VIRUS (HBV) AMONG PREGNANT WOMEN IN PERAK PUBLIC HEALTH CENTER, SURABAYA

Hepatitis B Virus (HBV) infection is still a major health problem in the world, as well as in Indonesia. Indonesia has a moderate to high endemic level of hepatitis B infection. Perinatal transmission mode is the most common transmission of hepatitis B infection in hyperendemic region. The prevalence of HBsAg in Indonesia was reported 5% - 10% among the general population (Khan et al, 2004).

Hepatitis B virus infection is diagnosed by hepatitis B surface antigen (HBsAg) assay. HBsAg is one of the serologic marker for HBV infection. HBsAg could be detected on the second week after HBV infection. Occult hepatitis B infection (OBI) defined as the presence of HBV DNA in serum with undetectable HBsAg (Raimondo et al, 2008). Failure of HBsAg detection may be due to mutations in *'a'* determinant region, and/or the low level of viral load titer. This study was used polymerase chain reaction (PCR) method, because PCR was the gold standard to detect the HBV DNA (Weber et al, 2003).

The HBV genotypes, subgenotypes, and subtypes were associated with geographical distribution, clinical characteristics and historical information on the migration pattern of local's ancestor. HBV genotype B and C to be predominant in Asia including in Indonesia. This study was aimed to determine the prevalence of OBI and to indentify the genotype, subgenotype, subtype and mutations in the *'a'* determinant region among pregnant women from Perak Public Health Center, in Surabaya. The serum samples were stored in the Hepatitis Laboratory, Institute of Tropical Disease, Airlangga University. All samples in this study were obtained from persons with HBsAg-negative and positive for anti-HBc.

HBV DNA was amplified by nested polymerase chain reaction (PCR) with one or two pairs of primers that specific for surface region of HBV. Amplification products were visualized on a 2% agarose gel, then they were purified, labeled, and sequenced. The HBV nucleotide sequences were compared with those from international DNA data bank (DDBJ/GenBank) for HBV genotypes and subgenotype determination. The amino acids of the HBV were deducted from the nucleotide sequences. HBV subtype was aligned and compared with the amino acid substitution at positions 122, 127, 134, 143, 159, 160, 177 and 178 of the S genes. Mutations in the *'a'* determinant region were determined by analyzing of amino acid substitution at positions 121 to 149.

This study identified 9 of 50 (18%) HBsAg-negative and anti-HBc positive samples who were positive for HBV DNA. The HBV/B3-*adw2* was predominant in 7 (77.7%), followed by HBV/B9-*ayw1* (11.1%), and HBV/C7-*adrq+* (11.1%). Interestingly, isolate which classified into HBV/C7-*adrq+* was came from Madura ethnic, in contrast with the previous study had reported subtype *adrq+* was predominant in Papua.

Three (33,3%) of 9 isolates have amino acid mutations within *'a'* determinant. Mutations T126I, T140I, and Q129H, T131N, M133S. Six of 9 (66,7%) isolates had HBV DNA with the wild-type S gene. Failure of HBV detection by HBsAg assay in this study might be due to amino acid mutations in the *'a'* determinant which could alter HBsAg antigenic property. Another factor could be the low titer of viral load as a result of suppressing of viral replication and keeping the infection under control (Brechot et al, 2001). This is supported by the fact that HBV DNA in these samples was detectable only in the second-round of nested PCR.