

Analisis efektivitas metode NAT untuk uji saring virus hepatitis B pada darah asal donor dengan hepatitis B occult = Analysis of NAT effectivity for hepatitis B virus screening on occult hepatitis B blood donors

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Deskripsi Lengkap: <https://lib.ui.ac.id/detail?id=20404326&lokasi=lokal>

Abstrak

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Latar belakang. Hepatitis B merupakan salah satu masalah kesehatan yang serius, diperkirakan lebih dari 2 milyar orang didunia telah terinfeksi virus hepatitis B (VHB). Dari jumlah ini kira-kira 360 juta orang mengalami infeksi khronis. Kematian terutama disebabkan karena sirosis hepatis dan karsinoma hepatoseluler. Salah satu upaya pencegahan penularan infeksi VHB adalah uji saring darah donor terhadap hepatitis B surface antigen (HBsAg) yang merupakan pemeriksaan skrining yang dilakukan oleh Unit Transfusi Darah (UTD) di negara berkembang seperti Indonesia. Banyak peneliti membuktikan bahwa darah HBsAg negatif masih berpotensi menularkan infeksi VHB. Untuk itu meningkatkan keamanan darah, beberapa negara menambahkan parameter pemeriksaan antibodi terhadap hepatitis B core (anti-HBc) sebagai petanda paparan terhadap infeksi VHB dan pemeriksaan antibodi terhadap hepatitis B surface (anti-HBs) sebagai tanda respon imun terhadap infeksi VHB. Dengan berkembangnya teknologi biologi molekuler, masa jendela infeksi VHB dengan seronegatif dapat di ketahui lebih cepat melalui deteksi DNA VHB dengan metode Nucleic Acid Test (NAT) multipleks yang dilanjutkan dengan NAT discriminatory. Beberapa penelitian terdahulu menunjukkan, didapatnya DNA VHB pada spesimen darah donor yang seronegatif dengan metoda NAT. Darah dengan HBsAg negatif, DNA VHB positif dengan atau tanpa anti-HBc dan atau anti-HBs merupakan darah asal donor dengan Hepatitis B Occult (HBO). Prevalensi donor dengan HBO di Indonesia berkisar antara 8- 10%.

Metodologi. Penelitian ini menggunakan desain potong lintang (cross sectional) yang dilakukan di UTDP dan Lembaga Biologi Molekul Eijkman, dengan jumlah sampel 4.973 asal subyek donor darah dari 4 UTD daerah DKI, Kota Tangerang, kota Depok dan Kabupaten Tangerang. Terhadap sampel penelitian dilakukan pemeriksaan serologis HBsAg, anti-HBc, anti-HBs, NAT, dan PCR kuantitatif dan kualitatif, selanjutnya pada sampel yang HBsAg negatif, NAT positif dan didapatkan hasil PCR kuantitatif positif dilakukan pemeriksaan lanjutan sequencing asam amino untuk mengetahui ada tidaknya mutan HBsAg penyebab lolosnya deteksi serologi HBsAg oleh reagensia HBsAg yang digunakan untuk uji saring darah donor.

Hasil. Didapatkan hanya 20 subjek (0,40%) mempunyai hasil HBsAg negatif dan NAT positif multipleks, dan hanya 16 subyek (80%) HBsAg negatif dan NAT discriminatory positif. Lebih lanjut hasil pemeriksaan anti-HBc negatif dan anti- HBs positif/negatif didapatkan hanya 1 subyek (6,25%), anti-HBc positif dan anti-HBs negatif didapatkan 9 subyek (56,25%), hasil pemerksaan anti-Hbc dan anti- HBs positif 5 subyek (31,25%). Lebih lanjut dilakukan pemeriksaan PCR kualitatif dan didapatkan 3 subyek (18,75%) tidak terdeteksi, , 6 (37,5%) subyek menunjukkan hasil viral load yang low detection (dibawah sensitivitas alat), dan 7 subyek (43,75%) menunjukkan hasil viral load dapat di ketahui. Pada pemeriksaan PCR kualitatif dan sequencing didapatkan 2 subyek (28,57%) ditemukan mutasi pada gen S pada posisi 143 dimana terjadi

substitusi asam amino T143M.

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ABSTRACT

Background. Hepatitis B is one of the most serious health problem. It is estimated that more than 2 billion people have been infected by this virus, of which 360 million are chronically infected with severe and fatal risk especially of cirrosis and hepatocellular carcinoma. One of the main ways to prevent transfusion transmitted HBV infection is blood screening for HBsAg. However, many studies have proven that HBsAg negative blood can still be infection. Therefore to enhance blood safety same countries have added antibody parameters in blood screening of donors – antibody for hepatitis B core antigen (anti-HBc) as marker for HBV infection and antibody for hepatitis B surface antigen (anti-HBs) as marker for immunological response to HBV infection.

And with the development of molecular biology technology, HBV infection can be knowing faster in seronegative windows period with HBV DNA examination inspection by methods Nucleic Acid Test (NAT) multiplex and discrimenatory. In fact the results of seronegative blood is still there HBV virus with NAT, and this result we can called with the Occult hepatitis B (HBO). Prevalence of donors with HBO ranges 8-10% in Indonesia. This study aims to determine the efectivity analized by NAT method blood donor with HBO to see continuity with the examination of anti-HBc, anti-HBs, viral load, and the cause of the HBO that mutations in the gene encoding HBsAg.

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Result. There was 20 samples (0.40%) had results of HBsAg negative and positive NAT multiplex, and only 16 samples (80%) HBsAg negative and positive discrimenatory NAT. Furthermore, the results of the examination of anti-HBc and anti-HBs negative positive / negative obtained only 1 samples (6.25%), anti-HBc positive and negative anti-Hbs obtained 9 samples (56.25%), and anti-HBc , anti- HBs positive are 5 samples (31.25%). and then , qualitative PCR examination and had 3 samples (18.75%), is not detected, 6 (37.5%) samples, and some samples showed a low viral load results detection (sensitivity under tools value), and 6 samples (43.75%) shows viral load is positive. In qualitative PCR and sequencing obtained 2 samples (28.57%) found a mutation in the S gene at position 143 where there T143M amino acid

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