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Growıng Threat: Vaccıne Escape Mutant Hepatıtıs B Vıruses

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Abstract: *HBV is a member of Hepadnavirus genus. HBV is a DNA virus with a diameter of 42 nm consisting of an outer lipid envelope which contains proteins HBsAg, pre-S1 and pre-S2. HBsAg is a product of 's' gen. The s geneinvolves 'a' epitope- 124. -147. codones and this region is the affecting region for anti-HBs antibody binding. Vaccine /Immunglobulin escape mutations occur at HBV DNA s gene's 'a determinant' region, 127-149. codones. These mutations causes inability to bind of anti HBs antibody to HBsAg. Also, these variants are transmissible among humans although individuals were vaccined. The result of our literature investigation, the number of the cases which caused by vaccine escape mutant variants were found to be increased, especially in high endemicity region. Because of that, we think vaccine escape mutants are growing very important public health problem.*

Keywords: *Hepatitis B virus infection; Vaccine escape;Hepatitis Bsurface antigen mutants*.

1. INTRODUCTION

Hepatitis B virus is a DNA virus which is a member of hepadnaviridae. It is responsible nearly 300 million chronic HBV infection and over 1 million deaths per year due to HBV-related end-stage liver disease, liver cirrhosis and liver cancer.HBV viruses can be present all of the body fluids and blood of the infected person. So, it can be transmitted such as perinatally, sexually, blood transfusions, unsafe injection, injecting drug with using mutual syringes and occupational exposure of health care workers [1].

Currently, HBV viruses have been classified into 10 genotypes [A-J]which can be further sub-divided into over 40 sub-genotypes [2-4].Genotype D and subgenotype D1 HBV virus infections are common in Turkey[5,6,7].

The worldwide prevalence of chronic HBV infection in the general population is 5%, but it differs from one geographical area to another, example 0.1%-2.0% in the United States and Western Europe, 2.0%-8.0% in Eastern Mediterranean countries [such as Turkey] and Japan, and 5.0%-20.0% in South-Eastern Asia and sub-Saharan Africa.In highly endemic areas the majority of chronic carriers acquire HBV infection at birth or in the first decade of life, whereas in countries with a low endemicity, HBV transmission occurs mostly in adulthood due to unsafe sexual contact, using mutual syringes or parenteral exposure to contaminated medical equipment or blood transfusion[8].

Because of it's caused diseases, some strategies have been devoloped to fight HBV infection such as treating the chronically infected patients as much as possible, preventing the transmission and immunizing susceptible individuals.Among them, vaccination is the most effective by preventing individuals from contracting HBV infection[9]. Thirty years ago the first HBV vaccine was presented to use in fight against HBV. The vaccine contains highly antigenic the subviral particle"; HBsAg [10].

HBV vaccines have highly antigenic effect on immun system. It cause creating antibodies against to HBV surface antigen [HBsAg]directly.HBsAg is a product of 's' gen. The 'a' epitope involves 124. -147. codones and this region is the affecting region for anti-HBs antibody binding. The mutations in this area can cause conformational changes at HBsAg and create *'vaccine escape mutant variants*'[11]. This situation is a public health threat and these mutants can cause infections at vaccined individuals. Such mutant variants may be dominant variant HBV virus in future. The aim of this review is providing attention to this point.

2. HBV VIROLOGY

HBV is a member of Hepadnavirus genus. HBV is a DNA virus with a diameter of 42 nmol/L consisting of an outer lipid envelope which contains proteins HBsAg, pre-S1 and pre-S2 . These are provide the viral binding, entry into the hepatocytes [12,13].

The HBV genome consists of ~3.2 kb, 3200 base pairs ofpartially double-stranded circular DNA containing four[P, C, S and X] overlapping open reading frames [ORF]with a nucleotide diversity of ≥ 8% in different genotypes[14].The core [C] gene encodes the capsid protein, HBcAg, which is the major structural protein of the nucleocapsid. The preC/C ORF is transcribed into a precore/core fusion protein. The X gene gives rise to a protein of not completely understood function but it has transactivating functions. The P gene codes for the viral polymerase/reverse transcriptase.In addition, the HBV polymerase [pol] gene completely overlaps with the envelope [S] gene.The three envelope proteins are encoded by a single ORF with three in frame start codons. Consequently, they share a common C-terminus that provides the membrane anchor and differ in length at their N-termini. They are designated the Large [L], Middle [M-] and Small [S-] surface protein, respectively, with the first-mentioned playing the major role in this proposal. Compared to the S-protein [the classic HBs-Ag], the M-protein is extended to the 55 preS2- and the L-protein additionally to the 108 preS1-encoded amino acids [15].

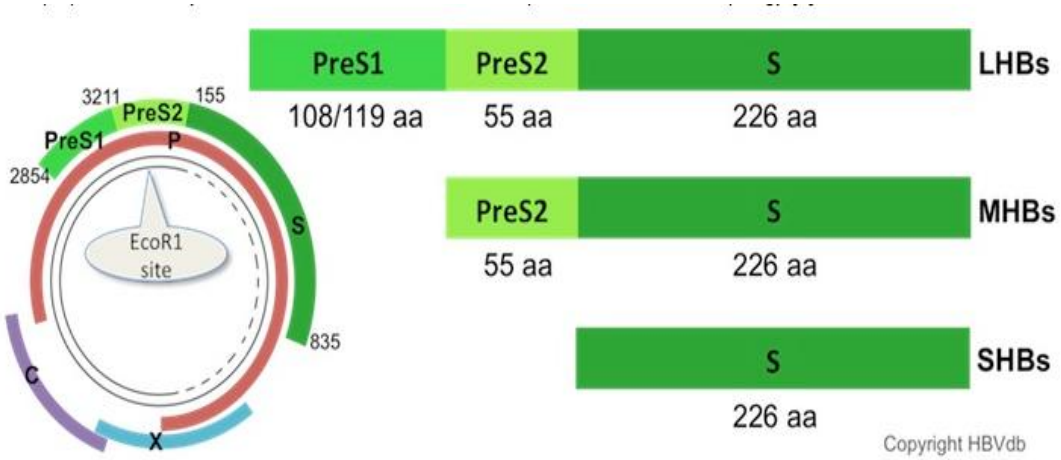

Figure1. *HBV Envelope structure and envelope proteins[16].*

Currently the primary element for diagnosis and target of immunoprophylaxis of HBV infection is HBsAg. The dominant epitopes of HBsAg, which are the targets of neutralizing B-cell responses, reside in the ‘‘a’’ determinant [aa 124-147] within the major hydrophilic region [MHR]. It is in a form of two major and one minor loops with cysteine-disulphide bonds, protruding from the outer surface of the virus. The second hydrophilic loop [aa 139 to 147 or 149] is the major target for neutralizing anti-HBs produced following natural infection or vaccination[17].The most common pattern mutations are sP120T, sS143L, sD144A/E, sE164D, sL127P, sG129H, sM133I/T, sP/T134A/L, sS140L, sG145A/R, sW172, and sW182 in the literature [18]. But the variant sG145R was identified mostly as a vaccine escape mutant in these literature [54%]. It has been demonstrated that it is also associated with HBV-related chronic liver disease in non-vaccinated subjects[19]. On the other hand, the horizontal transmission of the sG145R mutant strain has been reported[20]. Naturally occurring surface gene variants have also been reported around the world in persons who have not been immunized[21,22].

The ‘a’ determinant is common all genotypes. Neutrolizing antiHBs response which vaccine caused,provides protection against all HBV genotypes andsubtypes.Alterations in this region of the surface antigen can determine conformational changes thatcan allow replication of mutated viruses in vaccinated people [vaccine escape mutants or VEMs][23].

3. LITERATURE REVIEW

Currently, we can use five oral anviral regimens [Lamivudine, Telbuvidine, Adefovir, Entecavir, Tenofovir] and pegile interferons subcutaneously for the treatment of chronic hepatitis B. Especially, low potential antiviral drugs are responsible of drug resistance at *p* genes. Because of *s* genes and *p* genes overlap, a mutation at *p* genes, especially mutual region for *p* and *s* genes, can effect eachother.

So, drug associated *p* gene mutations, can cause *s* gene mutant variants. If mutation occurs at 'a determinant' of *s* genes, the mutation effects the response to vaccine. Out of this regions,occurred mutations in the area which including 161, 164, 172, 173, 175, 176, 182, 193, 194, 195, 196. codones also cause vaccine escape mutations [24].In addition, the mutations in *"a"* epitope correlate with the absence of detectable anti-HBsAg. These mutations also called as Antiviral Drug Associated Potential Vaccine Escape Mutation-ADAPVEM at the patient who received orally antiviral drugs previously.

The mutation occured during antiviral treatment cause NUC resistances. These mutations can cause changes in the overlapping reading frame and alter the C-terminal region of HBsAg [25]. LAM-resistant HBV [rtV173L + rtL180M + rtM204V] has been shown to cause two amino acid changes in the overlapping surface gene [sE164D + sI195M],and cause reduction at anti-HBs binding similar with the vaccine escape mutant sG145R [34, 35]. This situation significantly reduces antiHBs binding due to changes in the HBsAg and effects vaccine and HB-Ig response [18]. As a result, antiviral drug resistance can effect the HBsAg protein structure and anti HBs binding in these patients. ADAPVEM's are important potential threat for public health. Because these mutant variants can be dominant variant among the HBV viruses and it can be transmissible an individual to another although the vaccinewas applied[26].

Since 1990, vaccine escape mutant has been reported as an important public health threat. A literature investigation about vaccine escape mutation [VEMs] was done from 1990 to 2015. We obtained total 69reports about vaccine escape mutant HBV variants. Turkey has nine reports[18,24,26-32]. China has eight reports[33-40]and Taiwan has seven reports[41-47]. England have five reports[48-52]. Thailand, Japan, Italy USA, India, Iran have four reports from each country[53-76]. France has three reports[77-79]. Argentina have two reports[79-81] and the other countries have one report from each country total 11 reports[82-92].

Firstly, an Italian infant has been established as having a vaccine escape mutant [G145R] variants [61]. In 1990-2000 years total 14 report, in 2001-2010 years total 30 report and in 2011-2015 years 24 report.Most of the reports were including G145A/R mutations and most of the reports were from high endemicity regions.

It was observed that, this problem is growing too fast in the world wide. Especially in the countries which are situated in high endemicity region such as China, Thailand, Taiwan, China.Monitoring for more than a decade in these countries has shown that hepatitis B immunization programmes have increased the incidence of HBV vaccine escape variants [35,54].

In China, the prevalence of "α" determinant mutants in the children increased from 6.5% in 1992 to 14.8% in 2005, where the G145R mutant occurred most frequently. In contrast, mutation frequencies showed little difference between 1992 [9.4%] and 2005 [9.9%] in adults[93].

Similarly, in Taiwan, the prevalence of hepatitis B surface gene a determinant mutants increased from 7.8% before the vaccination program, to 19.6%, 28.1% and 23.1% at 5, 10 and 15 years after the program[44].

Turkey is an middle endemicity area [2–7%]. A recent study in Turkey found an overall prevalence of 4.19% for HBsAg, but the prevalence rates are highest in Southeastern Turkey [9–11%]. In 1997, all of the new borns were vaccined according to universal vaccination program of WHO and CDC in the Worldwide. By 1998, vaccination against to HBV was started in Turkey[94]. In Turkeyinterest in this subject is increasing every day. Especially, Sayan contributesmany studies to Turkish literature in this subject.

First HBV vaccine escape mutation [sT143stop codon] was observed in Turkey in a child with CHB[27].After that,Ozaslan et al.[28]sequencing the amplified surface gene region has suggested sM125T and sT127P mutations as HBsAg escape mutations in Turkish patients with CHB and their family members.They had investigated 40 HBV DNA positive patients among 132 HBsAg carriers.Ten kinds of point mutations had been identified within the S region. The highest rates of mutation had been obtained in chronic hepatitis patients and their family members. The amino acid mutations 125 [M -> T] and 127 [T -> P] had been found on the first loop of 'a'-determinant [28]. Another study has been described a diagnostic HBsAg escape mutation [sS143L, sQ101H, sS117N, sT118R, sP120Q] causing chronic HBV infection in a previously vaccinated treatment-naı̈ve Turkish patient[29].

The most comprehensive study about this subject is Sayan ‘s study in Turkey. Sayan et al [18]had evaluated 142 patients who are undergoing treatment and 185 patient who are treatment naive. The sP120T, sG130R, sM133I, sY134N, sD144E, sS143L and sG145R mutations had been obtainedin their study. The prevalence of typical HBsAg escape mutations had beendetected treatment naive and treatment groups 8.1% and 8.5% respectively. In both groups sG145R escape mutation had been observed. The sG145R escape mutation prevalence had been detected at a low frequency [1.2%] in Turkish patients with CHB.

Sayan et al. [24] also had been evaluated 94 patients who HBsAg-positive and having hemodialysis. They had been observed ADAPVEM mutations which located in 161, 164, 172, 173, 175, 176, 182, 193, 194, 195, 196. codones at 43 patients of these 94 hemodialysis patients.

A case report who has sS143T, sD144E, sG145R, sE164D and sI195M mutations all together.had been presented by Sayan recently. It was emphasized that the patient as a first case including G145R mutation in Turkey[37].

Kaymakoğlu et al. [31] reported a case about an acute hepatitis B caused by immune escape variants in the absence of any immunosuppression or cytotoxic chemotherapy.

Also, we evaluated recently a case who experienced with pegile interferon, lamivudine and entecavir 0.5 mg and 1 mg. Despite treatment his viral load increased, we applied sequence analysing and observed T127P vaccine escape mutations[32]

4. CONCLUSION

Vaccine /Immunglobulin escape mutations occur at HBV DNA *s* gene’s ‘a determinant’ region, 127-149. and 161-195codones. It can be a result of previous NUC treatment. These mutant variants are capable to prevent anti HBs binding. Also, these variants are transmissible among humans. Because of that, ADAPVEM formation should be monitored. In addition, such mutated viruses can be undetectable by the current diagnosticassays. Because ot that, we think, VEMs are growing very important public heath problem. Required precaution methods should be taken immediately. New vaccines development studies should be done as soon as possible.

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