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Resistant Mutations Against Nucleot(s)ide Analogues (NAs) in RT Domain of HBV Genome: A Review

HBV Genomunun RT Alanındaki Nükleotid Analoglarına (NA) Karşı Dirençli Mutasyonlar: Bir Derleme

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ABSTRACT

Drug resistance is a significant hurdle in the control and treatment of chronic hepatitis B virus (HBV) infection. This resistance is caused by some mutations in the viral genome which enable alternative ways for viral replication; which otherwise is blocked by nucleot(s)ide analogues (NAs). To overcome this hurdle, the correct drug selection is required because a specific mutation may bestow upon the virus resistance against a particular drug, but it remains susceptible to others. Significant literature has been published on the topic since the start of NAs use as a treatment option for chronic HBV patients. This review summarizes all the literature published on resistant mutations in the reverse transcriptase domain of the HBV genome, most of which have been reported. After a detailed screening, 36 studies were included in the final review. It is concluded that the most frequent mutations related to resistance are rtM204V/S/I/Y, rtM180C/L/I/T, rtN236T, rtA181T/V/S and rtV173L. Mutations rtT184S, rtN238D/S/R, rtA194T, rtL80V/G/I, rtS202I/G/C, rtV214A/T/I, rtV207L/M, rtI169T/P, and rtM250V/I/L are less common but are also reported to be associated with viral resistance.

Keywords: HBV resistance, HBV RT domain, resistant mutations, nucleot(s)ide analogues

ÖZ

İlaç direnci, kronik hepatit B virüsü (HBV) enfeksiyonunun kontrol ve tedavisinde büyük ve zorlu bir engeldir. Bu dirence, viral replikasyona alternatif yollar sağlayan viral genomdaki bazı mutasyonlar neden olur, aksi takdirde nükleotid analogları (NA) tarafından bloke edilir. Bu engelin üstesinden gelmek için doğru ilaç seçimi gereklidir, çünkü belirli bir mutasyon virüse belirli bir ilaca karşı direnç kazandırabilir ancak diğer bazı ilaçlara karşı duyarlı olmaya devam edebilir. NA'ların kronik HBV hastalarında tedavi seçeneği olarak kullanılmaya başlanmasından bu yana konuyla ilgili pek çok literatür yayınlanmıştır. Bu derleme, HBV genomunun ters transkriptaz alanındaki dirençli mutasyonlar hakkında yayınlanmış ve çoğunun rapor edildiği tüm literatürü özetlemektedir. Ayrıntılı bir taramanın ardından 36 çalışma nihai incelemeye dahil edildi. Dirençle ilgili en sık görülen mutasyonların: rtM204V/S/I/Y, rtM180C/L/I/T, rtN236T, rtA181T/V/S ve rtV173L olduğu sonucuna varılmıştır. rtT184S, rtN238D/S/R, rtA194T, rtL80V/G/I, rtS202I/G/C, rtV214A/T/I, rtV207L/M, rtI169T/P ve rtM250V/I/L mutasyonları daha az yaygındır ancak aynı zamanda viral dirençle ilişkili olduğu da bildirilmektedir.

Anahtar Kelimeler: HBV direnci, HBV RT alanı, dirençli mutasyonlar, nükleotid analogları

Introduction

Hepatitis B virus (HBV) is a pathogen infecting human liver cells and is a member of the hepadnaviridae family, of viruses (1). Undesirable effects caused by HBV include liver degeneration, liver cirrhosis, hepatocellular carcinoma (HCC), and end-stage liver disease (2). worldwide, approximately 257 million people are chronic carriers of HBV. The annual number of deaths caused by HBV-related infections was estimated to be 887,000 in 2015 (3).

Different types of interferon and nucleot(s)ide analogues (NAs) are clinically available treatments for chronic HBV patients. Interferon reduces the hepatitis B surface antigen level from blood while also having immunomodulatory effects, but it poses many adverse side effects. The process of reverse transcription is

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targeted by NAs acting on the reverse transcriptase (RT) domain of the viral genome, stopping the production of DNA from pre-genomic RNA (1,4). NAs therapy reduces the level of HBV-DNA to very low or untraceable amounts, which reduces the possibility of death and HCC (3). Lamivudine (LAM), adefovir dipivoxil (ADV), entecavir (ETV), telbivudine (Ldt), and tenofovir (TDF) are NAs approved and used to date for chronic HBV treatment. NAs therapy is considered to be more convenient and has fewer side effects than interferon (5).

As mutations occur at a high rate in the HBV genome, due to the lack of a proofreading mechanism, complications are also associated with long-term NAs therapy. Some mutations in the HBV genome give resistance to the virus against the NAs (2). Resistant mutations in viral genomes change the interaction of HBV polymerase and NAs, causing inhibitory effects on drug action. To restore replication activity, primary mutations occur in addition to compensatory and secondary mutations, thus increasing the viral resistance against the drug (5,6). RT domain mutations have specific names according to a continuous numbering system introduced by Stuyver et al. (7). In the current analysis, we aimed to compile the literature available on resistance-related mutations related to NAs in the RT domain of the polymerase region in the HBV genome.

Methodology

We conducted a literature search using databases like "Google Scholar", "PubMed", "Scopus", and "Web of Science" using keywords: "resistant mutations", "NAs", "HBV", "LAM", "TDF", "ETV", "adefovir", "Ldt". The inclusion criterion was set as a study analyzing resistant mutations in the HBV RT domain of treatment non-responders. We initially identified 269 records from the databases. Studies in languages other than English were not included in this review (n=4). Records that did not have full access were also excluded (n=121). A total of 30 studies were excluded, having been found to be duplicates on screening. A total of 114 full-text articles were accessed for eligibility. After full text assessment, 67 studies were excluded because they did not report the mutational analysis of the RT domain, while 11 articles were excluded because they reported mutations from treatment-naive patients and not from treatment non-responder patients. After completing the exclusion process (Figure 1), a total of 36 studies were included in the current review.

Review

A total of 36 studies fulfilling all inclusion criteria were finally selected in the current review (Table 1). In these 36 studies, 14 were found from China (8,9,10,11,12,13,14,15,16,17,18,19,20, 21), 4 from different states of the USA (22,23,24,25), 3 each from Australia (26,27,28), and Korea (2,29,30), 2 from Germany (31,32), and one from each of Iran (33), Iraq (34), Turkey (35), Cyprus (36), Italy (37), Sweden (38), Argentina (39), Hong Kong (40), Pakistan (41), and Japan (1) (Figure 2, Table 1).

Common Mutations Associated with HBV Resistance

Very much frequent mutations were reported on positions rt204 and rt180 of HBV genome in literature. The most frequent mutation associated with viral resistance was found to be rtM204I/V/S/Y, which was reported by 29 studies from different countries included in the current review (Figure 2, Table 1). It was found to be associated primarily with LAM and Ldt resistance; however, it is suspected to have some role in viral resistance against all other NAs too, i.e., adefovir, ETV and TDF. The mutation rtL180M/L/C/I/T was reported in 25 studies included in the current review (Figure 2, Table 1). This mutation has been reported to be primarily associated with LAM and Ldt resistance, but it is assumed that it also has some role in viral resistance against adefovir, ETV and TDF.

According to the reviewed literature, mutations at rt236, rt181, and rt173 are also commonly found in the RT domain of the HBV genome, but their frequency is slightly lower than the mutations at rt204 and rt180. Mutations rtN236T, rtA181T/V/S and rtV173L were reported by 13, 11 and 10 studies respectively, included in this review (Figure 2, Table 1). Mutations rtN236T and rt181T/V/S are reported to be generally associated with resistance against ADV therapy, while rtV173L is generally associated with resistance against LAM and Ldt therapy.

Apart from the above-mentioned common mutations at positions rt204, rt180, rt236, rt181, and rt173, which are now well known for their role in HBV resistance to NAs treatment, some mutations have also been reported in many studies from different parts of the world with a possible role in viral resistance. These mutations are: rtN238D/S/R, rtS202I/G/C, rtA194T and rtM250V/L/M, which are reported in 8, 6, 6 and 6 studies respectively (Figure 2, Table 1). Mutation rtN238D/S/R is commonly found to be associated with ADV resistance, while the mutation rtS202I/G/C has been reported to have an association with ETV resistance. Mutation rtA194T is reported to have a probable role in resistance against TDF, while the mutation rtM250V/L/M is reported to have an association with resistance to all NAs (Table 1).

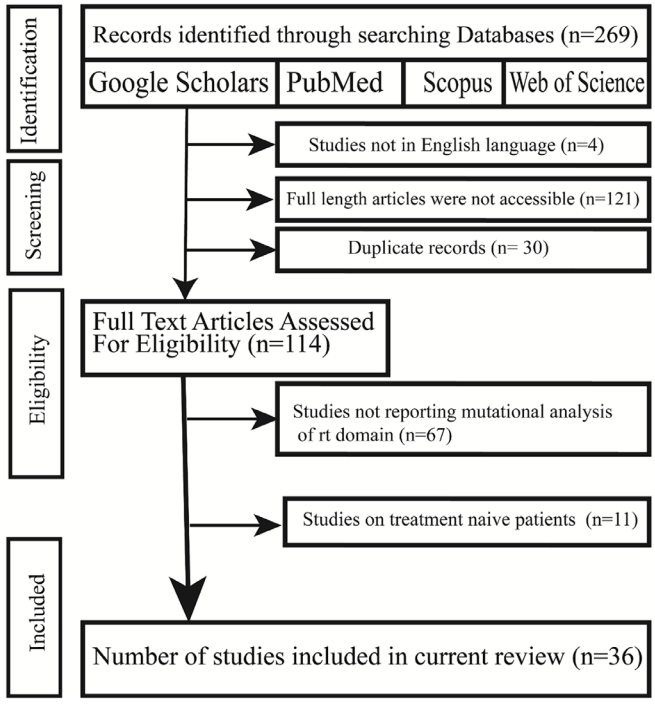

Figure 1. Flow chart of study methodology with detail of the articles included and excluded

Each of the mutations rtL80I/G and rtA184S has been reported in five studies (Figure 2) rtA184S was found to be associated with ETV resistance, while rtL80G/I is reported to be associated with

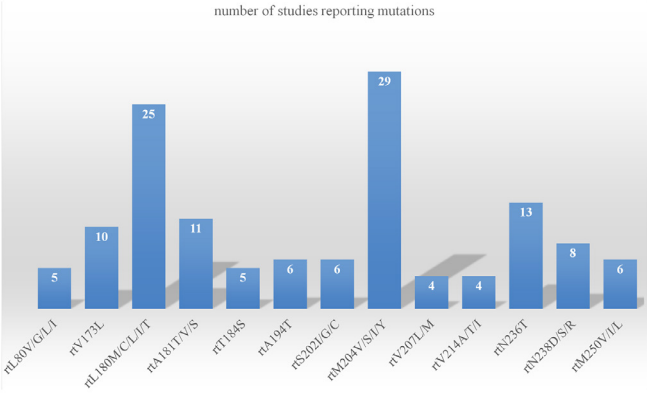

Figure 2. Detail of major resistant mutations in HBV RT domain with number of studies for each mutation reporting the particular mutation

LAM and Ldt resistance. Mutations rtV214A/T/I and rtV207L/M were each reported in four studies (Figure 2). these mutations are thought to have some association with resistance to all NAs, i.e., LAM, Ldt, adefovir, ETV, and TDF.

Overall, 13 mutations in the RT domain of the HBV polymerase region are reported in different studies from the samples of patients non-responders to different NAs (Figure 2). More commonly reported of these 13 are rtM204I/V/S/Y, rtL180M/L/C/I/T, rtN236T, rtA181T/V/S and rtV173L, which have been reported by at least 10 studies included in this review. Mutations less commonly reported include rtN238D/S/R, rtS202I/G/C, rtA194T, rtM250V/L/M, rtL80I/G, rtA184S, rtV214A/T/I and rtV207L/M, which are reported in less than 10 studies. The role of less common mutations in resistance is still to be established completely.

NAs used for the treatment of chronic HBV are not equally resistant to the virus. Some of these drugs have more mutations to resist while some have much fewer. A brief review of each is given below.

Table 1. List of studies included in the review along with the reported mutations associated to be resistance against NAs

Study reference	Country/state	Sample size	Drug	Non responder percentage	RT mutations reported
Westland et al. (22)	California	467	ADV	14%	S119A, H133L, V214A/T/I, H234Q
Angus et al. (26)	Australia	1	ADV	100%	N236T, N238D
Delaney et al. (23)	New Jersey	216	ADV	22%	V173L, L180M,204V
Tenney et al. (27)	Australia	500	ETV	NR	L18M, S78T, I169T, V173L, L180M, T184S/G, S202I, M204V, M250V
Pai et al. (34)	Turkey	1	LAM	100%	L180C, M204I
Marrone et al. (37)	Italy	14	LAM	57%	L180M, M204V/I,
Burnelle et al. (24)	California	NR	LAM, ADV	NR	L180 M, M204V, N236T
Sheldon et al. (31)	Germany	43	TDF	53%	A194T, L180M, M204V
Colonno et al. (25)	Wallingford CT	673	ETV, LAM	16%	L180M/L/I, S202G, M204I/V
Warner et al. (28)	Australia	1376	LAM	37%	L180M, M204I/V
Libberecht et al. (40)	Hong Kong	80	LAM	25%	L80V/I, V173L, L180M, M204V/I,
Malmström et al. (38)	Sweden	5	LAM	100%	L180M, M204I/V
Cassino et al. (39)	Argentina	1	ETV, ADV, TDF, LAM	100%	L180M, M204V
Qin et al. (8)	China	5	ADV	60%	M204Y, N238R, N248H
Lei et al. (9)	China	179	LAM, ADV, LdT	89%	M204I, L80I/V, L180M, N236T
Jiang et al. (10)	China	79	ADV	63.29%	L180M, A181T, T184L, M204V, 207M/L/I, S213T, V214A, Q215S, N236T, P237H, N238T, M250L
Wang et al. (11)	China	3	ADV	100%	A181V, N236T,
Hua et al. (12)	China	620	LAM, ADV, LdT	2.7%	L180M, A181T, 204I/V, N236T
Yin et al. (13)	China	26	LdT	61%	L180I/V, M20I
Kim et al. (29)	Korea	83	ADV, LAM	NR	L180M, A181T, 204I/V, N236T
He et al. (14)	China	84	LAM, LdT, ADV, ETV	52.94%	V173L, L180M/I, A181T, M204I/V, V207M, S213T, V214A, N236A/T, N238T
Motahar et al. (33)	Iran	64	TDF	20%	A194T
Zhao et al. (15)	China	269	LAM, LdT, ADV	8.9%	L180M, T184G, S202I, 204V/I, M207I, N236T, M250V
Qian et al. (16)	China	139	LAM, LdT, ADV, ETV	41.7%	L80I/V, V173L, L180M/I, A181V/G, A194G, S202N, 204I/V, N236T, M250L/V

Table 1. Continued

Study reference	Country/state	Sample size	Drug	Non responder percentage	RT mutations reported
Fan et al. (17)	China	300	LAM, TDF, LdT, ADV, ETV	51.7%	L80V, V173L, V191I, A194T/S, L199V, A200V/S, V207L/M/I, A211S, K212R, V214A L220I/N, Y221H/S, A222S, L228I, I235I, P237H, N238T, T240I, R242S
Zhang et al. (18)	China	46	ADV, ETV	8.7%	R15Q, D134N, L145M/S, F151Y/L, P177G, A181T/V, A194T, M204I/V, S223A, N236T, F249A, D263E
Shirvani-Dastgerdi et al. (32)	Germany	2	TDF, ETV	100%	S78T
Mahmood and Anwar (41)	Pakistan	20	LAM, LdT, ETV, ADV	65%	L80G, Y135, I169P, V173L, L180M, A181V, T184Y, M204V, N248H
Yamada et al. (1)	Japan	70	ETV	50%	V173L, L180M, M204, N238H, L269I
Marhoon et al. (35)	Iraq	20	LAM, ETV, ADV, LdT, TDF	NR	L80I/V, V173L, L180M, A181S, A194T, S202I, M204V/I, N236T, M250L/V
Jiang et al. (19)	China	1	LAM, ETV, ADV	100%	L180M, T184L, A200V, M204V
Arikan et al. (36)	Cyprus	100	LAM, ETV, ADV	37%	L91I, Q149K, V173M, Q215H/P/S, N238D
Choe et al. (2)	Korea	232	ETV, TDF	16.7%	M204I
Zhang et al. (20)	China	435	ADV, LAM, ETV, TDF	54.7%	L180M, A181T, T184S M204I/V, I224V, N238H, F221Y
Hong-tao et al. (21)	China	406	ADV, LAM, ETV	NR	L180M, A181S, S202I, M204V/I, N236T, M250L/V, V207L/M/I
Park et al. (30)	Korea	2	TDF	100%	S106C, H126Y, D134E, D204I/V, L269I

RT: Nucleot(s)ide analogues, RT: Reverse transcriptase, NR: Not reached, ETV: Entecavir, TDF: Tenofovir, LAM: Lamivudine, ADV: Adefovir, Ldt: Telbivudine

LAM Associated Resistance

LAM was the first NA approved for oral use in 1998 for chronic hepatitis B (CHB) at a dosage of 100 mg/day. It was found to be effective in the recovery of CHB complications by inhibiting the activity of the RT enzyme, adding deoxycytidine triphosphate to the growing DNA chain, during viral replication. However, the major drawback associated with LAM was treatment resistance in patients, which was attributed to some mutations in the rt domain of the virus (4,6). The current review included 21 studies published from different parts of the world with different numbers of patients reporting mutations associated with LAM therapy (Table 1). The most frequent mutations reported to be associated with viral resistance against LAM are rtL180M and rtM204I/V (Table 1). Moreover, the mutations rtL80I/V and rtV173L are also reported to be associated with LAM resistance. In 2005, three studies were published reporting the mutations rtL180M and rtM204I/V from the patients having no response against LAM from Turkey (35), Italy (37), and California (24). In 2006 and 2007, four studies, including two with a larger number of patients, reported mutations rtL180M and rtM204I/V in the patients who were resistant to LAM (25,28,38,40). Additionally, the mutations rtL80I/V and rtV173L were also found in one of these studies 40 from the patients resistant to LAM therapy (Table 1). In 2011, mutations rtL180M and rtM204I/V were identified in a patient who was a non-responder to LAM, from Argentina (39). In 2013, mutations rtL80I/V, rtL180M, and rtM204I/V were found in another non-responder patient against LAM, along with another mutation rtN236T from patients who had developed resistance to Ldt and ETV (9). In 2015, three studies reported the mutations rtL180M, rtM204I/V and rtV173L from the patients having no response against LAM therapy from China (12,14), and Korea (29) while the mutations rtA181T, rtV207M, rtS213T, rtV214A, rtN236A/T and rtN238T were also found from the patients resistant against Ldt, ADV and ETV (Table 1). In 2016, three studies reporting mutations rtL80I/V, rtL180M, rt204I/V, and rtV173L from patients not responding to LAM treatment were published from China (15,16,17). Moreover, mutations rtV191I, rtA194T/S, rtL199V, rtA200V/S, rtS202N, rtV207L/M/I, rtA211S, rtK212R, rtV214A, rtL220I/N, rtY221H/S, rtA222S, rtL228I, rtL235I, rtP237H, rtN238T, rtT240I, rtR242S, rtN236T, rtA181V/G were also reported in these studies associated with adefovir, ETV, Ldt and TDF resistance (Table 1). In 2017, resistant mutations rtL80I/V, rtL180M, rt204I/V, and rtV173L were reported from the patients not responding to LAM therapy in a study from Pakistan (41). Additionally, the mutations rtY135, rtI169P, rtA181V, rtT184Y, and rtN248H were also reported in patients resistant to Ldt, adefovir, and ETV (Table 1). In 2018, a study from Iraq reported mutations rtL80I/V, rtV173L, rtL180M and rtM204V/I in patients treated with LAM who did not respond (34). In 2019, two studies from China (19,20) and one from Cyprus (36) reported mutations rtL180M, rtM204I/V and rtV173L with association to resistance against LAM. Some other mutations like rtL91I, rtQ149K, rtA181T, rtT184S, rtA200V, rtQ215H/P/S, rtN238D, rtI224V, rtN238H, and rtF221Y were also found in these studies associated with resistance to ETV, adefovir, and TDF (Table 1).

ADV Associated Resistance

ADV is an adenosine monophosphate NA, approved in 2003 for treating CHB at a dose of 10 mg/day. It was proved to significantly improve histological, virological, and biochemical parameters, and it was also effective against patients resistant to LAM (6). ADV inhibits elongation of the viral DNA strand by inhibiting the addition of deoxyadenosine triphosphate to the viral primer. Thus, the reverse transcription of the virus is stopped (4). However, soon

after its approval as a improved therapy for chronic HBV patients, reports of resistant mutations against it began to emerge. The current review included 23 studies reporting mutations associated with ADV therapy (Table 1). The mutations rtN236T/A, rtN238T, and rtA181T were most commonly reported to be associated with ADV resistance. Other putative mutations reported to be associated with ADV resistance were rtS119A, rtH133L, rtH234Q, rtN248H, and rtS213T (Table 1). In 2003, three studies including two with larger sample size reported the mutations rtN236T, rtN238D, rtS119A, rtH133L, rtV214A/T/I, rtH234Q, rtV173L, rtL180M and rtM204V in the patients having no response against ADV from California (22), Australia (26), and New Jersey (23). In 2005, rtN236T was detected in a patient from California, who was a non-responder against ADV (24). Additionally, the mutations rtL180M and rtM204V were found in association with LAM resistance (Table 1). In 2011, resistant mutations rtL180M and rtM204V were reported in patients from Argentina who were non-responders to ADV along with other drugs (39). In 2013, two studies from China reported the resistant mutations rtN236T, rtN238, and rtN248H from non-responder patients against ADV therapy (8,9). Additionally, the mutations rtM204I, rtL80I/V and rtL180M, associated with LAM and Ldt resistance, were also reported in the studies. Mutations rtL180M, rtA181T, rtT184L, rtM204V, rt207M/L/I, rtS213T, rtV214A, rtQ215S, rtN236T, rtP237H, rtN238T, and rtM250L were detected in a non-responder patient from China against Adefovir, in 2014 (10). In 2015, four studies reported the mutations rtA181T, rtS213T, rtV214A, rtN236A/T and rtN238T in the patients having no response against ADV therapy from China (11,12,14), and Korea (29) while the mutations rtV173L, rtL180M and rtM204I/V were also detected in the study from the patients non-responder to LAM, ETV and Ldt (Table 1). In 2016, four studies were published from China reporting mutations rtI235I, rtP237H, rtN238T, rtT240I, rtR242S, rtN236T, and rtA181V/G in patients who were non-responders to ADV (Table 1) (15,16,17,18). In 2017 and 2018, resistant mutations rtA181V, rtN236T, rtM250L/V and rtN238T were reported against ADV from Pakistan (41), and Iraq (34). In 2019, four studies including three from China (19,20,21), and one from Cyprus (36) reported the mutations rtA181T, rtV207L/M/I, rtN238D, rtI224V and rtN238H, from the patients non-responder against ADV therapy (Table 1). In conclusion, the most frequently reported mutations from patients unresponsive to ADV therapy are present on positions rt238, rt236, and rt181, which have been found in many studies from different parts of the world.

ETV Associated Resistance

ETV is a carboxylic analogue of guanosine and was approved in 2005 with a dosage of 0.5 mg/day and 1 mg/day for treatment naïve and LAM resistant CHB patients, respectively. Inside the cell, phosphorylation occurs at its active 5'triphosphate metabolite. ETV inhibits HBV-DNA polymerase by competing with its natural substrate guanosine triphosphate (6). This review included 16 studies published in different parts of the world, with different numbers of patients reporting mutations associated with ETV resistance (Table 1). The most frequently associated with resistance against ETV are rtI169P/T, rtT184S/G/F/C/A/L/M/S, and rtS202I/G/C. Moreover, the mutations rtS78T, rtP177G and rtD263E were also reported by some studies to have an association with ETV resistance (Table 1). In 2004, a study from Australia reported mutations rtL18M, rtS78T, rtI169T, rtV173L, rtL180M, rtT184S/G, rtS202I, rtM204V, and rtM250V from a patient who was a non-responder to ETV therapy (27). This was the first report on resistance against ETV. In 2006, another study, conducted in Wallingford CT on 673 patients, reported the mutation rtS202I associated with ETV resistance mutations (Table 1) (25). In 2011, resistant mutations rtL180M and rtM204I/V were also reported from patients who were non-responders against ETV, in a study from Argentina (39). In 2015, a study from China on 84 patients reported the mutation rtV207M and rtS213T to be associated with ETV resistance (14). Three more studies were published from China in 2016 on mutations associated with ETV resistance (16,17,18). These studies found rtS202N, rtP177G, and rtD263E from the patients having no response to ETV therapy (Table 1). Two studies in 2017, including one with a larger number of patients, reported mutations rtV173L, rtL180M, rtI169P, rtT184Y, rtM204, rtN238H and rtL269I in the patients who were resistant to ETV (1,41). Five studies, including three from China (19,20,21), one from Cyprus (36), and one from Korea (2) were published in 2019 on mutations from patients having no response to ETV. These studies reported mutations rtQ215H/P/S, rtT184S, rtA200V, rtS202I, and rtM250L/V associated with ETV resistance (Table 1).

Ldt Associated resistance

Ldt (L-deoxythymidine) is a NA of L-thymidine approved in 2006 for the treatment of CHB patients at the dosage of 600 mg/day. The stoichiometric structure of L-nucleoside is different from natural nucleoside, as the sugars and base moieties are arranged in L configuration rather than the D configuration. It inhibits the synthesis of the positive strand of HBV-DNA by competing with thymidine 5'-triphosphate. With the incorporation of Ldt , the nascent chain of HBV-DNA is terminated (4,6). This review included 9 studies published from different parts of the world with different numbers of patients reporting the mutations associated with Ldt (Table 1). The most frequent mutation reported to have an association with resistance against Ldt is rtM204V/I, with or without rtL80I/V and rtL180M. Moreover, the mutations rtV173L were also reported to have an association with Ldt resistance (Table 1). In 2013, a study from China (9) reported the mutations rtL180M and rtM204I/V from the patients having no response against Ldt therapy. Moreover, three studies from China in 2015 also reported the mutations rtV173L, rtL180M, and rtM204I/V in the patients having no response to Ldt therapy (Table 1) (12,13,14). In the next year (2016), three more studies were published from China who reported mutations rtL80I/V, rtL180M, rt204I/V and rtV173L from the patients non-responders to Ldt confirming the role of these mutations in Ldt resistance (15,16,17). Same mutations (rtL80I/V, rtL180M, rt204I/V and rtV173L) were again reported in 2017 from Pakistan (41), and in 2018 from Iraq (34) in the patients not responding to Ldt (Table 1). The role of mutations rtL80I/V, rtL180M, rt204I/V, and rtV173L in Ldt-associated resistance has been confirmed by many reports.

TDF Associated Resistance

TDF is an acyclic NA {9-[(R)-2-(phosphonomethoxy)-propyl] adenine} which undergoes phosphorylation to competitively inhibit the natural substrate deoxyadenosine 5'triphosphate. TDF has been available since 2002 for HIV treatment. It was approved for

the treatment of CHB in 2008 at the dosage of 300 mg/day. It was found to be superior to other NAs in terms of suppression of HBV-DNA and normalization of alanine transaminase (4,6). Few reports exist on mutations associated with TDF resistance. The current review included 9 studies reporting mutations possibly associated with TDF therapy (Table 1). Three mutations are frequently reported to have an association with resistance against TDF. These are: rtA194T, rtS78T and rtM204V/I. Moreover, the mutations rtS106C, rtH126Y, rtD134E, rtD204I/V, rtL269I, were also suspected to have a role in TDF resistance (Table 1). In 2005, a study reported the mutations rtA194T, rtL180M, and rtM204V in the patients who showed no response to TDF therapy, from Germany (31). In 2011, a study from Argentina reported that mutations rtL180M and rtM204V also have some association with TDF resistance in addition to their association with resistance against LAM, ETV, and ADV (Table 1) (39). Mutation rtA194T was also found in non-responder patients against TDF in Iran in 2016 (33). In 2017, another study from Germany on two patients reported that mutation rtS78T is associated with resistance against TDF (32). A study from Iraq in 2018 reported the mutations rtA194T and rtM204V/I in non-responder patients against TDF (34). Three more studies were published in 2019, reporting mutations that may have some association with TDF resistance. One of these studies from Korea (2) reported that the mutation rtM204I is associated with both TDF and ETV resistance, while the second study from Korea (30) reported the mutations rtS106C, rtH126Y, rtD134E, rtD204I/V and rtL269I from patients who were non-responders for TDF. The third study in 2019 reported the mutations rtL180M, rtA181T, rtT184S, rtM204I/V, rtI224V, rtN238H and rtF221Y from the patient non-responder of a patient from China against adefovir, LAM, ETV and TDF. It was suggested in the study that any of these mutations might also have a role in resistance against TDF (Table 1). The exact mutations conferring resistance against TDF therapy are, however, still to be confirmed.

Conclusion

The most frequent mutation associated with viral resistance was found to be rtM204I/V/S/Y and then the mutation rtL180M/L/C/I/T, both of which are considered to be associated with almost all of the NAs but primarily associated with LAM and Ldt resistance. Mutations rtN236T, rtA181T/V/S, and rtV173L are also commonly found in the RT domain of the HBV genome with a slightly lower frequency than the mutations at rt204 and rt180. Mutations rtN236T and rt181T/V/S are associated with ADV resistance, while rtV173L is generally associated with resistance against LAM and Ldt therapy. Relatively less common mutations are rtN238D/S/R, rtS202I/G/C, rtA194T, and rtM250V/L/M. Of these, the first three are associated with adefovir, ETV and TDF resistance respectively, while the lattermost is thought to be associated with resistance to all NAs. Mutations rtL80I/G, rtA184S, rtV214A/T/I, and rtV207L/M were also reported in a smaller number of studies with their association with different NAs.

Footnotes

Authorship Contributions

Concept: M.M., Design: M.M., Data Collection or Processing: Z.U.R., Analysis or Interpretation: M.M., Literature Search: Z.U.R., Writing: Z.U.R., M.M.

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References

1. Yamada N, Sugiyama R, Nitta S, Murayama A, Kobayashi M, Okuse C, Suzuki M, Yasuda K, Yotsuyanagi H, Moriya K, Koike K, Wakita T, Kato T. Resistance mutations of hepatitis B virus in entecavir-refractory patients. Hepatol Commun. 2017;1:110-121.
2. Choe WH, Kim K, Lee SY, Choi YM, Kwon SY, Kim JH, Kim BJ. Tenofovir is a more suitable treatment than entecavir for chronic hepatitis B patients carrying naturally occurring rtM204I mutations. World J Gastroenterol. 2019;25:4985-4998.
3. Tan M, Bhadoria AS, Cui F, Tan A, Van Holten J, Easterbrook P, Ford N, Han Q, Lu Y, Bulterys M, Hutin Y. Estimating the proportion of people with chronic hepatitis B virus infection eligible for hepatitis B antiviral treatment worldwide: a systematic review and meta-analysis. Lancet Gastroenterol Hepatol. 2021;6:106-119.
4. Keeffe EB, Dieterich DT, Pawlotsky JM, Benhamou Y. Chronic hepatitis B: preventing, detecting, and managing viral resistance. Clin Gastroenterol Hepatol. 2008;6:268-274.
5. Lim YS. Management of antiviral resistance in chronic hepatitis B. Gut Liver. 2017;11:189-195.
6. Fung J, Lai CL, Seto WK, Yuen MF. Nucleoside/nucleotide analogues in the treatment of chronic hepatitis B. J Antimicrob Chemother. 2011;66:2715-2725.
7. Stuyver LJ, Locarnini SA, Lok A, Richman DD, Carman WF, Dienstag JL, Schinazi RF. Nomenclature for antiviral-resistant human hepatitis B virus mutations in the polymerase region. Hepatology. 2001;33:751-757.
8. Qin B, Pei RJ, HE TT, Huang ZH, Pan GS, Tu CY, Lu MJ, Chen XW. Polymerase mutations rtN238R, rtT240Y and rtN248H of hepatitis B virus decrease susceptibility to adefovir. Chin. Sci. Bull. 2013;58:1760-1766.
9. Lei J, Wang Y, Wang LL, Zhang SJ, Chen W, Bai ZG, Xu LY. Profile of hepatitis B virus resistance mutations against nucleoside/nucleotide analogue treatment in Chinese patients with chronic hepatitis B. Virol J. 2013;10:313.
10. Jiang SW, Yao LP, Hu AR, Hu YR, Chen SX, Xiong T, Gao GS, Liang XY, Ding SX, Weng PJ. Resistant mutants induced by adefovir dipivoxil in hepatitis B virus isolates. World J Gastroenterol. 2014;20:17100-17106.
11. Wang Y, Liu S, Chen Y, Zheng S, Zhou L, Lu F, Duan Z. Delayed reduction of hepatitis B viral load and dynamics of adefovir-resistant variants during adefovir plus entecavir combination rescue therapy. Int J Med Sci. 2015;12:416-422.
12. Hua W, Zhang G, Guo S, Li W, Sun L, Xiang G. Microarray-based genotyping and detection of drug-resistant HBV mutations from 620 Chinese patients with chronic HBV infection. Braz J Infect Dis. 2015;19:291-295.
13. Yin F, Wu Z, Fang W, Wu C, Rayner S, Han M, Deng F, Du R, Liu J, Wang M, Wang H, Ning Q, Hu Z. Resistant mutations and quasispecies complexity of hepatitis B virus during telbivudine treatment. J Gen Virol. 2015;96:3302-3312.
14. He X, Wang F, Huang B, Chen P, Zhong L. Detection and analysis of resistance mutations of hepatitis B virus. Int J Clin Exp Med. 2015;8:9630-9639.
15. Zhao Y, Wu J, Sun L, Liu G, Li B, Zheng Y, Li X, Tao J. Prevalence of mutations in HBV DNA polymerase gene associated with nucleos(t)ide resistance in treatment-naive patients with Chronic Hepatitis B in Central China. Braz J Infect Dis. 2016;20:173-178.
16. Qian F, Qin J, Li D, Ma Z, Zhang H, Jin F, Wang W. Monitoring of genotypic resistance profile in chronic hepatitis B patients receiving nucleos(t)ide analogues in Huzhou, China. J Infect Dev Ctries. 2016;10:996-1002.

17. Fan J, Zhang Y, Xiong H, Wang Y, Guo X. Nucleotide analogue-resistant mutations in hepatitis B viral genomes found in hepatitis B patients. J Gen Virol. 2015;96:663-670.
18. Zhang X, Li M, Xi H, Zhang R, Chen J, Zhang Y, Xu X. Pre-existing mutations related to tenofovir in chronic hepatitis B patients with long-term nucleos(t)ide analogue drugs treatment by ultra-deep pyrosequencing. Oncotarget. 2016;7:70264-70275.
19. Jiang D, Wang J, Zhao X, Li Y, Zhang Q, Song C, Zeng H, Wang X. Entecavir resistance mutations rtL180M/T184L/M204V combined with rtA200V lead to tenofovir resistance. Liver Int. 2020;40:83-91.
20. Zhang X, Chen X, Wei M, Zhang C, Xu T, Liu L, Xu Z. Potential resistant mutations within HBV reverse transcriptase sequences in nucleos(t) ide analogues-experienced patients with hepatitis B virus infection. Sci Rep. 2019;9:8078.
21. Hong-tao C, Gui-rong H, Zhi Y, Run-zhang M, Shi-pin W. Investigation into drug-resistant mutations in the treatment of chronic hepatitis B with nucleos(t)ide analogues. Emerg. Infect. Dis. 2019;4:24-27. Available from: http://www.xfcrb2020.com/EN/abstract/abstract189.shtml
22. Westland CE, Yang H, Delaney WE 4th, Gibbs CS, Miller MD, Wulfsohn M, Fry J, Brosgart CL, Xiong S; 437 and 438 Study Teams. Week 48 resistance surveillance in two phase 3 clinical studies of adefovir dipivoxil for chronic hepatitis B. Hepatology. 2003;38:96-103.
23. Delaney WE 4th, Yang H, Westland CE, Das K, Arnold E, Gibbs CS, Miller MD, Xiong S. The hepatitis B virus polymerase mutation rtV173L is selected during lamivudine therapy and enhances viral replication in vitro. J Virol. 2003;77:11833-11841.
24. Brunelle MN, Jacquard AC, Pichoud C, Durantel D, Carrouée-Durantel S, Villeneuve JP, Trépo C, Zoulim F. Susceptibility to antivirals of a human HBV strain with mutations conferring resistance to both lamivudine and adefovir. Hepatology. 2005;41:1391-1398.
25. Colonno RJ, Rose R, Baldick CJ, Levine S, Pokornowski K, Yu CF, Walsh A, Fang J, Hsu M, Mazzucco C, Eggers B, Zhang S, Plym M, Klesczewski K, Tenney DJ. Entecavir resistance is rare in nucleoside naïve patients with hepatitis B. Hepatology. 2006;44:1656-1665.
26. Angus P, Vaughan R, Xiong S, Yang H, Delaney W, Gibbs C, Brosgart C, Colledge D, Edwards R, Ayres A, Bartholomeusz A, Locarnini S. Resistance to adefovir dipivoxil therapy associated with the selection of a novel mutation in the HBV polymerase. Gastroenterology. 2003;125:292-297.
27. Tenney DJ, Levine SM, Rose RE, Walsh AW, Weinheimer SP, Discotto L, Plym M, Pokornowski K, Yu CF, Angus P, Ayres A, Bartholomeusz A, Sievert W, Thompson G, Warner N, Locarnini S, Colonno RJ. Clinical emergence of entecavir-resistant hepatitis B virus requires additional substitutions in virus already resistant to Lamivudine. Antimicrob Agents Chemother. 2004;48:3498-3507.
28. Warner N, Locarnini S, Kuiper M, Bartholomeusz A, Ayres A, Yuen L, Shaw T. The L80I substitution in the reverse transcriptase domain of the hepatitis B virus polymerase is associated with lamivudine resistance and enhanced viral replication in vitro. Antimicrob Agents Chemother. 2007;51:2285-2292.
29. Kim J, Lee SH, Kim HS, Choi K, Jeong SW, Kim SG, Jang JY, Kim YS, Kim BS. Long-term outcomes and dynamics of mutants associated with lamivudine-adefovir rescue therapy in patients with lamivudine-resistant chronic hepatitis B. Gut Liver. 2015;9:103-108.
30. Park ES, Lee AR, Kim DH, Lee JH, Yoo JJ, Ahn SH, Sim H, Park S, Kang HS, Won J, Ha YN, Shin GC, Kwon SY, Park YK, Choi BS, Lee YB, Jeong N, An Y, Ju YS, Yu SJ, Chae HB, Yu KS, Kim YJ, Yoon JH, Zoulim F, Kim KH. Identification of a quadruple mutation that confers tenofovir resistance in chronic hepatitis B patients. J Hepatol. 2019;70:1093-1102.
31. Sheldon J, Camino N, Rodés B, Bartholomeusz A, Kuiper M, Tacke F, Núñez M, Mauss S, Lutz T, Klausen G, Locarnini S, Soriano V. Selection of hepatitis B virus polymerase mutations in HIV-coinfected patients treated with tenofovir. Antivir Ther. 2005;10:727-734.
32. Shirvani-Dastgerdi E, Winer BY, Celià-Terrassa T, Kang Y, Tabernero D, Yagmur E, Rodríguez-Frías F, Gregori J, Luedde T, Trautwein C, Ploss A, Tacke F. Selection of the highly replicative and partially multidrug resistant rtS78T HBV polymerase mutation during TDF-ETV combination therapy. J Hepatol. 2017;67:246-254.
33. Motahar M, Arabzadeh SAM, Mollaei H, Iranmanesh Z, Nikpour N, Soleimani F. Evaluation of HBV resistance to tenofovir in patients with chronic hepatitis B using ZNA probe assay in Kerman, southeast of Iran. Asian Pac J Trop Dis. 2016;6:513-516.
34. Pai SB, Bozdayi AM, Pai RB, Beker T, Sarioglu M, Turkyilmaz AR, Grier J, Yurdaydin C, Schinazi RF. Emergence of a novel mutation in the FLLA region of hepatitis B virus during lamivudine therapy. Antimicrob Agents Chemother. 2005;49:2618-2624.
35. Marhoon AA, Altaai MI, Ahmed AM. Drug-resistance associated mutations in polymerase (Pol) gene of hepatitis b virus isolated from Iraqi chronic hepatitis B patients (CHB). J. Pharm. Sci. & Res. 2018;10:1041-1044.
36. Arikan A, Sayan M, Sanlidag T, Suer K, Akcali S, Guvenir M. Evaluation of the pol/S gene overlapping muta tions in chronic hepatitis B patients in Northern Cyprus. Pol J Microbiol. 2019;68:317-322.
37. Marrone A, Zampino R, Karayannis P, Cirillo G, Cesaro G, Guerrera B, Ricciotti R, del Giudice EM, Utili R, Adinolfi LE, Ruggiero G. Clinical reactivation during lamivudine treatment correlates with mutations in the precore/core promoter and polymerase regions of hepatitis B virus in patients with anti-hepatitis B e-positive chronic hepatitis. Aliment Pharmacol Ther. 2005;22:707-714.
38. Malmström S, Hannoun C, Lindh M. Mutation analysis of lamivudine resistant hepatitis B virus strains by TaqMan PCR. J Virol Methods. 2007;143:147-152.
39. Cassino L, Benetti S, Fay F, Tanno H, Quarleri J. Unsuccessful therapy with adefovir and entecavir-tenofovir in a patient with chronic hepatitis B infection with previous resistance to lamivudine: a fourteen-year evolution of hepatitis B virus mutations. BMC Infect Dis. 2011;11:178.
40. Libbrecht E, Doutreloigne J, Van De Velde H, Yuen MF, Lai CL, Shapiro F, Sablon E. Evolution of primary and compensatory lamivudine resistance mutations in chronic hepatitis B virus-infected patients during long-term lamivudine treatment, assessed by a line probe assay. J Clin Microbiol. 2007;45:3935-3941.
41. Mahmood M, Anwar MA. Analysis of resistant mutations in reverse transcriptase domain of hepatitis B virus from patients from Islamabad, Pakistan. J Unexplored Medical Data. 2017;2:60-64.