MUTATIONS IN THE REVERSE TRANSCRIPTASE GENE ASSOCIATED WITH RESISTANCE TO REVERSE TRANSCRIPTASE INHIBITORS

Nucleoside and Nucleotide Analogue Reverse Transcriptase Inhibitors (nRTIs)[a]

Multi-nRTI Resistance: 69 Insertion Complex[b] (affects all nRTIs currently approved by the US FDA)

M	A	▼	K	L	T	K
41	**62**	**69**	**70**	**210**	**215**	**219**
L	**V**	**Insert**	**R**	**W**	**Y**	**Q**
					F	**E**

Multi-nRTI Resistance: 151 Complex[c] (affects all nRTIs currently approved by the US FDA except tenofovir)

A	V	F	F	Q
62	**75**	**77**	**116**	**151**
V	**I**	**L**	**Y**	**M**

Multi-nRTI Resistance: Thymidine Analogue-Associated Mutations[d,e] (TAMs; affect all nRTIs currently approved by the US FDA)

M	D	K	L	T	K
41	**67**	**70**	**210**	**215**	**219**
L	**N**	**R**	**W**	**Y**	**Q**
				F	**E**

	K	L	Y	M
Abacavir[f,g]	**65**	**74**	**115**	**184**
	R	**V**	**F**	**V**
	E			
	N			

	K	L
Didanosine[g,h]	**65**	**74**
	R	**V**
	E	
	N	

	K	M
Emtricitabine	**65**	**184**
	R	**V**
	E	**I**
	N	

	K	M
Lamivudine	**65**	**184**
	R	**V**
	E	**I**
	N	

	M	K	D	K	L	T	K
Stavudine[d,e,g,i,j,k]	**41**	**65**	**67**	**70**	**210**	**215**	**219**
	L	**R**	**N**	**R**	**W**	**Y**	**Q**
		E				**F**	**E**
		N					

	K	K
Tenofovir[l]	**65**	**70**
	R	**E**
	E	
	N	

	M	D	K	L	T	K
Zidovudine[d,e,j,k]	**41**	**67**	**70**	**210**	**215**	**219**
	L	**N**	**R**	**W**	**Y**	**Q**
					F	**E**

Nonnucleoside Analogue Reverse Transcriptase Inhibitors (NNRTIs)[a,m]

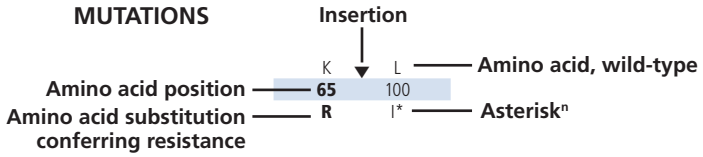
	L	K	K	V	V	Y	Y	G	P	M
Efavirenz	**100**	**101**	**103**	**106**	**108**	**181**	**188**	**190**	**225**	**230**
	I	**P**	**N**	**M**	**I**	**C**	**L**	**S**	**H**	**L**
			S			**I**		**A**		

	V	A	L	K	V	E	V	Y	G	M
Etravirine[n]	90	98	**100**	**101**	106	138	179	**181**	190	230
	I	G	**I***	E	I	A	D	**C***	S	L
				H		G	F	**I***	A	
				P*		K	T	**V***		
						Q				

	L	K	K	V	V	Y	Y	G	M
Nevirapine	**100**	**101**	**103**	**106**	**108**	**181**	**188**	**190**	**230**
	I	**P**	**N**	**A**	**I**	**C**	**C**	**A**	**L**
			S	**M**		**I**	**L**		
							H		

	L	K	E	V	Y	Y	H	F	M
Rilpivirine[o]	**100**	**101**	**138**	**179**	**181**	**188**	**221**	**227**	**230**
	I	**E**	**A**	**L**	**C**	**L**	**Y**	**C**	**I**
		P	**G**		**I**				**L**
			K		**V**				
			Q						
			R						

Amino acid abbreviations: A, alanine; C, cysteine; D, aspartate; E, glutamate; F, phenylalanine; G, glycine; H, histidine; I, isoleucine; K, lysine; L, leucine; M, methionine; N, asparagine; P, proline; Q, glutamine; R, arginine; S, serine; T, threonine; V, valine; W, tryptophan; Y, tyrosine.



MUTATIONS IN THE PROTEASE GENE ASSOCIATED WITH RESISTANCE TO PROTEASE INHIBITORS[p,q,r]

	L	G	K	L	V	L	E	M	M	G	I	F	I	D	I	I	A	G	V	I	I	N	L	I
Atazanavir +/- ritonavir[s]	10	16	20	24	32	33	34	36	46	48	**50**	53	54	60	62	64	71	73	82	**84**	85	**88**	90	93
	I	E	R	I	I	I	Q	I	I	V	L	L	L	E	V	I	V	C	A	V	V	S	M	L
	F		M			F		L	L			Y	V			L	I	S	T					M
	V		I			V		V					M			M	T	T	F					
	C		T										T			V	L	A	I					
			V										A											
	V				V	L			I		I		I				T	L		I		L		
Darunavir/ ritonavir[t]	11				32	33			**47**		**50**		**54**				74	**76**		**84**		89		
	I				I	F			V		V		M				P	V		V		V		
													L											
	L				V				M	I	I		I					G	L	V			L	
Fosamprenavir/ ritonavir	10				32				46	47	**50**		54					73	76	82			90	
	F				I				I	V	V		L					S	V	**84**			M	
	I								L				V							V				
	R												M							A				
	V																			F				
																				S				
																				T				
	L		K	L	V			M	M				I				A	G	L	V	V	I		L
Indinavir/ ritonavir[u]	10		20	24	32			36	**46**				54				71	73	76	77	**82**	**84**		90
	I		M	I	I			I	I				V				V	S	V	I	A	V		M
	R		R						L								T	A			F			
	V																				T			
	L		K	L	V	L			M	I	I	F	I			L	A	G	L		V	I		L
Lopinavir/ ritonavir[v]	10		20	24	**32**	33			46	**47**	50	53	54			63	71	73	**76**		**82**	84		90
	F		M	I	I	F			I	V	V	L	V			P	V	S	V		A	V		M
	I		R						L	A			L				T				F			
	R												A								T			
	V												M								S			
													T											
	L				D			M	M								A			V	V	I	N	L
Nelfinavir[u,w]	10				**30**			36	46								71			77	82	84	88	**90**
	F				N			I	I								V			I	A	V	D	M
	I								L								T				F		S	
																					T			
																					S			
	L			L						G			I		I		A	G		V	V	I		L
Saquinavir/ ritonavir[u]	10			24						**48**			54		62		71	73		77	82	84		**90**
	I			I						V			V		V		V	S		I	A	V		M
	R												L				T				F			
	V																				T			
																					S			
	L					L		M		K	M	I			I	Q		H		T		V	N	I		L	
Tipranavir/ ritonavir	10					33		36		43	46	**47**			54	**58**		69		**74**		**82**	**83**	**84**		89	
	V					F		I		T	L	V			A	E		K		P		L	D	V		I	
								L							M			R				T				M	
								V							V											V	

MUTATIONS IN THE ENVELOPE GENE ASSOCIATED WITH RESISTANCE TO ENTRY INHIBITORS

	G	I	V	Q	Q	N	N
Enfuvirtide[x]	**36**	**37**	**38**	**39**	**40**	**42**	**43**
	D	**V**	**A**	**R**	**H**	**T**	**D**
	S		**M**				
			E				
Maraviroc[y]	See User Note						

MUTATIONS IN THE INTEGRASE GENE ASSOCIATED WITH RESISTANCE TO INTEGRASE STRAND TRANSFER INHIBITORS[z]

	F	E	G	Q	N	R
Dolutegravir[aa]	121	138	140	**148**	155	263
	Y	A	A	**H**	H	K
		K	S	**R**		

	T	E	T	F	S	Q	N	R
Elvitegravir[bb]	**66**	**92**	97	**121**	**147**	**148**	**155**	263
	I	**Q**	A	**Y**	**G**	**H**	**H**	K
	A	G				**K**		
	K					**R**		

	L	E	T	F	E	G	Y	Q	N	R
Raltegravir[cc]	74	92	97	**121**	138	140	**143**	**148**	**155**	263
	M	Q	A	**Y**	A	A	**R**	**H**	**H**	K
					K	S	**H**	**K**		
							C	**R**		

User Notes

a. Some nucleoside (or nucleotide) analogue reverse transcriptase inhibitor (nRTI) mutations, like T215Y and H208Y,[1] may lead to viral hypersusceptibility to nonnucleoside analogue reverse transcriptase inhibitors (NNRTIs), including etravirine,[2] in nRTI-treated individuals. The presence of these mutations may improve subsequent virologic response to NNRTI-containing regimens (nevirapine or efavirenz) in NNRTI-naive individuals,[3-7] although no clinical data exist for improved response to etravirine in NNRTI-experienced individuals. Mutations at the C-terminal reverse transcriptase domains (amino acids 293-560) outside of regions depicted on the figure bars may prove to be important for nRTI and NNRTI HIV-1 drug resistance. The clinical relevance of these connection domain mutations arises mostly in conjunction with thymidine analogue–associated mutations (TAMs) and M184V and they have not been associated with increased rates of virologic failure of etravirine or rilpivirine in clinical trials.[8-10] K65E/N variants are increasingly reported in patients experiencing treatment failure with tenofovir, stavudine, or didanosine. K65E usually occurs in mixtures with wild type. K65N gives an approximately 4-fold decrease in susceptibility. Patient-derived viruses with K65E and site-directed mutations replicate very poorly in vitro; as such, no susceptibility testing can be performed.[11,12]

b. The 69 insertion complex consists of a substitution at codon 69 (typically T69S) and an insertion of 2 or more amino acids (S-S, S-A, S-G, or others). The 69 insertion complex is associated with resistance to all nRTIs currently approved by the US Food and Drug Administration (FDA) when present with 1 or more TAMs at codons 41, 210, or 215.[13] Some other amino acid changes from the wild-type T at codon 69 without the insertion may be associated with broad nRTI resistance.

c. Tenofovir retains activity against the Q151M complex of mutations.[13] Q151M is the most important mutation in the complex (ie, the other mutations in the complex [A62V, V75I, F77L, and F116Y] in isolation may not reflect multidrug resistance).

d. Mutations known to be selected by TAMs (ie, M41L, D67N, K70R, L210W, T215Y/F, and K219Q/E) also confer reduced susceptibility to all currently approved nRTIs.[14] The degree to which cross-resistance is observed depends on the specific mutations and number of mutations involved.[15-18]

e. Although reverse transcriptase changes associated with the E44D and V118I mutations may have an accessory role in increased resistance to nRTIs in the presence of TAMs, their clinical relevance is very limited.[19-21]

f. The M184V mutation alone does not appear to be associated with a reduced virologic response to abacavir in vivo. When associated with TAMs, M184V increases abacavir resistance.[22,23]

g. As with tenofovir, the K65R mutation may be selected by didanosine, abacavir, or stavudine (particularly in patients with nonsubtype-B clades) and is associated with decreased viral susceptibility to these drugs.[22,24,25] Data are lacking on the potential negative impact of K65R on clinical response to didanosine.

h. The presence of 3 of the following mutations—M41L, D67N, L210W, T215Y/F, K219Q/E—is associated with resistance to didanosine.[26] The presence of K70R or M184V alone does not decrease virologic response to didanosine.[27]

i. K65R is selected frequently (4%–11%) in patients with some nonsubtype-B clades for whom stavudine-containing regimens are failing in the absence of tenofovir.[28,29]

j. The presence of M184V appears to delay or prevent emergence of TAMs.[30] This effect may be overcome by an accumulation of TAMs or other mutations.

k. The T215A/C/D/E/G/H/I/L/N/S/V substitutions are revertant mutations at codon 215 that confer increased risk of virologic failure of zidovudine or stavudine in antiretroviral-naive patients.[31,32] The T215Y mutant may emerge quickly from one of these mutations in the presence of zidovudine or stavudine.[33]

l. The presence of K65R is associated with a reduced virologic response to tenofovir.[13] A reduced response also occurs in the presence of 3 or more TAMs inclusive of either M41L or L210W.[13] The presence of TAMs or combined treatment with zidovudine prevents the emergence of K65R in the presence of tenofovir.[34-36]

m. There is no evidence for the utility of efavirenz, nevirapine, or rilpivirine in patients with NNRTI resistance.[37]

n. Resistance to etravirine has been extensively studied only in the context of coadministration with ritonavir-boosted darunavir. In this context, mutations associated with virologic outcome have been assessed and their relative weights (or magnitudes of impact) assigned. In addition, phenotypic cutoff values have been calculated, and assessment of genotype-phenotype correlations from a large clinical database have determined relative importance of the various mutations. These 2 approaches are in agreement for many, but not all, mutations and weights.[38-40] Asterisks (*) are used to emphasize higher relative weights with regard to reduced susceptibility and reduced clinical response compared with other etravirine mutations.[41] The single mutations L100I*, K101P*, and Y181C*/I*/V* reduce clinical utility. The presence of K103N alone does not affect etravirine response.[42] Accumulation of several mutations results in greater reductions in susceptibility and virologic response than do single mutations.[43-45]

o. Fifteen mutations have been associated with decreased rilpivirine susceptibility (K101E/P, E138A/G/K/Q/R, V179L, Y181C/I/V, H221Y, F227C, and M230I/L).[46-48] A 16th mutation, Y188L, reduces rilpivirine susceptibility 6 fold.[48] K101P and Y181I/V reduce rilpivirine susceptibility approximately 50 fold and 15 fold, respectively, but are not commonly observed in patients receiving rilpivirine.[49-51] Mutations at position 138 (most notably 138A) may occur as natural polymorphisms, especially in non-B subtypes.[52] K101E, E138K, and Y181C, each of which reduces rilpivirine susceptibility 2.5 fold to 3 fold, occur commonly in patients receiving rilpivirine. E138K and to a lesser extent K101E usually occur in combination with the nRTI resistance–associated mutation M184I, which alone does not reduce rilpivirine susceptibility. When M184I is combined with E138K or K101E, rilpivirine susceptibility is reduced about 7 fold and 4.5 fold, respectively.[51,53-55] The combinations of reverse transcriptase–associated mutations L100I plus K103N/S and L100I plus K103R plus V179D were strongly associated with reduced susceptibility to rilpivirine. However, for isolates harboring the K103N/R/S or V179D as single mutations, no reduction in susceptibility was detected.[48,56]

p. Often, numerous mutations are necessary to substantially impact virologic response to a ritonavir-boosted protease inhibitor (PI).[57] In some specific circumstances, atazanavir might be used unboosted. In such cases, the mutations that are selected are the same as with ritonavir-boosted atazanavir, but the relative frequency of mutations may differ.

q. Resistance mutations in the protease gene are classified as "major" or "minor."

Major mutations in the protease gene (positions in bold type) are defined as those selected first in the presence of the drug or those substantially reducing drug susceptibility. These mutations tend to be the primary contact residues for drug binding.

Minor mutations generally emerge later than major mutations and by themselves do not have a substantial effect on phenotype. They may improve replication of viruses containing major mutations. Some minor mutations are present as common polymorphic changes in HIV-1 nonsubtype-B clades.

Mutations in the cytoplasmic tail of gp41 of *env* or mutations in *gag* cleavage sites may confer resistance to all PIs and may emerge before mutations in protease do.[58] A large proportion of virus samples from patients with confirmed virologic failure on a PI-containing regimen is not found to have PI resistance–associated mutations. Preliminary data from recent studies suggest that several mutations in the Gag protein[59] and in the cytoplasmic tail of the Env protein[58] may be responsible for reduced PI susceptibility in a subset of these patients.

r. Ritonavir is not listed separately, as it is currently used only at low doses as a pharmacologic booster of other PIs.

s. Many mutations are associated with atazanavir resistance. Their impacts differ, with I50L, I84V, and N88S having the greatest effect. Higher atazanavir levels obtained with ritonavir boosting increase the number of mutations required for loss of activity. The presence of M46I plus L76V might increase susceptibility to atazanavir when no other related mutations are present.[60]

t. HIV-1 RNA response to ritonavir-boosted darunavir correlates with baseline susceptibility and the presence of several specific PI resistance–associated mutations. Reductions in response are associated with increasing numbers of the mutations indicated in the figure bar. The negative impact of the protease mutations I47V, I54M, T74P, and I84V and the positive impact of the protease mutation V82A on virologic response to ritonavir-boosted darunavir were shown in 2 data sets independently.[61,62] Some of these mutations appear to have a greater effect on susceptibility than others (eg, I50V vs V11I). The presence at baseline of 2 or more of the substitutions V11I, V32I, L33F, I47V, I50V, I54L or M, T74P, L76V, I84V or L89V was associated with a decreased virologic response to ritonavir-boosted darunavir.[63]

u. The mutations depicted on the figure bar cannot be considered comprehensive because little relevant research has been reported in recent years to update the resistance and cross-resistance patterns for this drug.

v. In PI-experienced patients, the accumulation of 6 or more of the mutations indicated on the figure bar is associated with a reduced virologic response to ritonavir-boosted lopinavir.[64,65] The product information states that accumulation of 7 or 8 mutations confers resistance to the drug.[66] However, there is emerging evidence that specific mutations, most notably I47A (and possibly I47V) and V32I, are associated with high-level resistance.[67-69] The addition of L76V to 3 PI resistance–associated mutations substantially increases resistance to ritonavir-boosted lopinavir.[60]

w. In some nonsubtype-B HIV-1, D30N is selected less frequently than are other PI resistance–associated mutations.[70]

x. Resistance to enfuvirtide is associated primarily with mutations in the first heptad repeat (HR1) region of the gp41 envelope gene. However, mutations or polymorphisms in other regions of the envelope (eg, the HR2 region or those yet to be identified) as well as coreceptor usage and density may affect susceptibility to enfuvirtide.[71-73]

y. The activity of CC chemokine receptor 5 (CCR5) antagonists is limited to patients with virus that uses only CCR5 for entry (R5 virus). Viruses that use both CCR5 and CXC chemokine receptor 4 (CXCR4; termed dual/mixed [D/M] virus) or only CXCR4 (X4 virus) do not respond to treatment with CCR5 antagonists. Virologic failure of these drugs is frequently associated with outgrowth of D/M or X4 virus from a preexisting minority population present at levels below the limit of assay detection. Mutations in HIV-1 gp120 that allow the virus to bind to the drug-bound form of CCR5 have been described in viruses from some patients whose virus remained R5 after virologic failure of a CCR5 antagonist. Most of these mutations are found in the V3 loop, the major determinant of viral tropism.[74] There is as yet no consensus on specific signature mutations for CCR5 antagonist resistance, so they are not depicted in the figure. Some CCR5 antagonist–resistant viruses selected in vitro have shown mutations in gp41 without mutations in V3[75]; the clinical significance of such mutations is not yet known.

z. In site-directed mutants and clinical isolates, the mutation F121Y has a profound effect on susceptibility to elvitegravir and raltegravir and to a lesser extent to dolutegravir. Mutation R263K can be selected in vivo during treatment with dolutegravir and raltegravir and results in a 2- to 5-fold reduction in susceptibility to dolutegravir, elvitegravir, and raltegravir.[76-81]

aa. Several mutations are required in HIV integrase to confer high-level resistance to dolutegravir.[82] Cross-resistance studies with raltegravir- and elvitegravir-resistant viruses indicate that Q148H/R and G140S in combination with mutations L74I/M, E92Q, T97A, E138A/K, G140A, or N155H are associated with 5-fold to 20-fold reduced dolutegravir susceptibility [83] and reduced virologic suppression in patients.[84-87]

bb. Seven elvitegravir codon mutations have been observed in integrase strand transfer inhibitor treatment–naive and –experienced patients in whom therapy is failing.[88-94] T97A results in only a 2-fold change in elvitegravir susceptibility and may require additional mutations for resistance.[93,94] The sequential use of elvitegravir and raltegravir (in either order) is not recommended because of cross-resistance between these drugs.[93]

cc. Raltegravir failure is associated with integrase mutations in at least 3 distinct, but not exclusive, genetic pathways defined by 2 or more mutations including (1) a signature (major) mutation at Q148H/K/R, N155H, or Y143R/H/C; and (2) 1 or more additional minor mutations. Minor mutations described in the Q148H/K/R pathway include L74M plus E138A, E138K, or G140S. The most common mutational pattern in this pathway is Q148H plus G140S, which also confers the greatest loss of drug susceptibility. Mutations described in the N155H pathway include this major mutation plus either L74M, E92Q, T97A, E92Q plus T97A, Y143H, G163K/R, V151I, or D232N.[95] The Y143R/H/C mutation is uncommon.[96-100] E92Q alone reduces susceptibility to elvitegravir more than 20 fold and causes limited (< 5 fold) cross-resistance to raltegravir.[90,101-103] N155H mutants tend to predominate early in the course of raltegravir failure, but are gradually replaced by viruses with higher resistance, often bearing mutations G140S plus Q148H/R/K, with continuing raltegravir treatment.[96]

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