HIVDB Genotypic Drug Resistance Interpretation Program

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Disclosures

- Within the past three years:
  - Research funding from Janssen Pharmaceuticals, Vela Diagnostics
  - Consulting from Abbott Diagnostics

- Database Funding:
  - NIH / NIAID
  - Several grants since 2000
  - Most recently: R24AI136618

Outline

- **Principles of resistance interpretation**
  - **Approaches to developing an expert system**
- HIVDB drug resistance “knowledgebase”
- HIVDB drug resistance interpretation program

Expert Systems: Rule-Based vs. Machine Learning

- Rule-Based
  - Can use data from a wide variety of sources. Even if the raw data are not available
  - Can integrate expert opinion
  - Transparent / Educational
  - Subjective
- Machine learning
  - Requires massive amount of raw data
  - Lacks explanatory component
  - Can be optimized to a given dataset

Rules-Based Expert Systems

- Logical rules
  - Example: “At least 4 mutations among: M41L, E44D, D67N, T69D/N/S, L74V/I, L210W, T215A/C/D/E/G/H/I/L/N/S/V/Y/F” => TDF resistance (ANRS)
- Scores
  - Scores for individual mutations and for combinations of mutations
  - Adding up the scores leads to a level of drug resistance

Goals of Expert Systems

- Yields an optimal result
  - May not be correct
  - But is guaranteed to be optimal according to some criteria
- Mimics an expert
  - May not be correct
  - Expert often gives background information

Additional Limitations

- Doesn’t contain sufficient information to choose a regimen without knowledge of:
  - Principles of HIV therapy
  - Treatment guidelines
  - Other information about a patient
- Currently doesn’t recommend regimens

HIVDB Program: Levels of HIVDR

Resistance Level	Definition	Score range
Susceptible	No evidence of reduced susceptibility	<10
Potential low level resistance	DRMs consistent with previous ARV exposure or DRMs associated with resistance only when they occur with other DRMs	10-14
Low-level resistance	DRMs associated with a reduction in vitro ARV susceptibility or a suboptimal virological response to ARV treatment.	15-29
Intermediate resistance	A high likelihood that ARV activity would be reduced. However, the ARV would likely still retain antiviral activity.	30-59
High-level resistance	A level of resistance similar to that observed in viruses with the highest levels of reduced in vitro susceptibility or in viruses that have little or no virological response to ARV treatment.	≥60

How DRM Scores are Derived

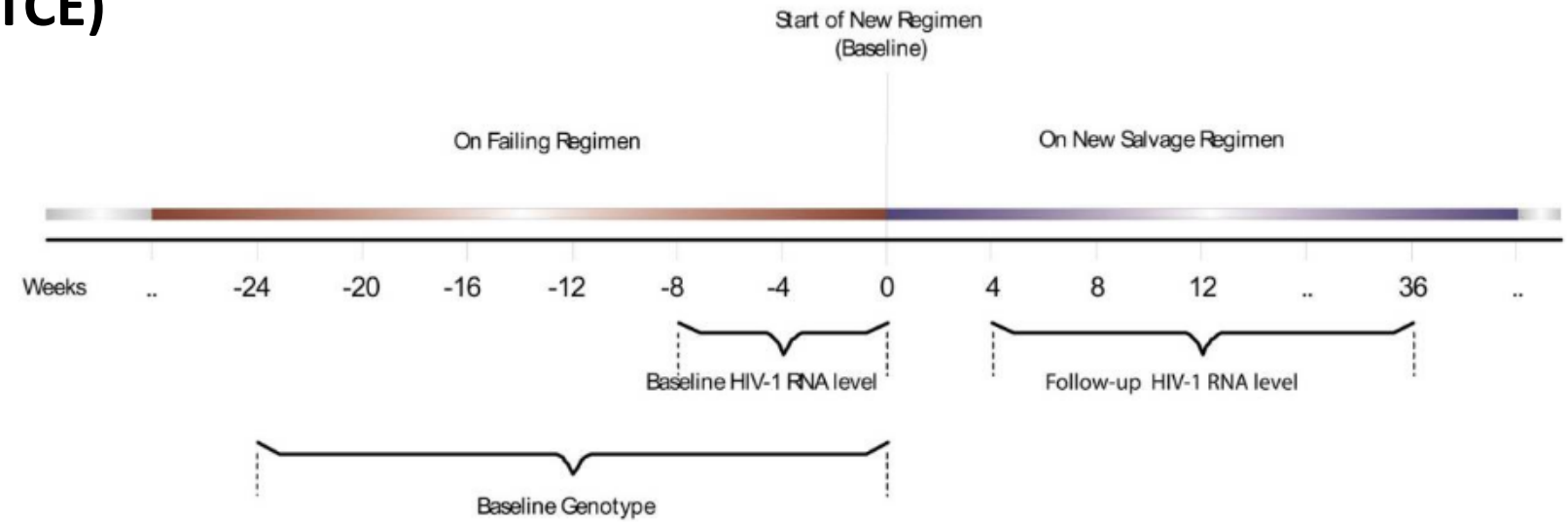
- Selected by drugs in vitro (“passage experiments”)
- Selected by drugs in vivo (persons developing virological failure)
  - Prevalence of mutations in ARV-naïve and ARV-experienced persons
- Reduce in vitro susceptibility (“phenotypic testing”)
  - Site-directed mutants
  - Clinical isolates
- Reduce response to a salvage therapy regimen
  - Clinical trials
  - Retrospective cohort studies

Virological Response Data

Treatment-change episode (TCE)

Pre-Genotype Factors:
Rx History
VL
CD4

Post-Genotype Factors:
Did care provider use genotype?
Other ARVs
Adherence



Analysis of TCE data

How DRM Scores are Derived - Expert Opinion

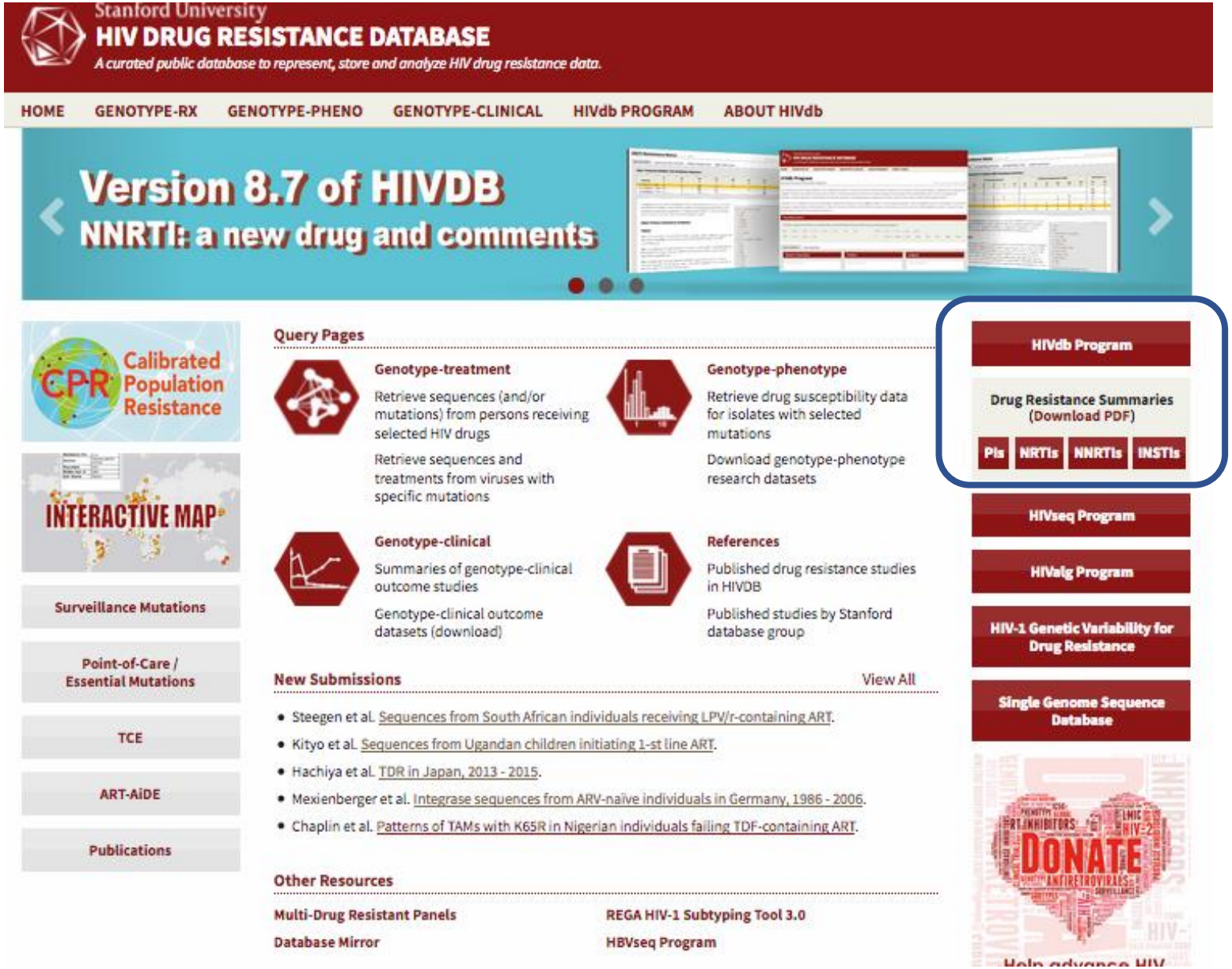
- Balancing the previous lines of evidence.
- Accounting for the fact that some ARVs are more potent than others or have performed better in clinical trials.

- Individual and combination scores are titrated so they produce the “correct” level of resistance: Susceptible, Potential low level, Low level, Intermediate, High level

Outline

- Principles of resistance interpretation and expert systems
- **HIVDB drug resistance "knowledgebase"**
  - **Scores, comments, and notes pages**
- HIVDB drug resistance interpretation program

HIVDB Home Page



HIVDB Program and Supporting Material

Query Pages

Genotype-treatment

Retrieve sequences (and/or mutations) from persons receiving selected HIV drugs

Retrieve sequences and treatments from viruses with

Genotype-phenotype

Retrieve drug susceptibility data for isolates with selected mutations

Download genotype-phenotype research datasets

HIVdb Program

Drug Resistance Summaries (Download PDF)

PIs **NRTIs** **NNRTIs** **INSTIs**

Notes (INSTIs)

HIV POL DRUG RESISTANCE SUMMARY

- DR Notes
- DR Comments
- Mutation Scores
- Pattern Scores
- Mut Scores Editor

INSTI Resistance Notes (PI · NRTI · NNRTI · INSTI)

HIVdb version 8.7 (last updated on 2018-10-19)
Notes last updated on 2018-07-16

Major Integrase Inhibitor (INSTI) Resistance Mutations

Consensus	66 T	92 E	118 G	138 E	140 G	143 Y	147 S	148 Q	155 N	263 R
Bictegravir (BIC)	K	Q	R	KAT	SAC			HRK	H	K
Dolutegravir (DTG)	K	Q	R	KAT	SAC			HRK	H	K
Elvitegravir (EVG)	AIK	Q	R	KAT	SAC		G	HRK	H	K
Raltegravir (RAL)	AIK	Q	R	KAT	SAC	RCH		HRK	H	K

The table lists the most common clinically significant INSTI-resistance mutations. Mutations in bold red are associated with the highest levels of reduced susceptibility or virological response to the indicated INSTI. Mutations in bold reduce INSTI susceptibility or virological response. Mutations in plain text contribute to reduced suceptibility in combination with other INSTI-resistance mutations.

- H51Y
- T66A/I/K
- L74M/I/F
- E92Q/G/V
- T97A
- G118R
- F121Y
- E138K/A/T
- G140S/A/C
- P142T
- Y143C/R/H/K/S/G/A
- P145S
- Q146P
- S147G
- Q148H/K/R/N
- G149A

H51Y

H51Y is a rare nonpolymorphic accessory mutation. It is selected in vitro by EVG (1,2,3) and DTG (4) and in patients receiving RAL (5,6) and EVG (7,8). It reduces EVG susceptibility by ~2-3-fold (1,3,4,9,10) but alone does not appear to reduce RAL, DTG, or BIC susceptibility (3,4,11,12).

T66A/I/K

T66A is a nonpolymorphic mutation selected in patients receiving EVG (5,13,8,14) and RAL (15,16,5). It reduces EVG susceptibility ~5-fold but has minimal effect on RAL, DTG or BIC susceptibility (17,3,18,19,20,10).

T66I is a nonpolymorphic mutation frequently selected in vitro (2,1,21,3,22) and in patients receiving

Notes (NRTI Table)

Major Nucleoside RT Inhibitor (NRTI) Resistance Mutations

	Discriminatory Mutations					Thymidine Analog Mutations (TAMs)						MDR Mutations	
	184	65	70	74	115	41	67	70	210	215	219	69	151
Consensus	M	K	K	L	Y	M	D	K	L	T	K	T	Q
3TC	VI	R										Ins	M
FTC	VI	R										Ins	M
ABC	VI	R	E	VI	F	L			W	FY		Ins	M
DDI	VI	R	E	VI		L			W	FY		Ins	M
TDF	***	R	E		F	L		R	W	FY		Ins	M
D4T	***	R	E			L	N	R	W	FY	QE	Ins	M
ZDV	***	***	*	*		L	N	R	W	FY	QE	Ins	M

Handout with Tables: NRTIs and NNRTIs

Major Nucleoside RT Inhibitor (NRTI) Resistance Mutations													
	Non-TAMs					TAMs						MDR	
	184	65	70	74	115	41	67	70	210	215	219	69	151
Cons	M	K	K	L	Y	M	D	K	L	T	K	T	Q
3TC	VI	R										Ins	M
FTC	VI	R										Ins	M
ABC	VI	R	E	VI	F	L			W	FY		Ins	M
TDF	***	R	E		F	L		R	W	FY		Ins	M
ZDV	***	***	*	*		L	N	R	W	FY	QE	Ins	M

Major Non-Nucleoside RT Inhibitor (NNRTI) Resistance Mutations								
	100	101	103	106	181	188	190	230
Cons	L	K	K	V	Y	Y	G	M
DOR	I	EP		AMI	CIV	LCH	SE	L
EFV	I	EP	NS	AM	CIV	LCH	ASEQ	L
ETR	I	EP			CIV	L	ASEQ	L
RPV	I	EP			CIV	L	ASEQ	L
NVP	I	EP	NS	AM	CIV	LCH	ASEQ	L

Handout with Tables: INSTIs and PIs

Major Integrase Inhibitor (INSTI) Resistance Mutations										
	66	92	118	138	140	143	147	148	155	263
Cons	T	E	G	E	G	Y	S	Q	N	R
BIC	K	Q	R	KAT	SAC			**HRK**	H	K
DTG	K	Q	R	KAT	SAC			**HRK**	H	K
EVG	**AIK**	**Q**	**R**	**KAT**	**SAC**		**G**	**HRK**	**H**	**K**
RAL	AI**K**	**Q**	**R**	**KAT**	**SAC**	**RC**		**HRK**	**H**	K

Major Protease Inhibitor (PI) Resistance Mutations											
	32	46	47	48	50	54	76	82	84	88	90
Cons	V	M	I	G	I	I	L	V	I	N	L
ATV/r	I	IL	V	**VM**	**L**	VTALM		ATFS	**V**	**S**	**M**
DRV/r	**I**		VA		**V**	**LM**	**V**	F	**V**		
LPV/r	**I**	IL	V**A**	VM	**V**	**VTALM**	**V**	**AFTS**	**V**		M

Bold underline: High-level reduced susceptibility or virological response. **Bold**: Reduced suceptibility or virological response. Plain text: Reduced susceptibility in combination with other PI resistance mutations

Comments and Mutation Classification (INSTIs)

HIV POL DRUG RESISTANCE SUMMARY

- DR Notes
- **DR Comments**
- Mutation Scores
- Pattern Scores
- Mut Scores Editor

INSTI Resistance Comments

(PI · NRTI · NNRTI · INSTI)

HIVdb version 8.7 (last updated on 2018-10-19)

Condition	Comment/ Mutation Type	Comment
50I	Other	M50I is a polymorphic mutation selected in vitro by DTG and BIC in combination with R263K. It appears to contribute to reduced DTG susceptibility in combination with R263K.
51Y	Accessory	H51Y is a rare non-polymorphic accessory mutation selected in patients receiving RAL and EVG and in vitro by DTG. H51Y minimally reduces EVG susceptibility (~2 to 3-fold). It does not reduce RAL or DTG susceptibility.
66A	Major	T66A is a non-polymorphic mutation selected in patients receiving EVG and RAL, usually in combination with other INSTI-resistance mutations. It causes a moderate reduction in EVG susceptibility but does not appear to reduce RAL, DTG, or BIC susceptibility.
66I	Major	T66I is a non-polymorphic mutation selected in patients receiving EVG, RAL, and DTG. It reduces EVG susceptibility about 10-fold but does not reduce RAL, DTG, or BIC susceptibility.
66K	Major	T66K is a non-polymorphic mutation selected in patients receiving EVG. It is associated with high-level EVG resistance, intermediate/high-level RAL resistance, and low-level DTG resistance. Its effect on BIC is not known.
74MIF	Other	L74M/I are polymorphic accessory mutations commonly selected by each of the INSTIs. In ARV-naive patients, L74M occurs in 0.5% to 10% of patients and L74I occurs in 3% to 20% of patients depending on subtype. Alone, L74M/I have minimal, if any, effect on INSTI susceptibility. However, they contribute reduced susceptibility to each of the INSTIs when they occur with major INSTI-resistance mutations. L74F is a rare nonpolymorphic mutation which also contributes reduced susceptibility when it occurs with other INSTI-resistance mutations.

Scores (INSTIs Individual Mutations)

HIV POL DRUG RESISTANCE SUMMARY

- DR Notes
- DR Comments
- **Mutation Scores**
- Pattern Scores
- Mut Scores Editor

INSTI Resistance Mutation Scores

(PI · NRTI · NNRTI · INSTI)

HIVdb version 8.7 (last updated on 2018-10-19)

Rule ▲	BIC	DTG	EVG	RAL
H51Y	10	10	15	15
T66A	0	0	60	15
T66I	5	5	60	15
T66K	15	15	60	60
E92G	0	0	30	15
E92Q	10	10	60	30
E92V	0	0	60	30
Q95K	0	0	10	10
T97A	0	0	10	10
G118R	15	15	30	30
F121Y	10	10	60	60
E138A	10	10	15	15
E138K	10	10	15	15
E138T	10	10	15	15
G140A	10	10	30	30
G140C	10	10	30	30
G140S	10	10	30	30

Scores (INSTIs, sorted by DTG)

INSTI Resistance Mutation Scores (PI · NRTI · NNRTI · INSTI)

HIVdb version 8.7 (last updated on 2018-10-19)

Rule	BIC	DTG ▼	EVG	RAL
Q148K	30	30	60	60
Q148H	25	25	60	60
Q148R	25	25	60	60
R263K	25	25	30	25
S230R	10	20	20	20
T66K	15	15	60	60
G118R	15	15	30	30
V151L	15	15	60	30
S153F	15	15	15	0
S153Y	15	15	15	0
H51Y	10	10	15	15
E92Q	10	10	60	30
F121Y	10	10	60	60
E138A	10	10	15	15
E138K	10	10	15	15
E138T	10	10	15	15
G140A	10	10	30	30
G140C	10	10	30	30
G140S	10	10	30	30
N155H	10	10	60	60

Scores (Mutation Combinations, INSTIs)

Combination Rule ▲	BIC	DTG	EVG	RAL
E138AKT + G140ACS	10	10	15	15
E138AKT + Q148HKR	10	10	0	0
G140ACS + Q148HKR	10	10	0	0
Y143ACGHRS + G163R	5	5	5	0
Y143ACGHRS + S230R	5	5	5	0
Q148HKR + N155H	10	10	0	0
Q148HKR + G163KR	5	5	0	0
E157Q + R263K	10	10	0	0
H51Y + R263K	10	10	15	0
L74FM + Y143ACGHRS	5	5	5	0
L74FM + Q148HKR	10	10	10	10
E92Q + N155H	5	5	0	0
T97A + Y143ACGHRS	0	0	5	0
T97A + Q148HKR	15	15	0	0

Published NRTI Mutation Patterns

NRTI Mutation Pattern Scores (PI · NRTI · NNRTI · INSTI)

HIVdb version 8.7 (last updated on 2018-10-1

Pattern	count ▼	ABC	AZT	D4T	DDI	FTC	3TC	TDF
M184V	6733	15	-10	-10	10	60	60	-10
M41L + M184V + T215Y	754	45	55	55	45	65	65	15
D67N + K70R + M184V + K219Q	623	60	55	40	40	70	70	15
A62V	591	5	5	5	5	5	5	5
M41L + M184V + L210W + T215Y	523	75	90	90	75	75	75	45
K70R + M184V	444	20	20	5	20	60	60	-5
M41L + T215Y	416	30	65	65	35	5	5	25
M184V + T215Y	403	25	30	30	25	60	60	0
M41L + L210W + T215Y	371	60	100	100	65	15	15	55
K65R + M184V	338	60	-25	50	70	90	90	50
K65R	330	45	-15	60	60	30	30	60
M41L	330	5	15	15	10	0	0	5
T215S	321	5	20	20	10	0	0	5
K70R	318	5	30	15	10	0	0	5
L74V + M184V	316	60	-10	-10	70	60	60	-10
A62V + M184V	312	20	-5	-5	15	65	65	-5
T215Y	283	10	40	40	15	0	0	10

Published INSTI Mutation Patterns

INSTI Mutation Pattern Scores (PI · NRTI · NNRTI · INSTI)

HIVdb version 8.7 (last updated on 2018-10-19)

Pattern	count ▼	BIC	DTG	EVG	RAL
E157Q	352	0	0	10	10
G140S + Q148H	195	45	45	90	90
T97A	173	0	0	10	10
N155H	146	10	10	60	60
N155H + G163R	33	10	10	75	75
N155H + E157Q	32	10	10	70	70
G163R	30	0	0	15	15
T97A + Y143R	23	5	5	25	70
E138K	21	10	10	15	15
E138A + G140S + Q148H	19	75	75	120	120
T97A + N155H	18	10	10	70	70
G163K	17	0	0	15	15
R263K	17	25	25	30	25
E92Q + N155H	16	25	25	120	90
Y143R	16	5	5	10	60
E138K + G140S + Q148H	15	75	75	120	120
E92Q	15	10	10	60	30
G140S + Q148R	13	45	45	90	90
Q148R	11	25	25	60	60

Published INSTI Mutation Patterns (Sorted by DTG)

INSTI Mutation Pattern Scores (PI · NRTI · NNRTI · INSTI)

HIVdb version 8.7 (last updated on 2018-10-19)

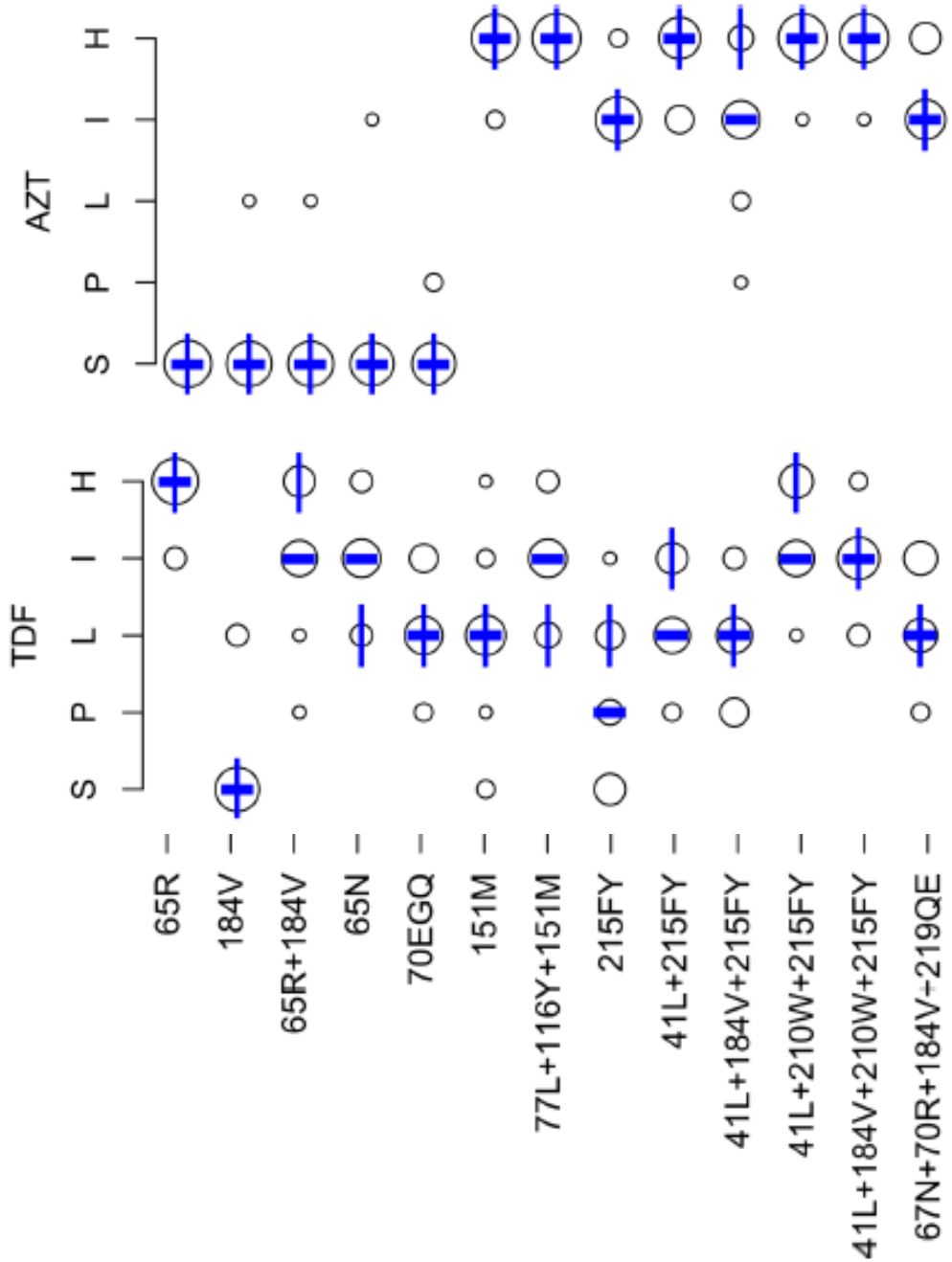
Pattern	count	BIC	DTG ▼	EVG	RAL
E138K + G140A + Q148K + N155H	1	100	100	180	180
H51Y + E92Q + G140S + Q148K + N155H + G163R	1	100	100	240	210
E138K + G140A + Q148R + N155H	2	95	95	180	180
H51Y + E92Q + G140S + Q148K + N155H	2	95	95	225	195
T97A + E138A + G140S + Q148H	1	90	90	130	130
E92Q + G140S + Q148K + N155H	2	85	85	210	180
L74M + E138K + G140C + Q148R + E157Q	1	85	85	140	140
L74M + E138T + G140S + Q148H	1	85	85	130	130
T97A + E138K + Q148R + N155H + G163K	1	85	85	160	160
T97A + G140S + Q148R + N155H + G163R	1	85	85	175	175
E138K + G140A + Q148K	3	80	80	120	120
E92Q + G140S + Q148R + N155H	2	80	80	210	180
E138A + G140S + Y143H + Q148H	1	80	80	130	180
E138K + G140S + Q148R + G163R	1	80	80	135	135
E138A + G140S + Q148H	19	75	75	120	120
E138K + G140S + Q148H	15	75	75	120	120
E138T + G140S + Q148H	6	75	75	120	120
E138A + G140A + Q148R	4	75	75	120	120

RESEARCH ARTICLE

Collaborative update of a rule-based expert system for HIV-1 genotypic resistance test interpretation

Roger Paredes[1], Philip L. Tzou[2], Gert van Zyl[3], Geoff Barrow[4], Ricardo Camacho[5], Sergio Carmona[6], Philip M. Grant[2], Ravindra K. Gupta[7], Raph L. Hamers[8], P. Richard Harrigan[9], Michael R. Jordan[10], Rami Kantor[11], David A. Katzenstein[2], Daniel R. Kuritzkes[12], Frank Maldarelli[13], Dan Otelea[14], Carole L. Wallis[15], Jonathan M. Schapiro[16], Robert W. Shafer[2]*

NRTIs



Expert Panel Paper: Susceptibility Data for NRTI Mutation Patterns

Table 5. In vitro susceptibilities associated with the 13 NRTI drug resistance mutation (DRM) patterns.

Overall Pattern*	Specific Pattern*	Exact†	Included†	3TC§	ABC§	AZT§	TDF§
M184V	M184V	19.03%	63.33%	**>200$_{175}$**	3.1$_{125}$	0.5$_{124}$	0.5$_{63}$
K65R	K65R	0.93%	5.64%	**8.9$_{30}$**	2.5$_{20}$	0.5$_{20}$	**1.8$_{17}$**
K65R, M184V	K65R, M184V	0.96%	2.88%	**>200$_{27}$**	**8.4$_{16}$**	0.4$_{16}$	1.2$_{16}$
K65N	K65N	0.02%	0.10%	**7.3$_{1}$**	2.1$_{1}$	-	**1.7$_{1}$**
K70EGQ	K70E	0.07%	0.85%	**5.3$_{5}$**	1.4$_{3}$	0.2$_{2}$	0.9$_{3}$
	K70G	0.00%	0.31%	-	-	-	-
	K70Q	0.03%	0.27%	-	-	-	-
T215YF	T215Y	0.80%	28.76%	2.4$_{19}$	1.6$_{12}$	**7.4$_{15}$**	**1.4$_{14}$**
	T215F	0.21%	10.29%	2.4$_{4}$	1.8$_{2}$	**5$_{2}$**	1.3$_{2}$
M41L, T215YF	M41L, T215Y	1.18%	23.87%	2$_{15}$	2$_{9}$	**12$_{12}$**	1.3$_{7}$
	M41L, T215F	0.23%	5.24%	2.6$_{1}$	3.2$_{1}$	**50$_{1}$**	-
M41L, M184V, T215YF	M41L, M184V, T215Y	2.13%	14.34%	**>200$_{55}$**	**5.1$_{41}$**	**6$_{41}$**	1.1$_{24}$
	M41L, M184V, T215F	0.45%	3.39%	**>200$_{6}$**	**5.4$_{7}$**	**3.5$_{7}$**	0.5$_{1}$
M41L, L210W, T215YF	M41L, L210W, T215Y	1.05%	16.44%	2.8$_{34}$	3.1$_{19}$	**164$_{21}$**	**3.1$_{18}$**
	M41L, L210W, T215F	0.05%	1.04%	**3.1$_{4}$**	3.2$_{1}$	**217$_{3}$**	**4.1$_{2}$**
M41L, M184V, L210W, T215YF	M41L, M184V, L210W, T215Y	1.48%	9.65%	**>200$_{69}$**	**6.5$_{48}$**	**18$_{51}$**	**1.6$_{38}$**
	M41L, M184V, L210W, T215F	0.10%	0.66%	**148$_{1}$**	-	**69$_{1}$**	**2.8$_{1}$**

RESEARCH ARTICLE

Collaborative update of a rule-based expert system for HIV-1 genotypic resistance test interpretation

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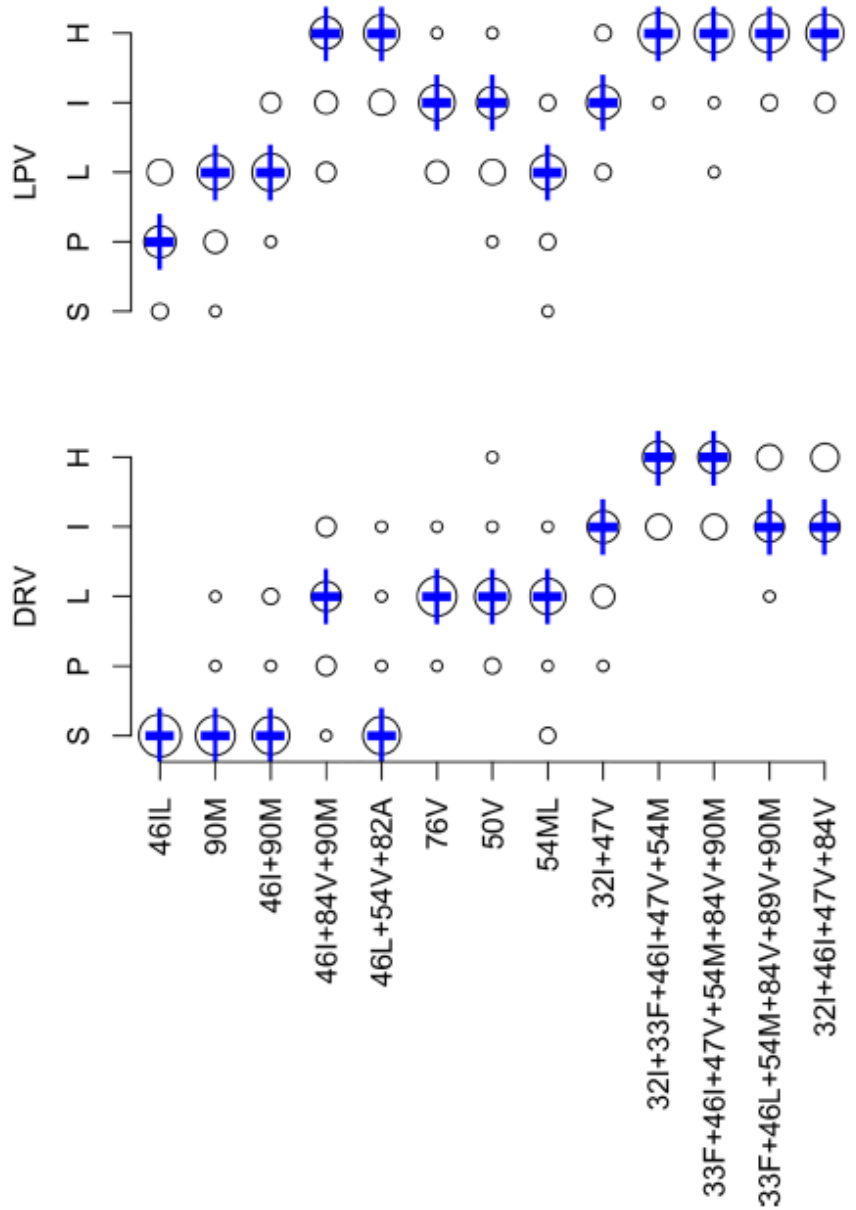
PIs



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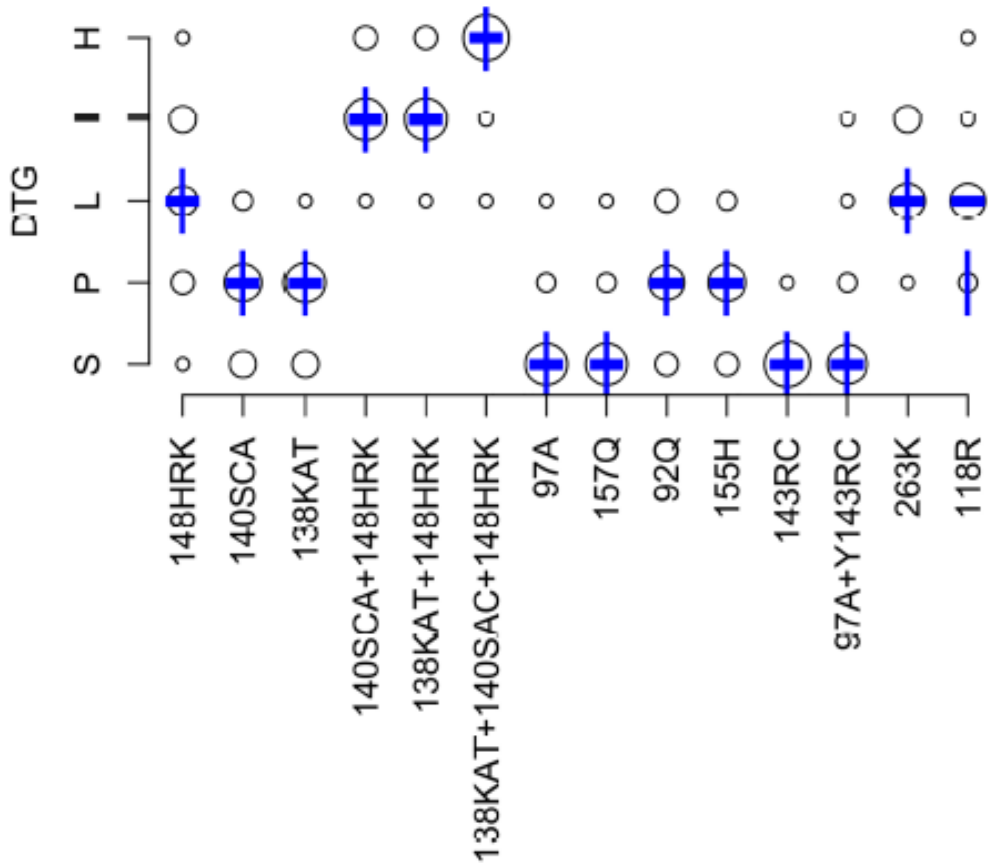
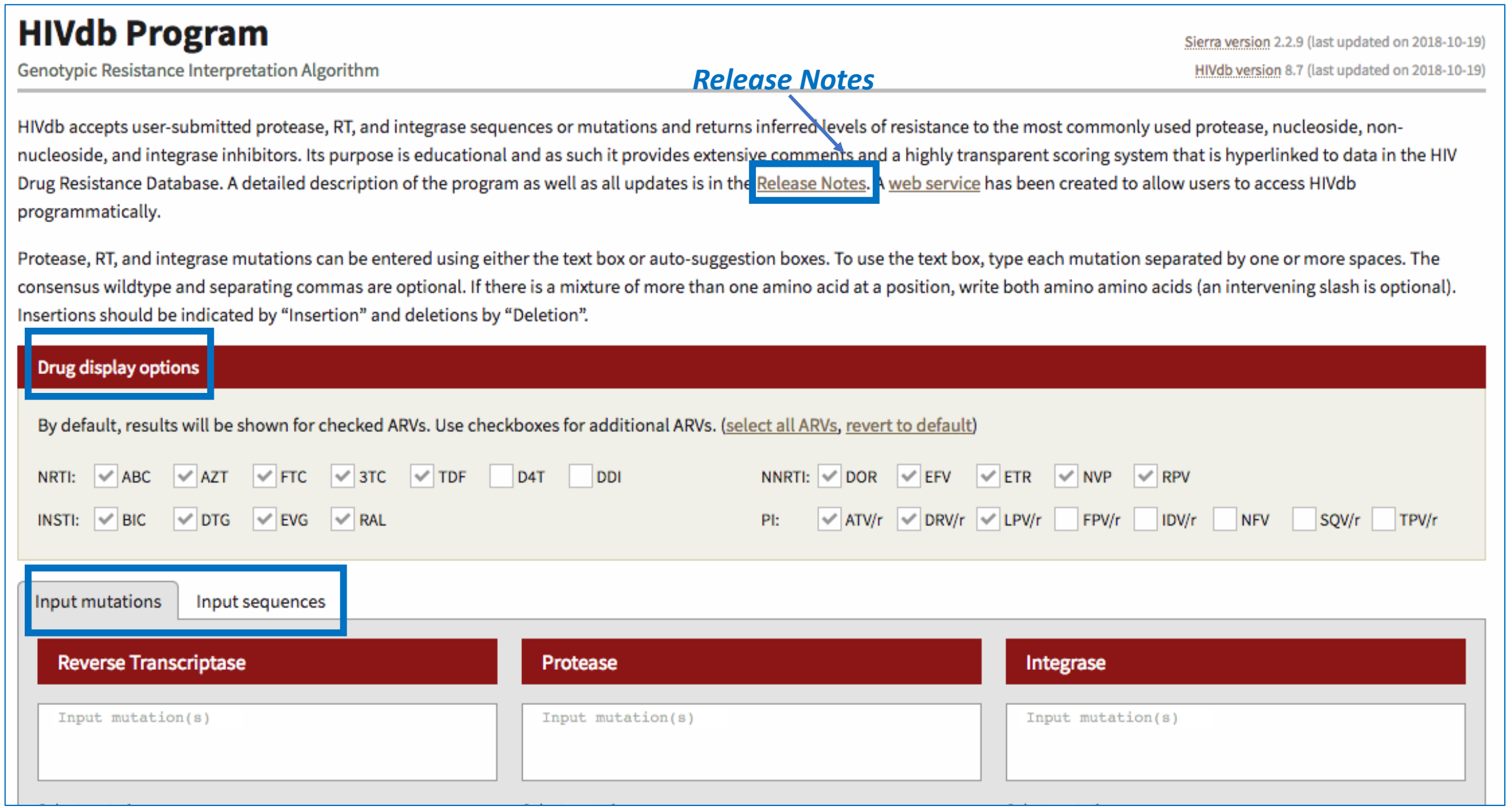
INSTIs



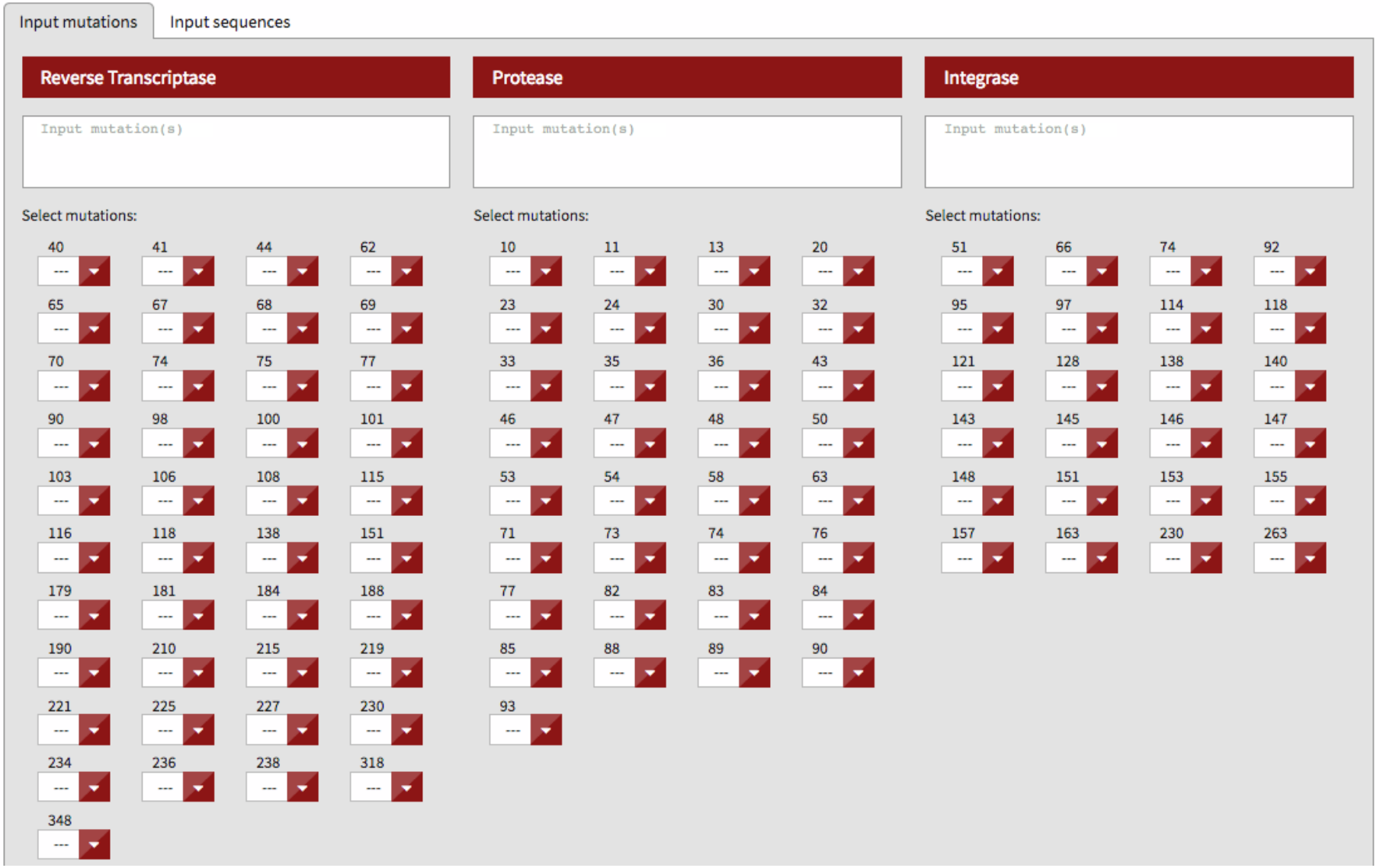
Outline

- Principles of resistance interpretation
- HIVDB drug resistance “knowledgebase”
- **HIVDB drug resistance interpretation program**

HIVDB Program Landing Page



HIVDB Program Landing Page: Mutation List Options



Mutation List Example

PI Interpretation - Mutation Classification, Levels, Comments

Drug Resistance Interpretation: PR

PI Major Resistance Mutations:	**M46I, I54V, L76V, V82A**
PI Accessory Resistance Mutations:	**L10F, L89V**
Other Mutations:	None

Protease Inhibitors	
atazanavir/r (ATV/r)	High-Level Resistance
darunavir/r (DRV/r)	Intermediate Resistance
lopinavir/r (LPV/r)	High-Level Resistance

PR Comments

PI Major

- **M46I/L** are relatively non-polymorphic PI-selected mutations. In combination with other PI-resistance mutations, they are associated with reduced susceptibility to each of the PIs except DRV.
- **I54V** is a non-polymorphic PI-selected mutation that contributes reduced susceptibility to each of the PIs except DRV.
- **L76V** is a non-polymorphic mutation selected by IDV, LPV and DRV. It reduces susceptibility to these PIs and to FPV and NFV. It increases susceptibility to ATV, SQV and TPV. **L76V** is included in the Tibotec DRV genotypic susceptibility score.
- **V82A** is a non-polymorphic mutation selected primarily by IDV and LPV. It reduces susceptibility to these PIs and contributes cross-resistance to each of the remaining PIs except DRV and TPV.

PI Accessory

- **L10F** is a common non-polymorphic, PI-selected accessory mutation associated with reduced susceptibility to DRV, FPV, IDV, LPV, and NFV.
- **L89V** is a non-polymorphic PI-selected accessory mutation that contributes reduced susceptibility to FPV, DRV, NFV, and IDV. **L89V** is included in the Tibotec DRV genotypic susceptibility score. L89T is a rare non-polymorphic PI-selected mutation that has not been well studied.

Dosage Considerations

- There is evidence for intermediate **DRV** resistance. If **DRV** is administered it should be used twice daily.

PI Interpretation - Scoring

Mutation Scoring: PR

PI	ATV/r	DRV/r	LPV/r
M46I	10	0	10
I54V	15	0	15
V82A	15	0	30
M46I + V82A	10	0	10
I54V + V82A	10	0	10
L10F	0	5	5
L76V	0	20	30
L89V	0	5	0
M46I + L76V	0	0	10
Total	60	30	120

RTI Interpretation - Mutation Classification and Levels

Drug Resistance Interpretation: RT

NRTI Resistance Mutations:	**K65R, M184IV**
NNRTI Resistance Mutations:	**Y181C, G190A**
Other Mutations:	None

Nucleoside Reverse Transcriptase Inhibitors	
abacavir (ABC)	High-Level Resistance
zidovudine (AZT)	Susceptible
emtricitabine (FTC)	High-Level Resistance
lamivudine (3TC)	High-Level Resistance
tenofovir (TDF)	Intermediate Resistance

Non-nucleoside Reverse Transcriptase Inhibitors	
doravirine (DOR)	Intermediate Resistance
efavirenz (EFV)	High-Level Resistance
etravirine (ETR)	Intermediate Resistance
nevirapine (NVP)	High-Level Resistance
rilpivirine (RPV)	High-Level Resistance

RTI Interpretation - Comments

RT Comments

NRTI

- **M184V/I** cause high-level in vitro resistance to 3TC and FTC and low-level resistance to ddI and ABC. However, **M184V/I** are not contraindications to continued treatment with 3TC or FTC because they increase susceptibility to AZT, TDF and d4T and are associated with clinically significant reductions in HIV-1 replication.
- **K65R** causes intermediate/high-level resistance to TDF, ddI, ABC and d4T and low/intermediate resistance to 3TC and FTC. **K65R** increases susceptibility to AZT.

NNRTI

- **Y181C** is a non-polymorphic mutation selected in patients receiving NVP, ETR and RPV. It reduces susceptibility to NVP, ETR, RPV, and EFV by >50-fold, 5-fold, 3-fold, and 2-fold, respectively. Although **Y181C** itself reduces EFV susceptibility by only 2-fold, it has been associated with a reduced response to an EFV-containing regimen in NNRTI-experienced patients. **Y181C** has a weight of 2.5 in the Tibotec ETR genotypic susceptibility score. Alone, it does not appear to reduce DOR susceptibility.
- **G190A** is a non-polymorphic mutation that causes high-level resistance to NVP and intermediate resistance to EFV. It has a weight of 1.0 in the Tibotec ETR genotypic susceptibility score but does not appear to be selected by ETR or RPV or to reduce their in vitro susceptibility in the absence of other NNRTI-resistance mutations. It also does not appear to reduce DOR susceptibility.

RTI Interpretation - Scoring

Mutation Scoring: RT

NRTI	ABC	AZT	FTC	3TC	TDF
K65R	45	-15	30	30	60
M184I	15	-10	60	60	-10
Total	60	-25	90	90	50

NNRTI	DOR	EFV	ETR	NVP	RPV
Y181C	10	30	30	60	45
Y181C + G190A	20	0	10	0	10
G190A	0	45	10	60	15
Total	30	75	50	120	70

INSTI Interpretation - Mutation Classification, Levels, Comments

Drug Resistance Interpretation: IN

IN Major Resistance Mutations:	**G140S, Q148H**
IN Accessory Resistance Mutations:	**T97A**
Other Mutations:	None

Integrase Strand Transfer Inhibitors	
bictegravir (BIC)	High-Level Resistance
dolutegravir (DTG)	High-Level Resistance
elvitegravir (EVG)	High-Level Resistance
raltegravir (RAL)	High-Level Resistance

IN Comments

IN Major

- **G140S/A/C** are non-polymorphic mutations that usually occur with Q148 mutations. Alone, they have minimal effects on INSTI susceptibility. However, in combination with Q148 mutations they are associated with high-level resistance to RAL and EVG and intermediate reductions in DTG and BIC susceptibility.
- **Q148H/K/R** are non-polymorphic mutations selected by RAL, EVG, and rarely DTG. **Q148H/R/K** are associated with high-level reductions in RAL and EVG susceptibility particularly when they occur In combination with E138 or G140 mutations. Alone, **Q148H/K/R** have minimal effects on DTG and BIC susceptibility. But in combination with E138 and G140 mutations they cause moderate and occasionally high-level reductions in DTG and BIC susceptibility.

IN Accessory

- **T97A** is a polymorphic INSTI-selected mutation that, depending on subtype, occurs in 1% to 5% of viruses from untreated persons. Alone, it has minimal effects on INSTI susceptibility but in combination with other major resistance mutations, it synergistically reduces susceptibility to EVG, RAL, DTG, and possibly BIC.

Dosage Considerations

- There is evidence for high-level **DTG** resistance. If **DTG** is used, it should be administered twice daily.

INSTI Interpretation - Scoring

Mutation Scoring: IN

INSTI	BIC	DTG	EVG	RAL
G140S	10	10	30	30
Q148H	25	25	60	60
T97A + Q148H	15	15	0	0
G140S + Q148H	10	10	0	0
T97A	0	0	10	10
Total	60	60	100	100

Release Notes: Table of Contents

Program Input Options

Input	Output	No. Samples	Input Format	Output Format
Mutation List	Mutation classification Predicted ARV activity Mutation comments Mutation penalty scores	1	Text box Drop-down menu	HTML
DNA Sequence	Mutation classification Predicted ARV activity Mutation comments Mutation penalty scores Sequence quality control	1 to 10000	Text box Upload FASTA File	HTML Spreadsheet XML
Sierra Web Service 2.0	Mutation classification Predicted ARV activity Mutation comments Mutation penalty scores Sequence quality control	Unlimited	User script	JSON

Program HTML Output

HTML Output

HTML output contains the output for either one sequence or for multiple sequences. Reports for sequences contain a menu bar that allows the user to choose the report for a specific sequence. The HTML output includes the following information:

1. **Header:** This contains the SequenceID, which is the fasta header and a Date field containing the date the program was run
2. **Summary Data:** This section shows which residues in PR, RT, and/or IN were present in the submitted sequence and the closest matching subtype. This section also contains two buttons. The "Pretty pairwise" button displays how each gene of a sequence aligns to the consensus reference sequence. The "SDRMs" button indicates the surveillance DRMs present in the sequence.
3. **Sequence Quality Assessment:** This section contains figures for each gene in which each mutation is indicated by a bar. Blue bars indicate DRMs, black bars indicate differences from the consensus amino acid sequence, and red bars indicate problematic mutations. Hovering over the bar displays the mutation text. This section will also contain warnings if there are indicators of overall or localized poor sequence quality including the presence of stop codons, frame shifts, unusual insertions or deletions, APOBEC-mediated G-to-A hypermutation, and an excess of highly unusual mutations.
4. **Mutation Classification:** PR mutations are classified into Major DRMs, Accessory DRMs, and mutations that do not receive mutation penalty scores (Other). RT mutations are classified into NRTI DRMs, NNRTI DRMs, and Other. IN mutations are classified into Major DRMs, Accessory DRMs, and Other.
5. **Drug Resistance Interpretation:** For PR, drug-resistance interpretations are provided for each of the ritonavir-boosted PIs. For RT, interpretations are provided for seven NRTIs and four NNRTIs. For IN, interpretations are provided for the three FDA-approved INSTIs.
6. **Comments:** Comments are provided for (i) All DRMs with a mutation penalty score, (ii) Unscored mutations that have been associated with drug resistance but are considered to have minimal or no impact on currently used ARVs, and (iii) Highly unusual mutations at known drug-resistance positions that are not established DRMs.
7. **Scoring Table:** There is one table for each ARV class. The first column indicates each of the DRMs and DRM combinations that contributed to the overall penalty score for one or more ARVs. The remaining columns contain the penalty scores for the ARVs indicated in the column header. The total penalty score for each ARV -- obtained by adding each of the individual scores -- is shown in the column header.

Program Spreadsheet Output Files

Spreadsheet output files

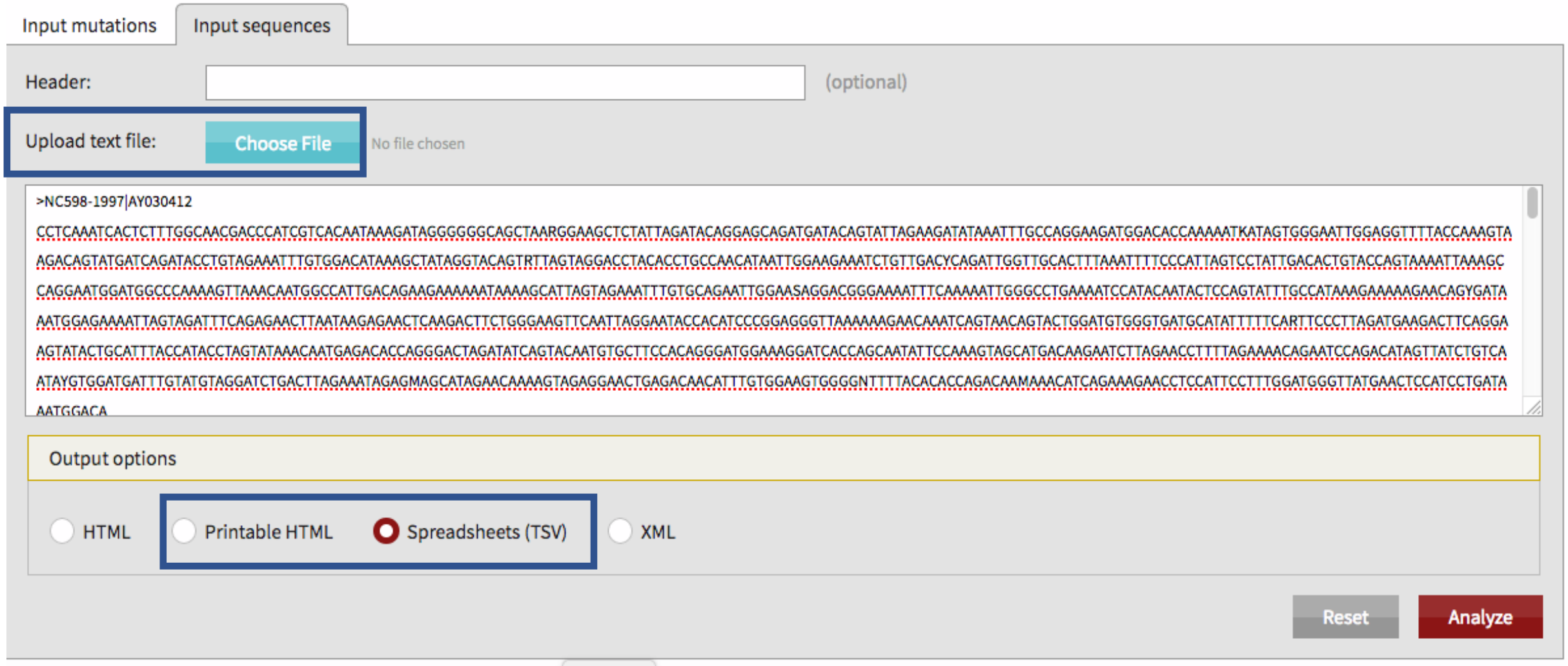
There are three types of spreadsheet / tabular output files for the HIVdb program: (i) Sequence summary; (ii) Resistance summary; and (iii) Formatted amino acid alignments for each gene. These files are useful for users submitting sets of sequences. These files contain tab-delimited text files that can readily be opened in Excel or compatible spreadsheet software. These files are downloaded into the user's download directory. If more than one output file is requested, the files are downloaded as a zip file.

Sequence summary

After the header row, each row contains one sequence. The fields are organized into the following types of information:

1. **SequenceID:** The fasta headers of the submitted sequences.
2. **Gene coverage:** The first and last residue of PR, RT, and/or IN.
3. **Subtype:** Subtype information including the best matching subtype and its genetic distance from one of 200 reference sequences. For more detailed information about the sequence references and decision making process, please refer to this page: HIV Subtyping Program.
4. **Percentage of ambiguities (Pcnt Mix):** Percentage of nucleotides with R (A/G), Y (C/T), M (A/C), W (A/T), S (G/C), or K (G/T).
5. **Mutation Classification:** PR mutations are classified into Major DRMs, Accessory DRMs, and mutations that do not receive mutation penalty scores (Other). RT mutations are classified into NRTI DRMs, NNRTI DRMs, and Other. IN mutations are classified into Major DRMs, Accessory DRMs, and Other. For each gene in a sequence, there are three comma-separated lists of mutations. Columns contain 'None' when there are no mutations belonging to the relevant classification. Columns contain 'NA' when the relevant gene was not sequenced.
6. **Surveillance Drug Resistance Mutations (SDRMs):** The SDRMs present in PR and RT.
7. **Additional treatment-selected mutations (TSMs):** TSMs are mutations that are non-polymorphic in ARV-naive individuals but occur with significantly increased frequency in ARV-experienced individuals. The most common TSMs are also DRMs. However, many TSMs are not established DRMs because they are either uncommon and/or they usually occur in sequences containing multiple DRMs and therefore have not been well studied.
8. **Sequence Quality Assessment:** For each gene, frame shifts, insertions and deletions, stop codons, mutations indicative of APOBEC-mediated G-to-A hypermutation, highly ambiguous nucleotides (B, D, H, V, N), and highly unusual amino acids.

HIVDB Program Landing Page



HIVDB Program: Input 10 Sequences. HTML Header

FOUND 10 SEQUENCES

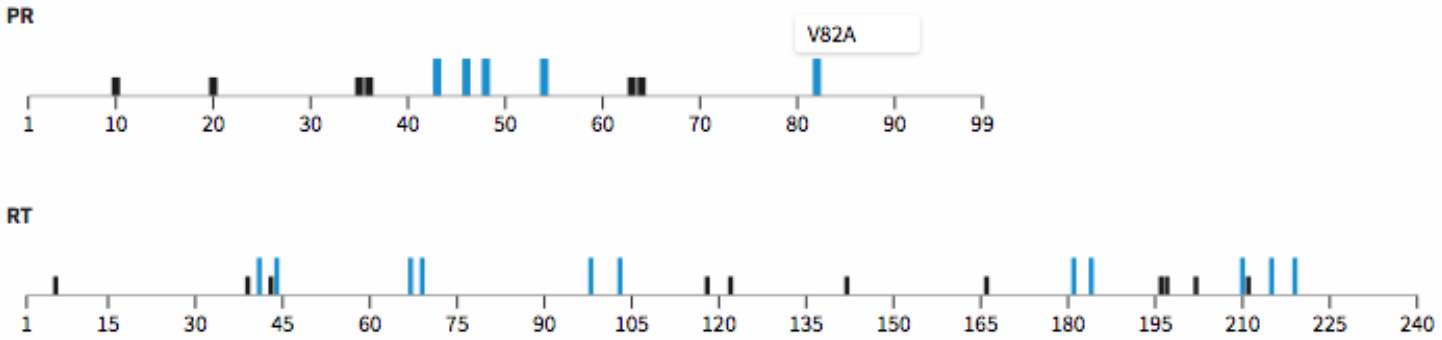
- **NC598-1997|AY030412**
- NC599-1997|AY030413
- NC602-1997|AY030414
- NC603-1997|AY030415
- NC4172-1997|AY031148
- NC798-1997|AY030428
- NC633-1997|AY030420
- NC636-1997|AY030421
- NC640-1997|AY030422
- NC2197-1998|

Sequence Summary

Sequence includes PR: codons 1 - 99
Sequence includes RT: codons 1 - 240
Subtype: ⊞ B (2.75%)

SDRMs | Pretty pairwise

Sequence Quality Assessment



HIVDB Program: Alignment, Subtype, Surveillance DRMs

Sequence Summary

SDRMs | Pretty pairwise

Sequence includes PR:	codons 1 - 99
Sequence includes RT:	codons 1 - 240
Subtype:	B (2.75%)

- D86069: France (1983); B (2.75%); best match
- AF256204: Spain (1989); B (2.85%)
- K03455: France (1983); B (2.95%)
- AF042100: Australia (1986); B (3.15%)
- EU839600: Haiti (2005); B (3.34%)
- U43096: Germany (1986); B (3.44%)
- DQ358805: Brazil (2002); B (3.44%)
- U63632: United States (1986); B (3.54%)
- AF156820: United States (1997); B (3.54%)
- EF694037: India (2006); B (3.54%)

PR SDRMs:	M46I, G48V, I54T, V82A
RT SDRMs:	M41L, D67N, T69D, K103N, Y181C, M184V, L210W, T215Y, K219N

Pretty pairwise of PR: Scroll right for more »

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
	P	Q	I	T	L	W	Q	R	P	L	V	T	I	K	I	G	G	Q	L	K	E	A	L	L	D
	CCT	CAA	ATC	ACT	CTT	TGG	CAA	CGA	CCC	ATC	GTC	ACA	ATA	AAG	ATA	GGG	GGG	CAG	CTA	ARG	GAA	GCT	CTA	TTA	GAT
	-	-	-	-	-	-	-	-	-	I	-	-	-	-	-	-	-	-	-	KR	-	-	-	-	-

Pretty pairwise of RT: Scroll right for more »

	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25
	P	I	S	P	I	E	T	V	P	V	K	L	K	P	G	M	D	G	P	K	V	K	Q	W	P
	CCC	ATT	AGT	CCT	ATT	GAC	ACT	GTA	CCA	GTA	AAA	TTA	AAG	CCA	GGA	ATG	GAT	GGC	CCA	AAA	GTT	AAA	CAA	TGG	CCA
	-	-	-	-	-	D	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-	-

HIVDB Program: PR Interpretation

Drug Resistance Interpretation: PR

PI Major Resistance Mutations:	**M46MI, G48V, I54T, V82A**
PI Accessory Resistance Mutations:	**K43T**
Other Mutations:	L10I, K20KR, E35D, M36I, L63P, I64V

Protease Inhibitors	
atazanavir/r (ATV/r)	High-Level Resistance
darunavir/r (DRV/r)	Susceptible
lopinavir/r (LPV/r)	High-Level Resistance

PR Comments

PI Major

- **M46I/L** are relatively non-polymorphic PI-selected mutations. In combination with other PI-resistance mutations, they are associated with reduced susceptibility to each of the PIs except DRV.
- **G48V** is a non-polymorphic mutation selected by SQV and, less often, by IDV and LPV. It confers high-level resistance to SQV, intermediate resistance to ATV, and low-level resistance to NFV, IDV and LPV.
- **I54A/T/S** are non-polymorphic PI-selected mutations that occur almost exclusively in viruses with multiple PI-resistance mutations. **I54A/T/S** are associated with reduced susceptibility to each of the PIs except DRV.
- **V82A** is a non-polymorphic mutation selected primarily by IDV and LPV. It reduces susceptibility to these PIs and contributes cross-resistance to each of the remaining PIs except DRV and TPV.

PI Accessory

- **K43T** is a non-polymorphic PI-selected accessory mutation. **K43T** is included in the Boehringer-Ingelheim TPV genotypic susceptibility score.

Other

- **L10I/V** are polymorphic, PI-selected accessory mutations that increase the replication of viruses with other PI-resistance mutations.
- **K20R** is a highly polymorphic PI-selected accessory mutation.

HIVDB Program: PI scores

Mutation Scoring: PR

PI	ATV/r	DRV/r	LPV/r
M46I	10	0	10
G48V	30	0	10
I54T	15	0	15
V82A	15	0	30
M46I + V82A	10	0	10
I54T + V82A	10	0	10
K43T	0	0	0
Total	90	0	85

HIVDB Program: RT Interpretation

Drug Resistance Interpretation: RT

NRTI Resistance Mutations:	**M41L, E44D, D67N, T69D, M184V, L210W, T215Y, K219KN**
NNRTI Resistance Mutations:	**A98G, K103N, Y181C**
Other Mutations:	E6D, T39A, K43EQ, V118VI, K122E, I142T, K166R, G196E, Q197QK, I202V, R211K

Nucleoside Reverse Transcriptase Inhibitors

abacavir (ABC)	High-Level Resistance
zidovudine (AZT)	High-Level Resistance
emtricitabine (FTC)	High-Level Resistance
lamivudine (3TC)	High-Level Resistance
tenofovir (TDF)	High-Level Resistance

Non-nucleoside Reverse Transcriptase Inhibitors

doravirine (DOR)	Intermediate Resistance
efavirenz (EFV)	High-Level Resistance
etravirine (ETR)	Intermediate Resistance
nevirapine (NVP)	High-Level Resistance
rilpivirine (RPV)	High-Level Resistance

RT Comments

NRTI

- **M184V/I** cause high-level in vitro resistance to 3TC and FTC and low-level resistance to ddI and ABC. However, **M184V/I** are not contraindications to continued treatment with 3TC or FTC because they increase susceptibility to AZT, TDF and d4T and are associated with clinically significant reductions in HIV-1 replication.
- **L210W** is a TAM that usually occurs in combination with M41L and T215Y. The combination of M41, **L210W** and T215Y causes high-level resistance to AZT and d4T and intermediate to high-level resistance to ddI, ABC and TDF.
- **T215Y** is a TAM that causes intermediate/high-level resistance to AZT and d4T, low-level resistance to ddI, and potentially low-level resistance to ABC and TDF.
- **K219N/R** are accessory TAMS that usually occur in combination with multiple other TAMs.
- **M41L** is a TAM that usually occurs with T215Y. In combination, **M41L** plus T215Y confer intermediate / high-level resistance to AZT and d4T and contribute to reduced ddI, ABC and TDF susceptibility.
- **E44D** is a relatively non-polymorphic accessory mutation and E44A is a nonpolymorphic accessory mutation. Each usually occurs with multiple TAMs.
- **D67N** is a non-polymorphic TAM associated with low-level resistance to AZT and d4T. When present with other TAMs, it contributes reduced susceptibility to ABC, ddI, and TDF.
- **T69D** is a non-polymorphic mutation that reduces susceptibility to ddI and possibly d4T.

NNRTI

- **K103N** is a non-polymorphic mutation that causes high-level reductions in NVP and EFV susceptibility.

HIVDB Program: NRTI and NNRTI Scores

Mutation Scoring: RT

NRTI	ABC	AZT	FTC	3TC	TDF
M41L	5	15	0	0	5
D67N	5	15	0	0	5
M184V	15	-10	60	60	-10
L210W	5	15	0	0	5
T215Y	10	40	0	0	10
K219N	5	10	0	0	5
M41L + E44D + L210W + T215Y	5	5	0	0	5
M41L + D67N + T215Y	5	5	5	5	5
M41L + L210W	10	10	0	0	10
M41L + L210W + T215Y	5	0	5	5	5
M41L + T215Y	15	10	5	5	10
D67N + T215Y + K219N	5	5	0	0	5
L210W + T215Y	10	10	5	5	10
T69D	0	0	0	0	0
Total	100	130	80	80	70

NNRTI	DOR	EFV	ETR	NVP	RPV
A98G	15	15	10	30	15
Y181C	10	30	30	60	45
A98G + Y181C	5	5	5	5	5
K103N + Y181C	10	0	0	0	0
K103N	0	60	0	60	0
Total	40	110	45	155	65