HIV Drug Resistance Testing Using the Stanford University Drug Resistance Database

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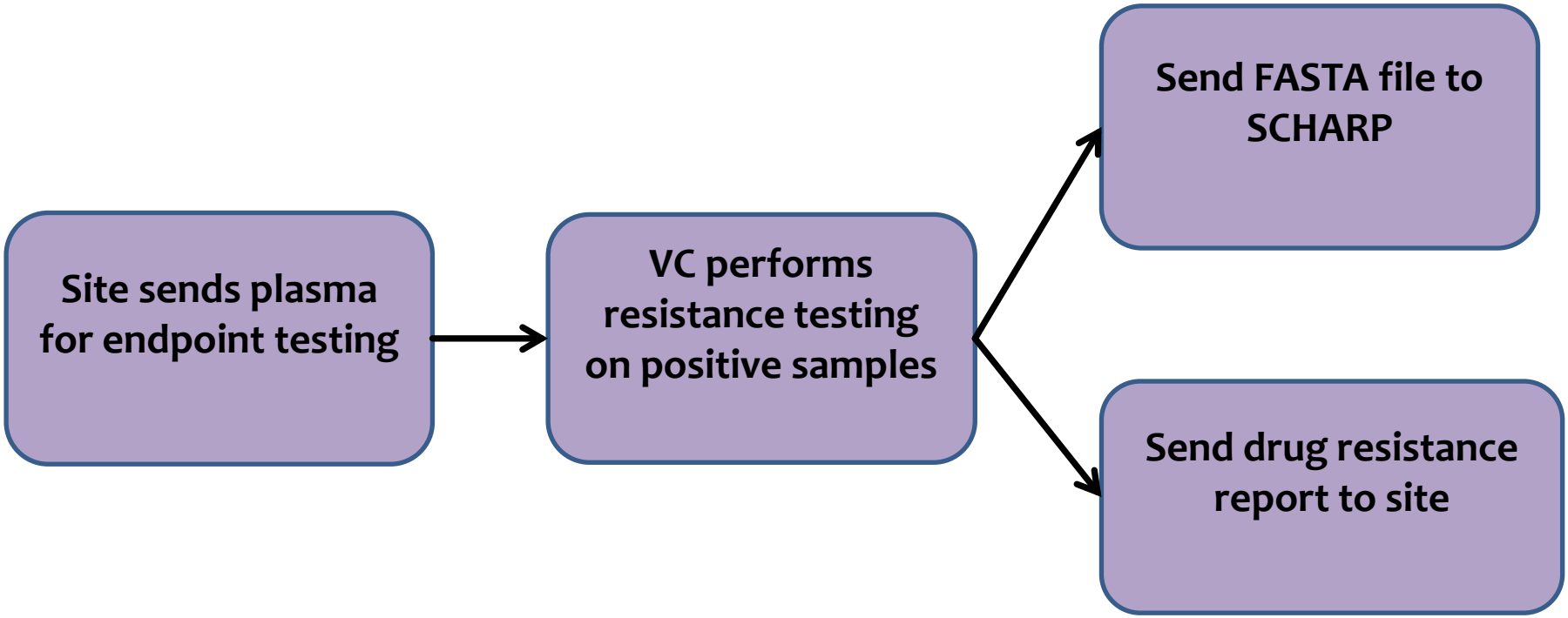
Overview

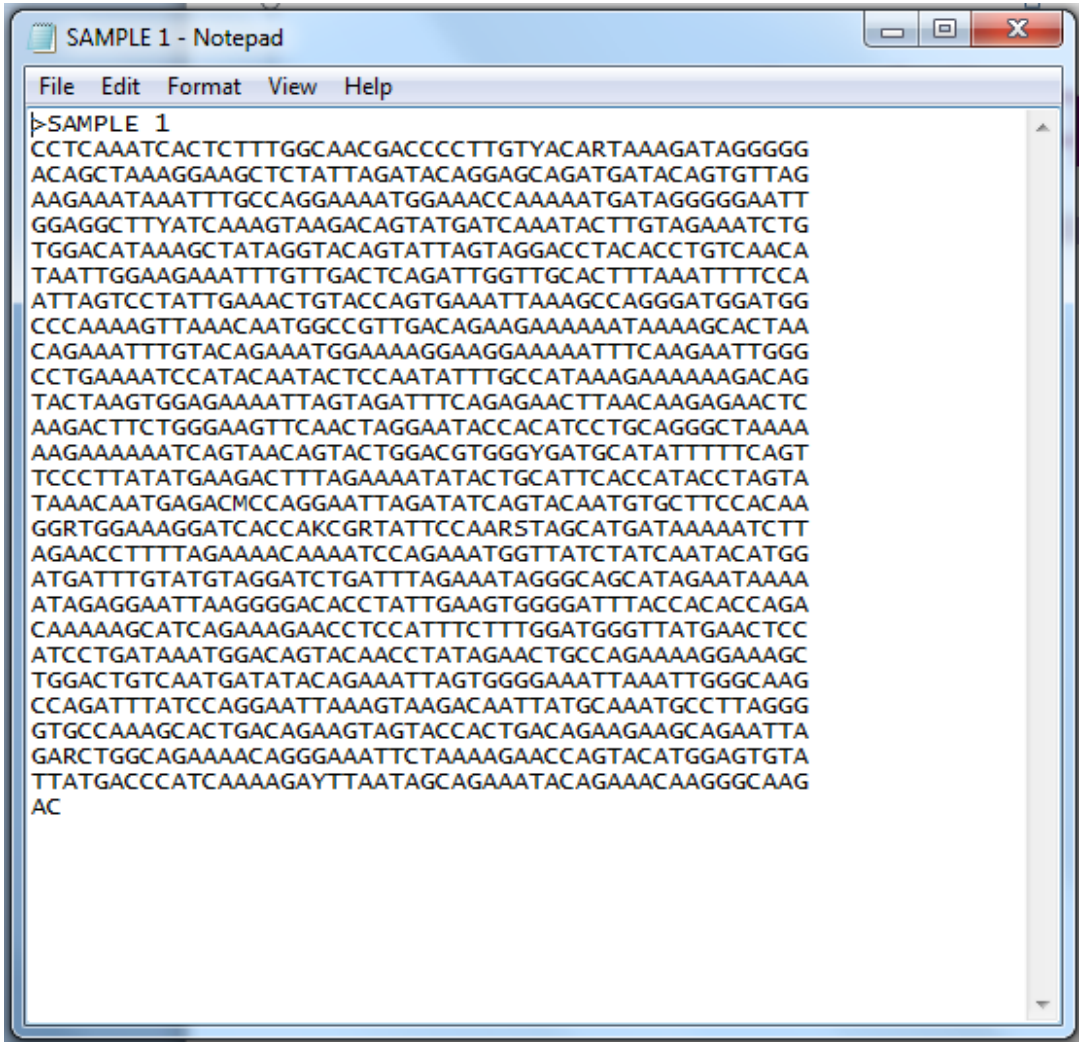
- How do we generate sequences at the VC?
- What is the Stanford Resistance Database?
- How do we generate the resistance report that is sent to sites?

Virology Core Resistance Testing



Final Result – FASTA file

- FASTA file



Stanford University HIV Drug Resistance Database Website

- An online database created from published HIV sequences.
- Three main types of content:
  1. Database queries and references
     - Info relating to genotype-treatment correlations, genotype-clinical outcome correlations, references, new submissions to database
  2. Interactive programs
     - HIVdb program, HIValg program, HIVseq program, ART-AiDE, Rega HIV-1 subtyping tool
  3. Educational resources.
     - Drug resistance summaries, surveillance drug resistance mutation list

Stanford Database User Guide- http://hivdb.stanford.edu/pages/documents.html

Interactive Programs

- HIVdb Program
  1. May enter a list of mutations
  2. May enter a complete sequence containing protease, RT and/or integrase
- VC uses option 2 to generate drug resistance report which contains:
  1. **Summary Data**
  2. **Sequence Quality Assessment**
  3. **Drug resistance Interpretation**
  4. **Mutation scoring table**

Stanford Database User Guide- http://hivdb.stanford.edu/pages/documents.html

Drug Resistance Report

- Stanford HIV Drug Resistance Database Output
    1. **Summary Data** → Defines sample subtype, region of HIV, QC summary

Summary Data

Sequence includes PR: codons: 1 - 99
Sequence includes RT: codons: 1 - 334

There are no insertions or deletions

Subtype and % similarity to closest reference isolate:

1. *PR: D (97.0%)*
2. *RT: D (95.9%)*

http://hivdb.stanford.edu/pages/documents.html

Drug Resistance Report

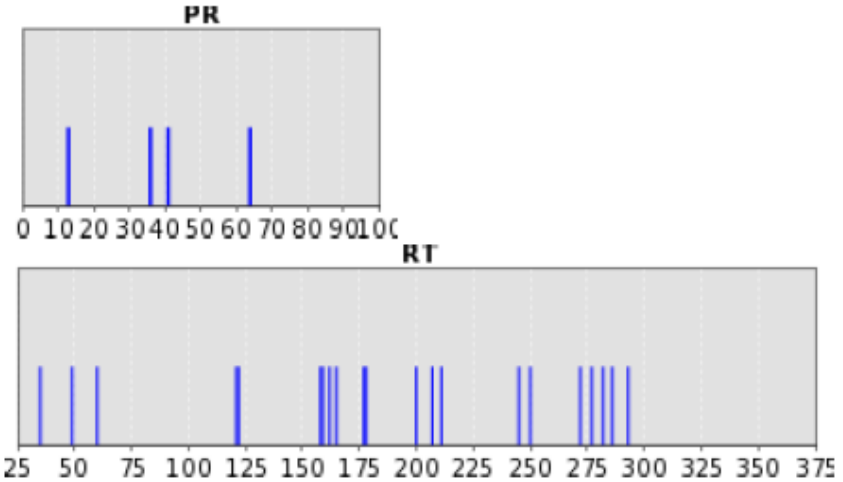
- Stanford HIV Drug Resistance Database Output

  2. Sequence Quality Assessment → a quality assessment sequence analysis (insertions, deletions, unusual bases)

Sequence Quality Assessment

Gene	QA Problem	Codons
PR	Stop Codons, Frame Shifts:	*None*
PR	Ambiguous Positions:	*None*
PR	Unusual Residues:	*None*

Gene	QA Problem	Codons
RT	Stop Codons, Frame Shifts:	*None*
RT	Ambiguous Positions:	*None*
RT	Unusual Residues:	*None*



Blue lines indicate differences from consensus B; tall blue lines indicate sites associated with drug resistance. Red lines indicate QA problems.

http://hivdb.stanford.edu/pages/documents.html

Drug Resistance Report

- Stanford HIV Drug Resistance Database Output

  3. Drug resistance Interpretation → Scans sequence for any mutations that are associated with 19 commonly used protease, nucleoside reverse transcriptase and non-nucleoside reverse transcriptase inhibitors.

Drug Resistance Interpretation: PR

PI Major Resistance Mutations:	None
PI Minor Resistance Mutations:	None
Other Mutations:	I13IV, M36I, R41K, I64V

Protease Inhibitors	
atazanavir/r (ATV/r)	Susceptible
darunavir/r (DRV/r)	Susceptible
fosamprenavir/r (FPV/r)	Susceptible
indinavir/r (IDV/r)	Susceptible
lopinavir/r (LPV/r)	Susceptible
nelfinavir (NFV)	Susceptible
saquinavir/r (SQV/r)	Susceptible
tipranavir/r (TPV/r)	Susceptible

PR Comments

Drug Resistance Interpretation: RT

NRTI Resistance Mutations:	None
NNRTI Resistance Mutations:	None
Other Mutations:	V35T, K49R, V60I, D121Y, K122E, A158AS, I159IV, S162AGST, T165I, D177E, I178M, T200I, Q207G, R211K, V245E, D250E, A272P, K277R, L282C, T286A, I293V

Nucleoside RTI		Non-Nucleoside RTI	
lamivudine (3TC)	Susceptible	efavirenz (EFV)	Susceptible
abacavir (ABC)	Susceptible	etravirine (ETR)	Susceptible
zidovudine (AZT)	Susceptible	nevirapine (NVP)	Susceptible
stavudine (D4T)	Susceptible	rilpivirine (RPV)	Susceptible
didanosine (DDI)	Susceptible		
emtricitabine (FTC)	Susceptible		
tenofovir (TDF)	Susceptible		

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Drug Resistance Report

- Stanford HIV Drug Resistance Database Output

  4. Mutation scoring → each mutation is given a score based on Stanford's internal algorithm. The score determines the level of resistance.

  (higher score = greater resistance)

Genotypic Score	
0 – 9	Susceptible
10 – 14	Potential Low-Level Resistance
15 – 29	Low-Level Resistance
30 – 59	Intermediate Resistance
≥ 60	High-Level Resistance

Mutation Scoring

PR	ATV/r	DRV/r	FPV/r	IDV/r	LPV/r	NFV	SQV/r	TPV/r
Total:	0	0	0	0	0	0	0	0

RT	3TC	ABC	AZT	D4T	DDI	FTC	TDF	EFV	ETR	NVP	RPV
K103N	-	-	-	-	-	-	-	60	0	60	0
Y188L	-	-	-	-	-	-	-	60	15	60	60
Total:	0	0	0	0	0	0	0	120	15	120	60

http://hivdb.stanford.edu/pages/documents.html

Examples

http://hivdb.stanford.edu/