

geno2pheno[ngs-freq]: a web service for identifying drug resistance in HIV-1 and HCV next-generation sequencing samples

Matthias Döring¹, Joachim Büch¹, Georg Friedrich¹, Alejandro Pironti¹, Prabhav Kalaghatgi¹, Elena Knops², Eva Heger², Martin Obermeier³, Martin Däumer⁴, Alexander Thielen⁴, Rolf Kaiser², Thomas Lengauer¹ and Nico Pfeifer^{1,5,6}

¹Max Planck Institute for Informatics,
Saarland Informatics Campus

²Institute of Virology,
University Hospital Cologne

³MVZ Medizinisches
Infektionsszentrum (MIB),
Berlin

⁴Seq-IT,
Kaiserslautern

⁵Department of Computer Science,
University of Tübingen

⁶Medical Faculty,
University of Tübingen

Background

Genotypic resistance testing based on Sanger sequencing is routinely used for the determination of drug resistance in patients infected with viruses such as HIV-1 or HCV. Since the level of detection of Sanger sequencing typically ranges from 15% to 20% [1], resistant variants that are present only at low population frequencies (minority variants) may be missed. Next-generation sequencing (NGS), enables the identification of resistance mutations even at low abundances of 1% [2]. Thus, genotypic resistance testing based on NGS data provides additional information that can improve clinical decision making. By considering minority resistant variants that are associated with treatment failure, it is possible to improve treatment outcomes [3]. Likewise the detection of minority resensitizing mutations could allow for the selection of drug combinations with enhanced synergy.

Existing web services for the interpretation of drug resistance based on NGS data such as PASeq and HyDRA [4] requires the input of raw sequencing data whose processing typically exceeds half an hour per sample. These services provide only rules-based interpretations. Here, we present geno2pheno[ngs-freq], a genotypic resistance interpretation system for NGS samples from HIV-1 and HCV. For samples from HIV-1, the statistical models of geno2pheno[resistance] are used, while predictions for HCV samples are based on the rules-based approach of geno2pheno[hcv]. By relying on the input of frequency files rather than raw NGS data, geno2pheno[ngs-freq] can provide results within seconds.

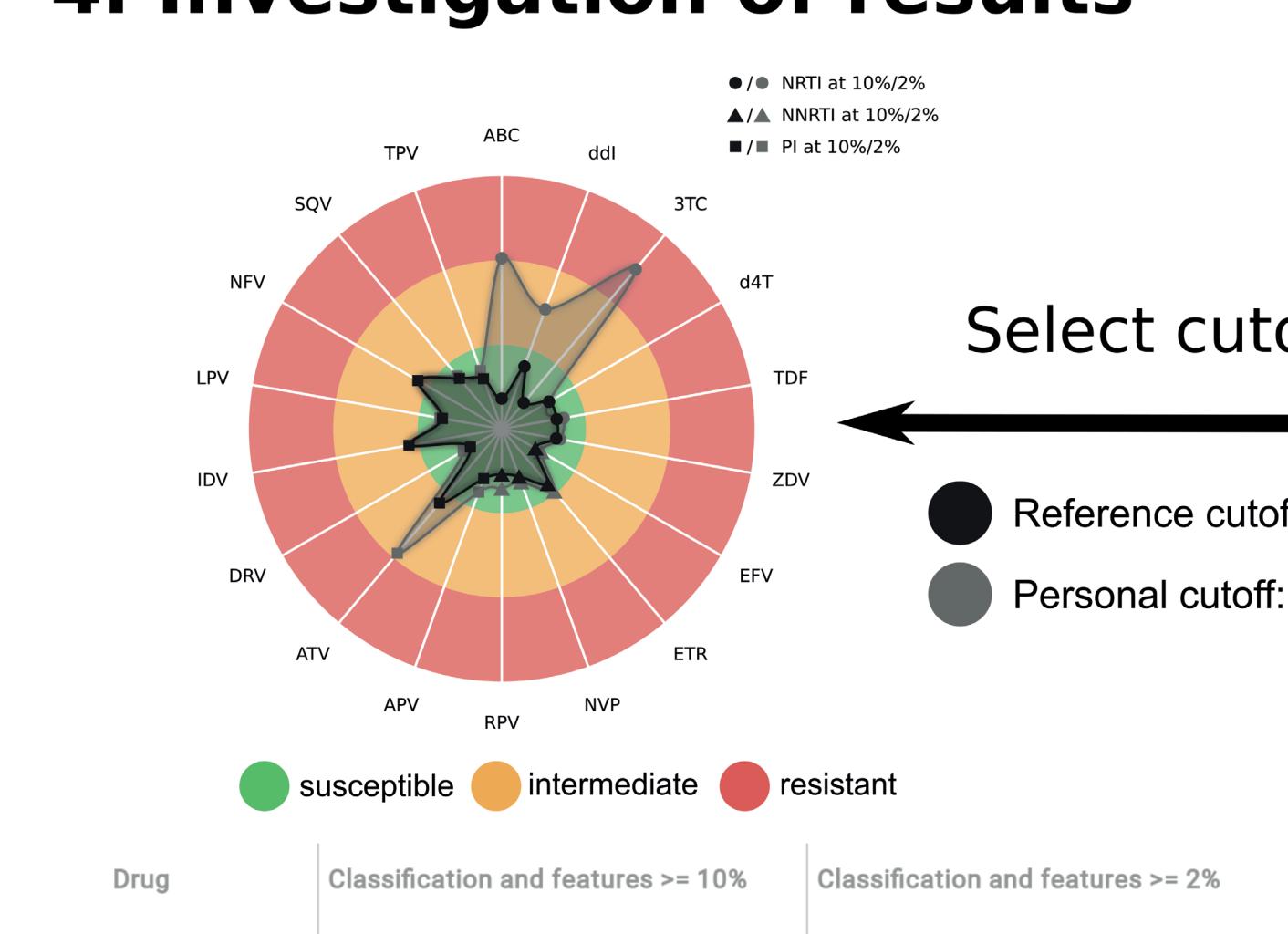
Workflow

1. Input of frequency files

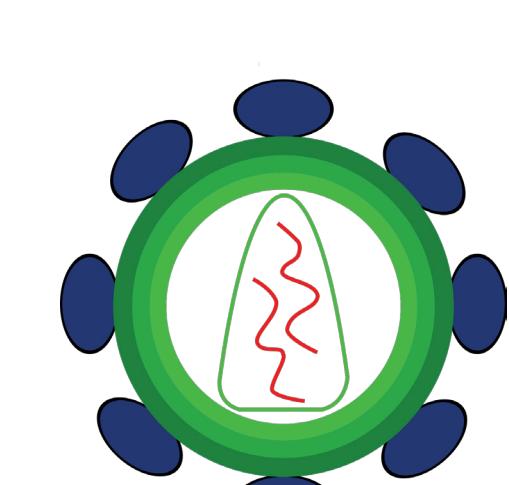
pos	1	2	3	4	5	6	7	8	9	10
A	5	4	18	4	2	16011	12041	15780	2000	10
C	14964	14862	15359	46	12959	2	2	0	61	16016
G	6	1	0	3	500	35	3989	129	1	0
T	18	15	1000	15517	11	31	38	4	15782	14



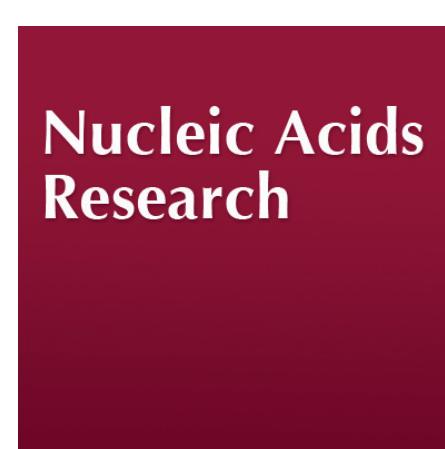
4. Investigation of results



Availability

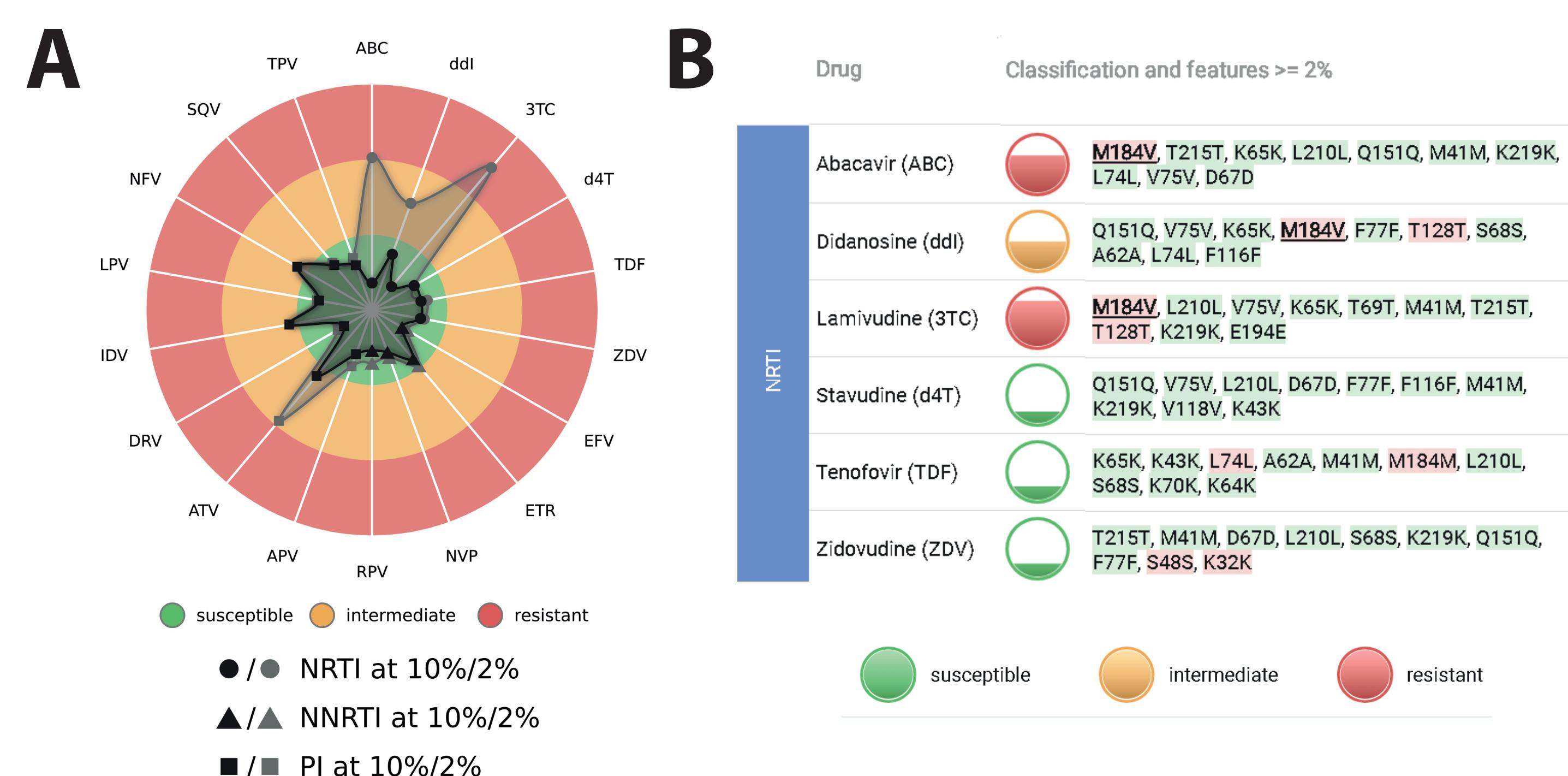


geno2pheno[ngs-freq] is freely available via geno2pheno.org



Manuscript available at NAR:
<https://doi.org/10.1093/nar/gky349>

Case Study: HIV-1 NRTI Resistance



(A) Radar plot of estimated HIV-1 drug resistance. The two points on each axis indicate standardized estimated resistance factors for individual drugs based on two cutoffs. The inner surface shows the estimated level of resistance for the consensus sequence at a cutoff of 10%, while the outer surface indicates the level of resistance for the consensus sequence at a cutoff of 2%. The three colored circle sectors indicate the estimated levels of drug resistance, from inside to outside: green for susceptibility, orange for intermediate resistance, and red for resistance. (B) Table of model features for the consensus sequence at a cutoff of 2%. Circles indicate levels of drug resistance. Displayed features are ordered with decreasing impact on predictions. Features with a red background increase the level of resistance, features with a green background decrease the level of resistance. Mutations are shown in bold. Underlined features are only present in the minor viral population (i.e. at a prevalence < 10%).

Interpretation

- M184V at 2.36% prevalence
- Possible treatment: TDF + FTC + DRV

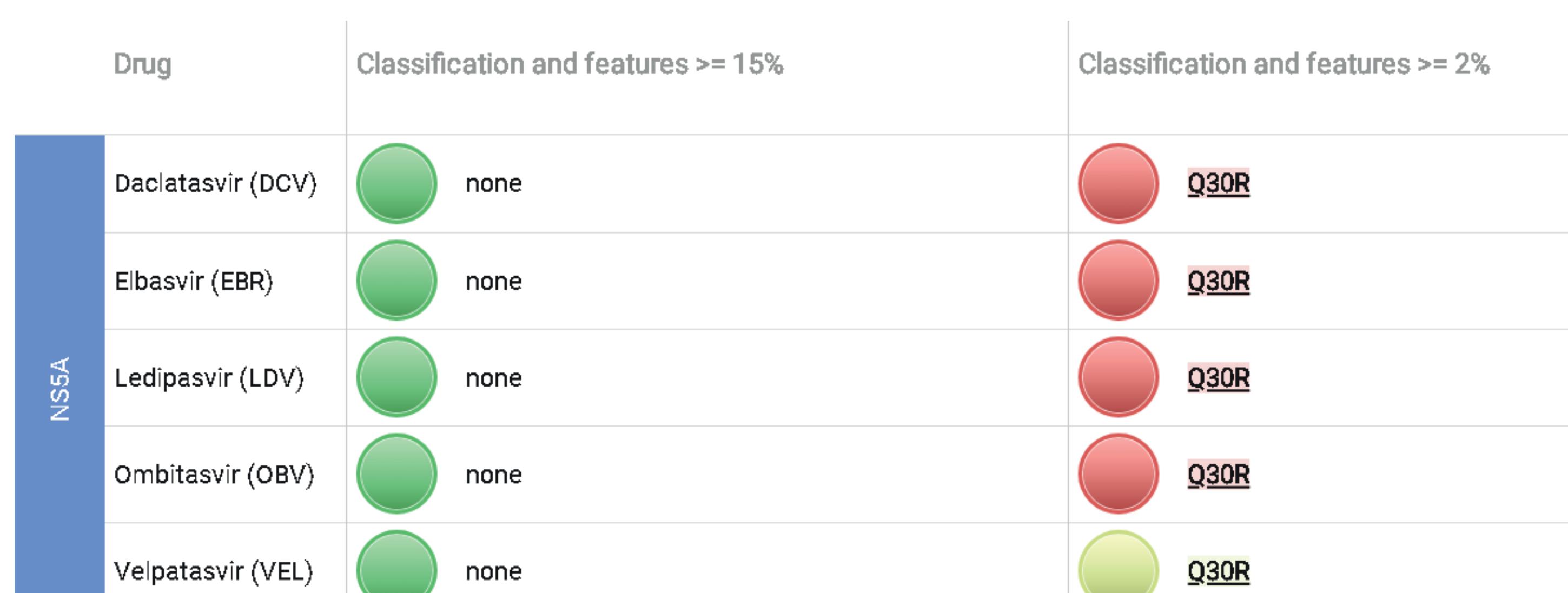
Context

- M184V enhances susceptibility to ZDV, d4T, and TDF
- Impact on activity of FTC:
 - *In vitro*: inactive [5]
 - *In vivo*: residual activity [6]

Implications

- FTC stabilizes M184V
- M184V ensures susceptibility to TDF
- FTC would still be residually active even if M184V were to proliferate

Case Study: HCV NS5A Resistance



Interpretation

- Q30R at 6.13% prevalence
- Possible treatment: VEL
- DCV, EBR, LDV, or OBV could lead to failure
- VEL should be fully active

References

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