

580-T Incidence and Nature of Phenotype-Genotype Discordance in a Clinical Setting: Maximizing the Utility of Resistance Testing.

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ABSTRACT

Background: Resistance testing, using either genotype (GT) or phenotype (PT), has become standard of care in the management of HIV treatment failure. GT relies on accurate interpretation algorithms, while PT provides quantitative data but requires a clinical cut-off value for maximum utility.

Methods: GT and PT tests were performed on approximately 200 patients participating in a pilot program to evaluate the impact of combining both resistance assays in a single report (PhenoSenseGT). Genotypic interpretations were based on an updated algorithm reflecting state of the art knowledge. Discordance was defined for drugs with a fold-change (FC) in IC₅₀ over the PhenoSenseTM assay cut-off but scored as GT sensitive (PT-R/GT-S), or vice versa (PT-S/GT-R).

Results: Most patients were highly treatment-experienced (85% had 2 or more treatment failures). PT/GT discordance was commonly observed: in 75, 54, 33, and 22% of samples at least 1, 2, 3 or 4 drugs, respectively, were discordant. The drugs with discordance in over 10% of samples were ddI (37%), ddC (25%), ABC (38%), 3TC (16%), SQV (15%), APV (12%), LPV (12%), IDV (12%), d4T (11%), and RTV (10%). Two-thirds of the PT-S/GT-R results were associated with mixtures at resistance-associated positions. After accounting for this, only ddI (29%), ddC (20%), 3TC (14%), ABC (14%) and APV (11%) had discordance rates over 10%. For ddI and ddC, most of the discordance was related to the presence of the M184V mutation, which causes GT to be called resistant, although not all have FC > 1.7. For SQV, many samples with the L90M mutation, which is interpreted as GT resistance when present with at least two secondary mutations, retained susceptibility to SQV (FC < 2.5). PT-R/GT-S was observed most commonly for 3TC, APV, and LPV. In the case of 3TC, PT loss of susceptibility (FC > 2.5) was frequently observed in samples with many "ZDV mutations" but lacking M184V, K65R, or E44D/V118I. For APV and LPV, complex patterns of PI mutations that did not meet the resistance criteria in the GT algorithm led to PT FC values > 2.5 or 10-fold, respectively.

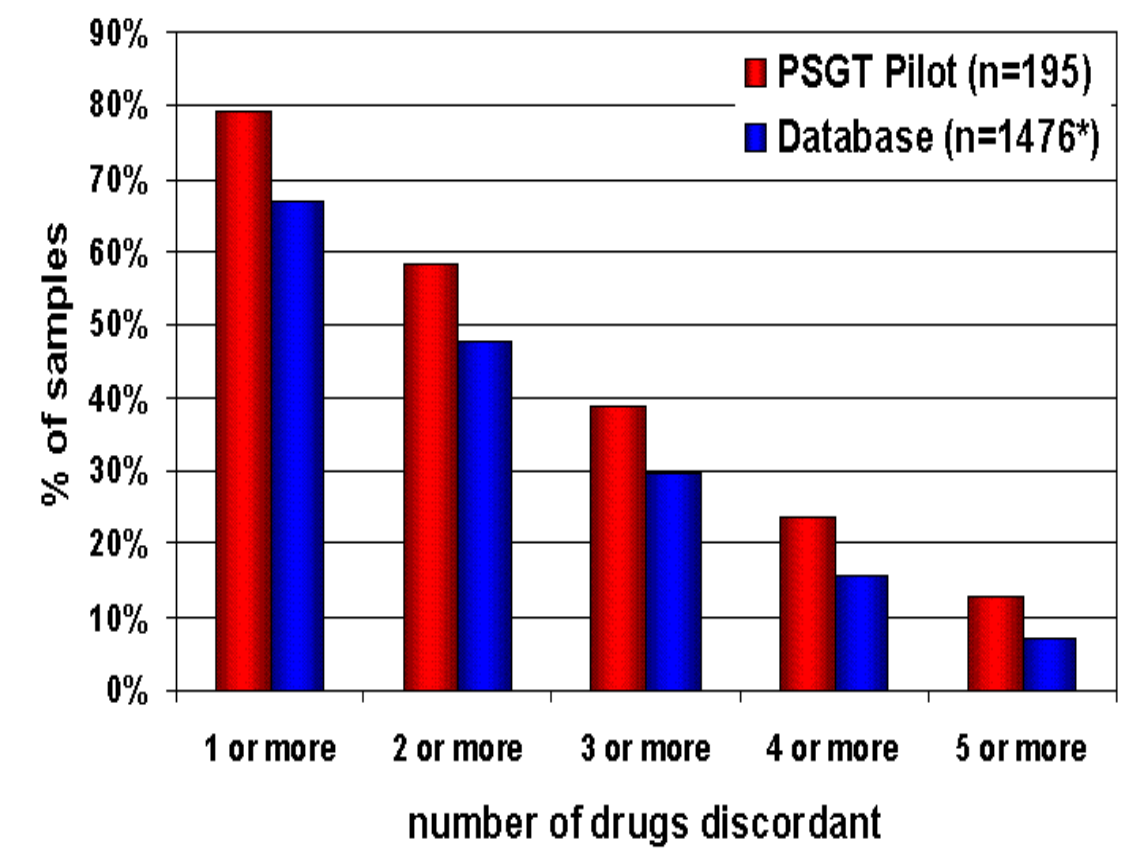
Conclusions: In this pilot program of combined PT/GT, the majority of samples showed PT/GT discordance in at least 2 of 15 drugs. GT results added valuable information when mixtures helped to explain the PT-S/GT-R result, while PT was important for interpretation of PT-R/GT-S results and for providing quantitative assessment of the degree of resistance.

PHENOSENSE GT REPORT FORM

PHENOSENSE GT PILOT PROGRAM

- Genotype and phenotype can provide complementary information
- Many physicians order both, often from separate labs
- Need to combine both test results in one report and provide interpretation where needed ("PhenoSenseGT" report)
- Test marketed 7/01-11/01
- 195 patients tested (29 physicians)
- Majority of patients heavily treatment experienced: 85% failed 2 or more previous HAART regimens

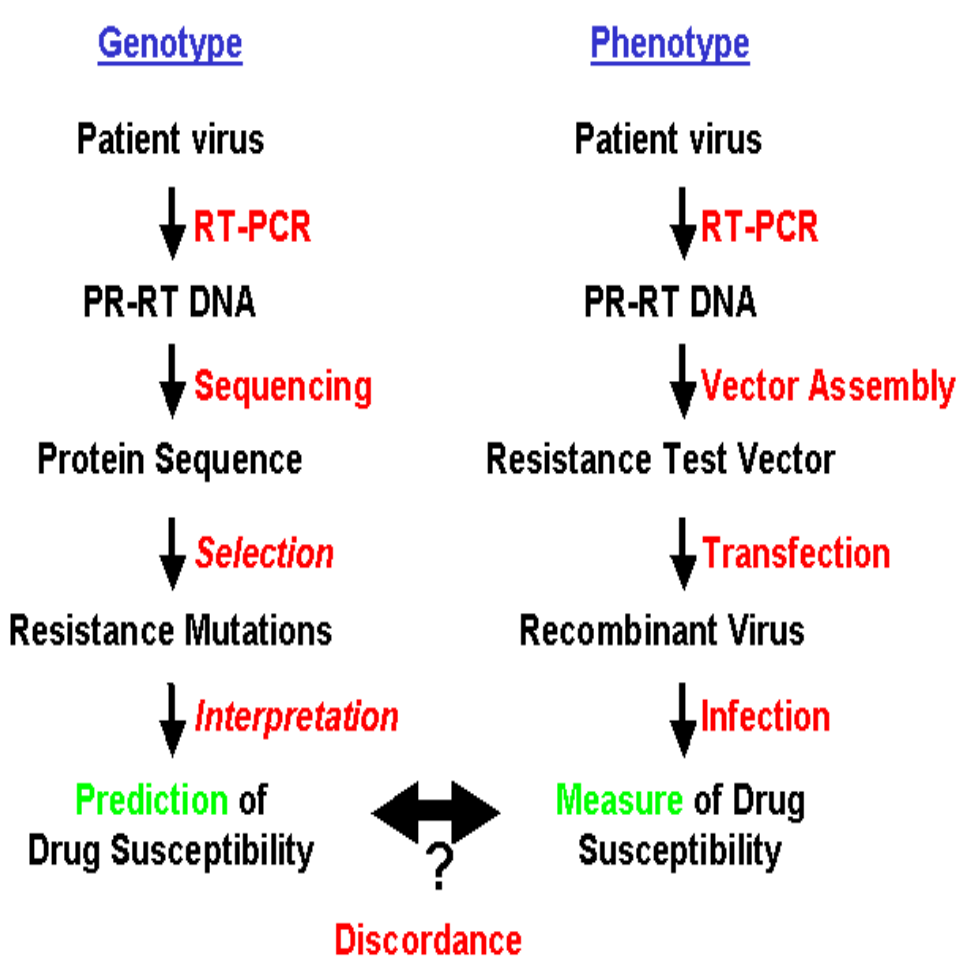
INCIDENCE OF DISCORDANCE



CAUSES FOR DISCORDANCE (EXCLUDING MIXTURES)

- Note: If discordance was possibly explained by mixtures or by another cause, it was assumed to be due to mixtures
- ddI, ddC: 96% and 77%, respectively, of PT-S/GT-R samples (n=47 and 31, respectively) have M184V. 4% and 13%, respectively, have L74V. The remaining ddC discordance is associated with T69D.
- TFV, d4T: 89% and 87%, respectively, of PT-S/GT-R samples (n=19 and 8, respectively) explained by suppression by M184V
- ZDV: all PT-S, GT-R samples (n=13) explained by suppression, either by M184V (62%), combinations of M184V with L74V and L100I, or L74V and Y181C (15% each), or Y181C alone (8%)
- SQV, IDV: 45% and 67%, respectively, of PT-S/GT-R samples (n=11 and 9, respectively) have L90M. 36% and 11%, respectively, have V82A, F, S or T.
- 3TC, TFV samples that are PT-R, GT-S: see poster 569-T (NRTI cross-resistance)
- LPV discordance: see poster 581-T (incomplete rules)

DISCORDANCE MAY BE CAUSED BY INACCURATE GENOTYPE INTERPRETATION



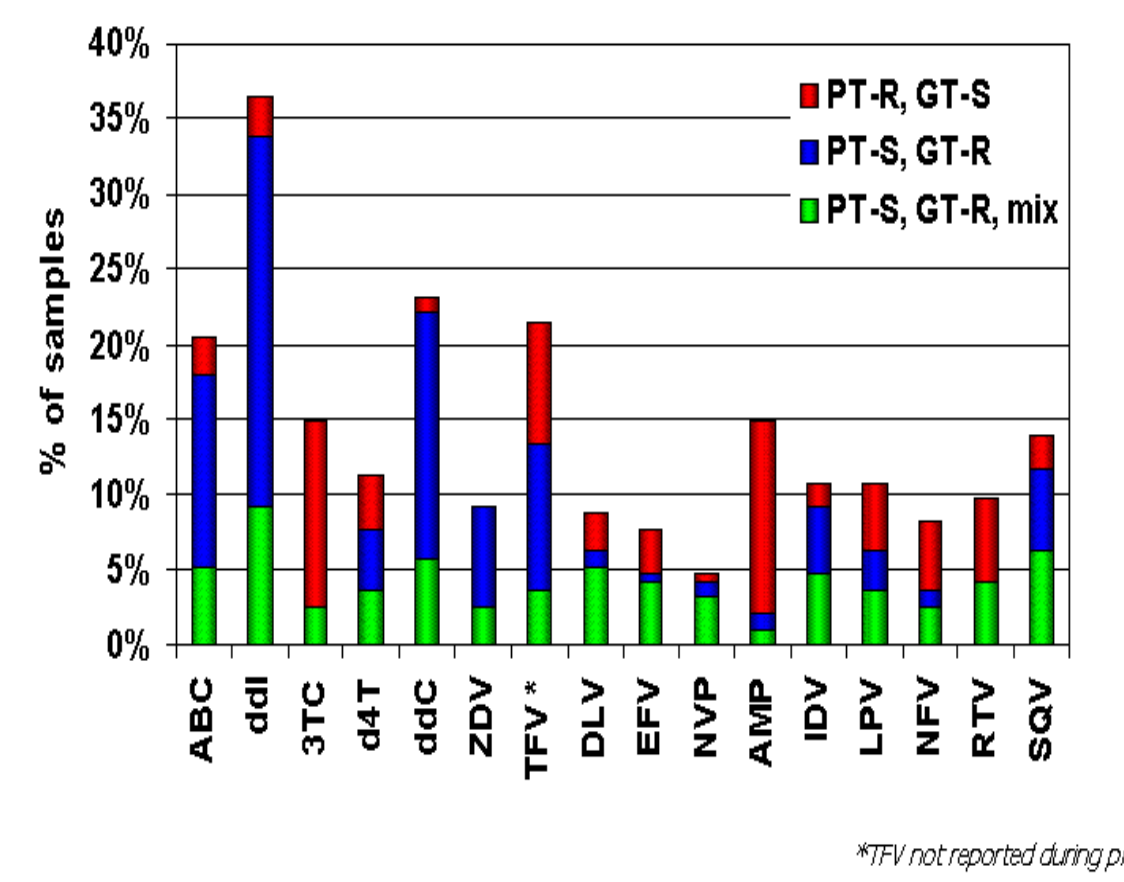
EXAMPLES OF DISCORDANCE

A. PT-R, GT-S discordance

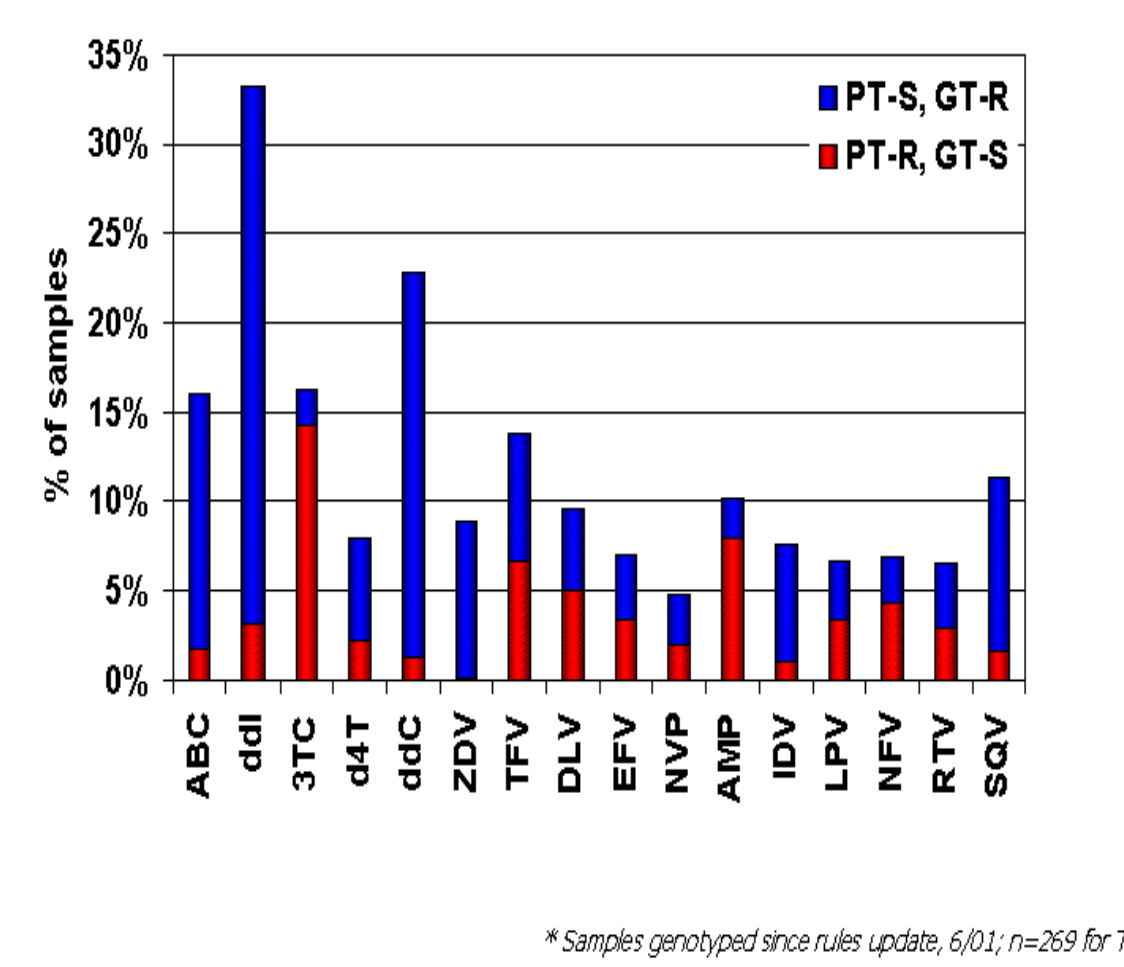
B. PT-S, GT-R, no mixtures

C. PT-S, GT-R, mixtures

DISCORDANCE RATES BY DRUG (PSGT PILOT, n=195)



DISCORDANCE RATES BY DRUG (VIROLOGIC DATABASE, n=1476*)



TYPES OF PHENOTYPE-GENOTYPE DISCORDANCE

- A Phenotype resistant, Genotype sensitive (PT-R/GT-S)**
 - Cross-resistance
 - New mutations
 - Incomplete rules
- B Phenotype sensitive, Genotype resistant (PT-S/GT-R), no mixtures**
 - Suppression or "re-sensitization"
 - Mutations associated with clinical failure but insufficient to cause phenotypic resistance on their own
 - Incomplete rules
- C Phenotype sensitive, Genotype resistant (PT-S/GT-R), mixtures present**
 - PT and GT test population of viruses present in plasma
 - Sometimes genotype is more sensitive e.g. M184V and 3TC
 - Sometimes phenotype is more sensitive e.g. G190S and NVP/EFV

CONCLUSIONS

- PT-GT discordance common
- PT contributes information that cannot be acquired with GT alone (quantitative, measures the effect of each drug on the folded 3-D structure of the viral enzymes, whereas GT relies on imperfect and incomplete interpretation algorithms)
- New drugs benefit most from phenotype
- Genotype helps most when mixtures are present, but may also provide an indication of potential for development of resistance in the future

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