Genotypic resistance testing

yes or **NO**

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Why do we use Resistance Testing?

- Baseline resistance predicts virologic response
- Resistance testing may tell us whether treatment failure is due to viral drug resistance vs. other factors
- We expect it to assist us in the choice of the optimal regimen

Why Do We Use Resistance Testing?

- **Resistance testing is in the treatment guidelines**
- **7 prospective trials have been reported and shown some benefit, however,**
  - **methodology not consistent** : patient population, assays, transmission of test results, interpretation/evaluation, expert advice, treatment choices available

Disadvantages of genotype

- Indirect measurement of susceptibility
- Disregards mutational interactions
- Result needs expert interpretation

Technical limitations of genotypic resistance testing

- **Unreliable or less reliable at low viral load**
  (low copy number may bias genotype sampling)
- **Lack of standardisation of technologies**
- **Underdiagnosis of resistance mutations even in complete mutant population**
- **Insensitive to minor species**
  - 50% mutant population only consistently detected by approximately 50% of the laboratories
  - 25% mutant can not be detected consistently
- **Inter-laboratory differences of results are extensive**

Biological limitations of genotypic resistance testing

- **Tests performed off-therapy can be misleading**.Genotype may not detect resistance to drugs not currently being given.
- **Tests are better at predicting inactive drugs than identifying active ones.**
- **Test results may be influenced by HIV subtype**.
- **Interpretation of test results is complex**.

Prospective studies of resistance guided therapy

Study	Design	Primary endpoint (ITT)	
VIRADAPT	G Vs SOC	ΔVL W12: -1.04 vs -0.46	(diff: 0.58 log)
		ΔVL W12: -1.15 vs -0.67	(diff: 0.48 log)
GART	G+EA SOC	ΔVL W4+8: -1.9 vs 0.61	(diff: 0.85 log)
KAISER	P vs SOC	ΔVL W12: -0.2 log vs -0.4 log	
VIRA 3001	P vs SOC	%<400 W24: 45% vs 34%	(diff: 11%)
NARVAL	P vs G vs SOC	%<200 W12: 35% vs 44% vs 36%	
HAVANA	G vs SOC	%<400 W24: 49% vs 36%	(diff: 13%; p<0.05)
	EA+vs EA-	%<400 W24: 47% vs 37%	(diff: 10%; p=NS)
ARGENTA	G+EA vs SOC+EA	%<500 W12: 27% vs 12%	(diff: 15%)
CCTG575	P vs SOC	%<400: 48% vs 48%	
		ΔVL: -0.71 vs -0.69	

Prospective studies : Why such a limited benefit

- **Modest short-term virological benefit seen in arms either genotype or phenotype**

- **Differences between resistance arms and SOC**
  - **△VL - 0.5-0.6 log**
  - **patients with VL<200-500 copies/ml = 15%-20%**
  - **modest benefit in part due to improper interpretation of genotypic results (algorithms) and unknown clinical cut-off for phenotypes**
  - **cross-resistance with present drugs: lack of new active drugs**
  - **possible role of undetected resistant minority species**

Genotypic (and phenotypic) resistance testing: usefull or not?

NARVAL

- Large, randomized prospective study comparing Genotyping, Phenotyping and Standard of Care
- Heavily pretreated population enrolled
- **No virological benefit of phenotyping**
- **Benefit to genotyping only in some of endpoints.**
- **No benefit in heavily PI-experienced patients.**

Narval : Possible conclusions

- **Resistance testing doesn't work in France**

  or …

  French clinicians are excellent …!

- **Resistance testing has probably limited benefit in salvage therapy**

  nevertheless …

  it may result in fewer drug being used for the same effect, thus saving options

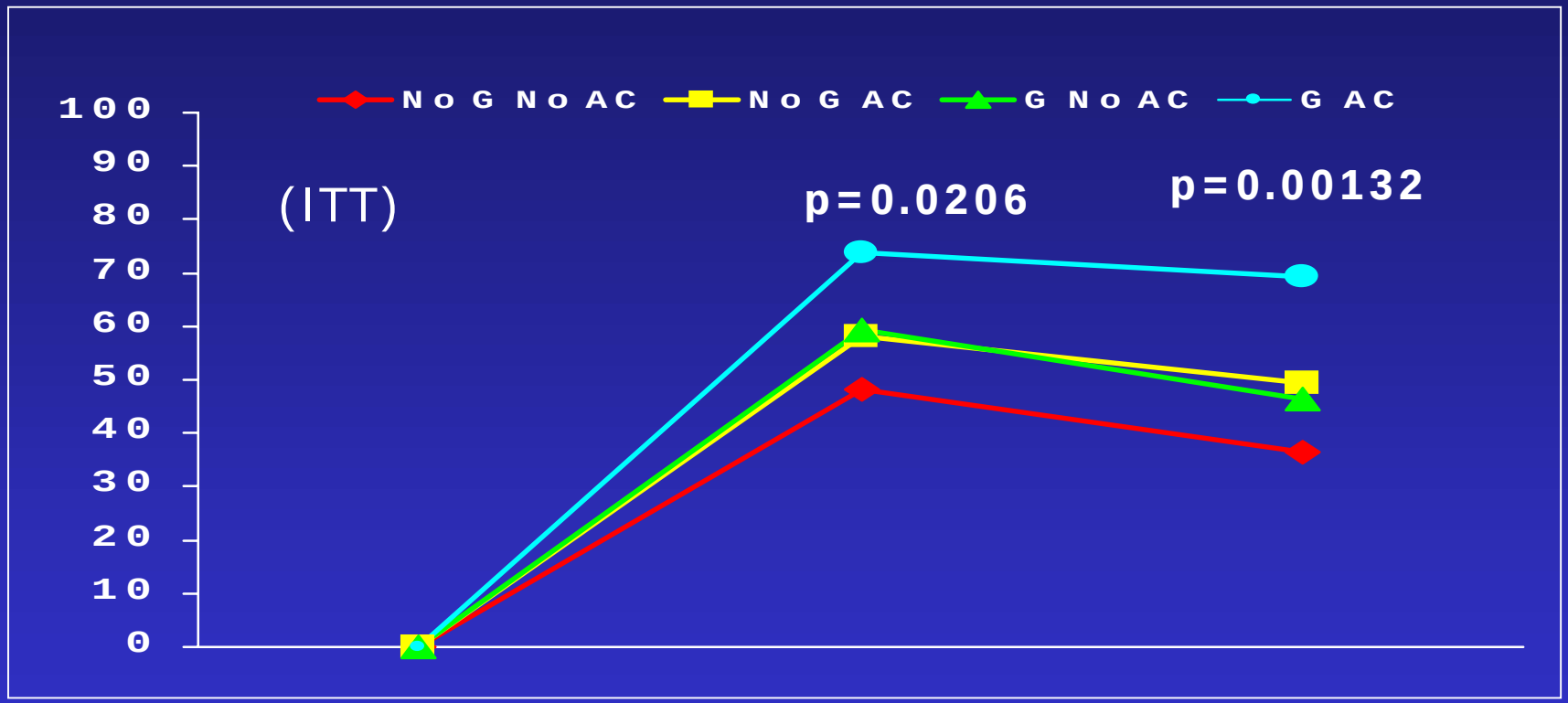
(Adapted from E. Van Wijngaerden)

Havana : study design

- Randomized, prospective, multicenter study
- Randomization 1 : Genotyping vs. SOC
- Randomization 2 : with or without Expert Advice
- Stratified by ARV experience

Havana : results (1)

% of Patients with HIV-1 RNA <400 copies/ml



	BL	Wk12	Wk24
NO G/ NO AC *(N=77)*		*48.1%*	*36.4%*
NO G/ AC *(N=67)*		*58.2%*	*49.3%*
G NO AC *(N=69)*		*59.4%*	*46.4%*
G/AC *(N=65)*		*73.8%*	*69.2%*

What can we do with the Havana results ?

Do genotyping, then sit together and think

- Past ARV History
- Adverse Events/Intolerance
- Adherence
- CD4/VL
- Concomitant medications
- Genotype

(adapted from E. Van Wijngaerden)

What else can we do with the Havana results ?

Sit together and think

- Adherence
- Adverse Events / Intolerance
- CD4 / VL
- Past ARV History
- Concomitant medications

Perform genotype if indicated

and …

Sit together and think again…!

What are We Receiving From the Resistance Assay Report ?

Does the Resistance Assay report show us raw data, interpretation of the data or actual clinical advice ?

- **Raw Data** : List of Mutations (L90M, V82A)
- **Interpretation** : What this list of mutations means for each drug (Sensitive, Resistant)
- **Clinical Advice** : Which drugs are now best to give our patient

Interpretation of Genotypic Reports

Interpretation tells us in concept :

« *if you have this mutation and you receive this drug, this will be your response* »

Interpretation actually tells us :

« *if you have these mutations and you receive this drug as part of your regimen, it will contribute this much to your response* »

(Shapiro, 2002)

GT algorithms and virological response

- **Many algorithms are available, but few based on clinical response data sets**
  - 16 different sets examined (Shapiro et al)
  - Different levels of concordance
  - Important to realize that concordance does not necessarily mean good interpretation regarding response
- **Accuracy of predicting response to ABC was tested using 16 different algorithms (Lanier et al)**
  - Only 9/16 algorithms correlated well with virological response (> 70% accuracy)

Example of algorythm disagreement (1)

Comparison of 5 algorythms (I = S)

Agreement	Drug	Median
	zalcitabine	0,26
	stavudine	0,29
	didanosine	0,35
Poor	zidovudine	0,49
	abacavir	0,54
	amprenavir	0,56
	saquinavir	0,68
Moderate	nelfinavir	0,76
	efavirenz	0,76
	indinavir	0,76
	ritonavir	0,81
Good	delavirdine	0,82
	nevirapine	0,84
	lamivudine	0,97

Overall median kappa : 0,72 (IQR : 0,50-0,80)

(From Boulmé et al)

Example of algorythm disagreement (2)

Comparison of 5 algorythms (I = R)

Agreement	Drug	Median
	zalcitabine	0,35
	didanosine	0,38
	stavudine	0,50
Poor	abacavir	0,59
	lamivudine	0,67
	amprenavir	0,70
	nelfinavir	0,71
Moderate	saquinavir	0,72
	zidovudine	0,73
	ritonavir	0,75
	indinavir	0,78
Good	efavirenz	0,79
	delavirdine	0,80
	nevirapine	0,82

Overall median kappa : 0,72 (IQR : 0,61-0,77)

(From Boulmé et al)

Resistance Assay Interpretation

- Optimal interpretation of genotypic and phenotypic resistance assays still in evolution
- All current interpretation systems still need improving
- Good prospective data and clinical validation are a necessity for improving these systems
- Large comprehensive databases and collaborations between clinicians, researchers, companies and regulatory agencies are required

Genotype: Yes or No ? (1)

- At this time, genotype has only shown a short term benefit in some of the published studies and not in all patients groups …
- As with any other medical procedures, evidence based medicine should recommend that genotypic resistance testing be restricted until evidence of its benefit has been unequivocally proven.

Genotype: Yes or No ? (2)

Major scientific and ethical issues

with genotypic interpretation :

- Most available systems are based on interpretations which have not been validated by a peer review process
- The present commercial environment, in which genotypic interpretation is not open to discussion or examination, results in differences in quality between systems which is not helpful to physicians and could be of doubtful benefit to patients

CONCLUSIONS

Whatever the usefulness of genotypic resistance testing could be, the following considerations must be taken into account.

1) **Focus specific clinical question.**

Focused questions may be more often answered by genotypic assay than general questions such as “What do I do now?”

Example : Do I use the specific drug now or do I move to a new class?

CONCLUSIONS

Whatever the usefulness of genotypic resistance testing could be, the following considerations must be taken into account.

2) Use genotypic testing only if it can be applied
(i.e. if potentially active drugs remain available)

CONCLUSIONS

Whatever the usefulness of genotypic resistance testing could be, the following considerations must be taken into account.

3) Consider optimal timing of sample

CONCLUSIONS

Whatever the usefulness of genotypic resistance testing could be, the following considerations must be taken into account.

4) **Be clear as to what resistance test can provide and how interpretation information was derived**

CONCLUSIONS

Whatever the usefulness of genotypic resistance testing could be, the following considerations must be taken into account.

5) Be sure that your laboratory is OK

CONCLUSIONS

Whatever the usefulness of genotypic resistance testing could be, the following considerations must be taken into account.

6) **Interpret report in light of patient's individual clinical situation** (patient history, treatment history, previous resistance tests …)