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Short communication

Predictors of having a resistance test following confirmed virological failure of combination antiretroviral therapy: data from EuroSIDA

Zoe V Fox[1,2], Alessandro Cozzi-Lepri[2,3], Antonella D'Arminio Monforte[4], Anders Karlsson[5], Andrew N Phillips[2], Gitte Kronborg[6], Jesper Kjaer[1], Bonaventura Clotet[7], Jens D Lundgren[1], EuroSIDA*

[1]Copenhagen HIV Programme, Copenhagen, Denmark
[2]University College London Medical School, London, UK
[3]Division of Biostatistics, University of Minnesota, Minneapolis, MN, USA
[4]Istituto di Malattie Infettive e Tropicali, Università di Milano, Milan, Italy
[5]Venhälsan, Södersjukhuset, Stockholm, Sweden
[6]Department of Infectious Diseases, Hvidovre Hospital, Copenhagen, Denmark
[7]University Hospital Germans Trias i Pujol, Badalona, Spain

*Corresponding author e-mail: z.fox@ucl.ac.uk

Background: Guidelines suggest that patients on continuous antiretroviral therapy for >4 months with current viral load (VL)>1,000 copies/ml should be tested for resistance. There are limited data showing the frequency of resistance testing in routine clinical practice following these recommendations.

Methods: In EuroSIDA, virological failure (VF) was defined as confirmed VL>1,000 copies/ml after ≥4 months continuous use of any antiretroviral in a ≥3-drug regimen started during or after 2002. We assessed whether a resistance test was performed around VF (from 4 months before to 1 year after VF) and used logistic regression analysis to assess factors associated with having a resistance test.

Results: A total of 1,090 patients experienced VF a median 8.1 months (range 4 months to 6.3 years) after starting their regimen. There were 395 (36.2%; 95% CI 33.4–39.1) patients with a resistance test around the time of VF. Predictors of having a resistance test following VF include availability of a resistance test earlier than 4 months before VF (OR 2.20, 95% CI 1.77–2.75 for yes versus no; *P*<0.0001), region (OR 0.29, 95% CI 0.14–0.62 for Eastern Europe versus Northern Europe and OR 0.64, 95% CI 0.48–0.85 for Southern Europe versus Northern Europe; global *P*=0.0006) and current calendar year (OR 0.45, 95% CI 0.30–0.68 for ≥2007 versus 2004; global *P*=0.003).

Conclusions: This analysis suggests a delay in genotypic testing after VF that seems longer than expected given current treatment guidelines. This delay is highly variable across Europe.

Introduction

Potent combination antiretroviral therapy (cART) has significantly reduced HIV-related morbidity and mortality in the developed world [1], but responses to therapy may diminish over time and lead to virological failure (VF). Treatment guidelines recommend starting a new regimen containing ≥2 active antiretrovirals (ARVs) [2,3] following VF, where resistance tests in combination with knowledge of the patient's treatment history should be used to determine which ARVs are likely to be active.

HIV resistance testing guidelines have been regularly updated since their introduction in 1998 [4–6]. It is uncertain how these guidelines are followed in practice across Europe and whether resistance tests are conducted comprehensively and promptly following VF. Because mutations may predict long-term virological and clinical outcomes [7–10], appropriate use of resistance testing following VF is crucial. In this study, data from EuroSIDA are used to assess the probability of having a genotypic resistance test at or

Table 1. Patients with VF from all of the countries contributing data to these comparisons[a]

North (*n*=222)	Central West (*n*=247)	South (*n*=436)	Central East (*n*=120)	East (*n*=65)
Denmark (*n*=71, 32.0%)	Belgium (*n*=48, 19.4%)	Greece (*n*=25, 5.7%)	Hungary (*n*=13, 10.8%)	Russia (*n*=35, 53.9%)
UK (*n*=58, 26.1%)	France (*n*=85, 34.4%)	Italy (*n*=195, 44.7%)	Czech Republic (*n*=23, 19.2%)	Estonia (*n*=10, 15.4%)
Finland (*n*=4, 1.8%)	Germany (*n*=89, 36.0%)	Portugal (*n*=45, 10.3%)	Slovakia (*n*=5, 4.2%)	Latvia (*n*=10, 15.4%)
Germany (*n*=18, 8.1%)	Luxembourg (*n*=20, 8.1%)	Spain (*n*=151, 34.6%)	Bulgaria (*n*=3, 2.5%)	Lithuania (*n*=10, 15.4%)
Ireland (*n*=14, 6.3%)	Austria (*n*=5, 2.0%)	Israel (*n*=20, 4.6%)	Poland (*n*=54, 45.0%)	
The Netherlands (*n*=8, 3.6%)			Romania (*n*=15, 12.5%)	
Sweden (*n*=29, 13.1%)			Croatia (*n*=7, 5.8%)	
Norway (*n*=20, 9.0%)				

[a]The numbers shown relate to the number of patients with virological failure (VF) and these are shown as a percentage of patients with VF in each region.

following VF and to examine the predictors of such a test being done.

Methods

We included patients who started a new cART regimen (that is, ≥3 drugs from ≥2 drug classes) at/after 2002 (not necessarily their first regimen) and had VF while still on cART. A patient was considered to have VF and an indication for a resistance test if they had confirmed viral load (VL)>1,000 copies/ml (confirmed at the subsequent visit) after ≥4 months continuous use of any drug in the regimen. This included both patients with continuous VL>1,000 copies/ml in the 4 months after starting cART, and those with VL rebound to >1,000 copies/ml ≥4 months after starting cART. We included the first VF at/after 2002 in each patient and only included one episode per patient.

Patients were followed-up in EuroSIDA [11]. EuroSIDA is an ongoing, longitudinal study that currently includes 16,599 HIV-infected patients. Data on $CD4^+$ T-cell counts and VL levels measured since the last follow-up visit and dates of starting and stopping each ARV were collected at each follow-up visit. Data collection on the utilization and findings of resistance tests has been standardized since January 2002; therefore, our analysis explores resistance tests performed on-site at/after this date. Sites that reported at least one resistance test were included in these analyses. Argentina was excluded in order to study regional variations across Europe. Data are presented according to predefined zones of Europe, as used previously (Table 1) [12].

We investigated the characteristics of patients at VF and explored the proportion of patients with a resistance test from 4 months prior to and up to 1 year post-VF. Baseline covariates and time-updated factors were explored using logistic regression analysis. Statistical analyses were performed using STATA software (version 10.1; StataCorp., College Station, TX, USA).

Results

There were 6,947 (43.1%) patients who started a new cART regimen at/after January 2002 and had ≥4 months of follow-up thereafter. VLs were recorded a median (IQR) 4 (2–5.5), 4 (2–5), 3 (2–4), 4 (2–5), 3 (2–4) and 2 (1–4) times per patient who was followed in 2002, 2003, 2004, 2005, 2006 and ≥2007, respectively. Of patients who started a new cART regimen in 2002, 2003, 2004, 2005, 2006 or ≥2007, VF was seen in 375/1,557 (24.1%), 345/1,569 (22.0%), 208/1,354 (15.4%), 167/1,461 (11.4%), 85/1,141 (7.5%) and 34/1,877 (1.8%), respectively.

Overall, we observed VF in 1,090 (15.1%) patients, where 552 (50.6%) had previously experienced VL<500 copies/ml on their regimen. Patients had used their regimens for a median (range) 8.1 months (4.0 months to 6.3 years) at VF; 928 (85.1%) patients were followed for ≥1 year following VF. The population was primarily male (74.7%), White (88.0%) and had cART-experience prior to 2002 (79.8%; Table 2). There were 393 (36.1%) patients with a pre-VF resistance test (that is, >4 months preceding VF) and 395 (36.2%; 95% CI 33.4–39.1) with a resistance test from 4 months prior to and up to 1 year following VF. There were 287/395 (72.7%) patients with a resistance test and 369/695 (53.1%) of those without a resistance test who changed ARVs in the year following VF.

Overall, the proportions of patients with a resistance test from 4 months before the date of VF to 1 year afterwards varied according to different risk factors (Figure 1A). The probability of having a resistance test was 47.8% (95% CI 41.2–54.3) in Northern Europe, 37.7% (95% CI 31.6–43.7) in Central West, 35.8% (95% CI 31.3–40.3) in Southern Europe, 24.2% (95% CI 16.5–31.9) in Central East and 16.9% (95% CI 7.7–26.1) in Eastern Europe.

In unadjusted logistic regression analysis, variables related to having a resistance test included current calendar year, race, region, prior drug use and availability of a pre-VF test. After adjustment, predictors

Table 2. Characteristics of patients, according to resistance test availability in the year following VF

Characteristic	Patients without a resistance test (*n*=695)	Patients with a resistance test (*n*=395)	All (*n*=1,090)
Male gender, *n* (%)	507 (73.0)	307 (77.7)	814 (74.7)
Race			
White, *n* (%)	625 (89.9)	334 (84.6)	959 (88.0)
Black, *n* (%)	42 (6.0)	34 (8.6)	76 (7.0)
Other/unknown, *n* (%)	28 (4.0)	27 (6.8)	55 (5.0)
Risk group			
MSM, *n* (%)	261 (37.6)	159 (40.3)	420 (38.5)
IDU, *n* (%)	158 (22.7)	79 (20.0)	237 (21.7)
Heterosexual, *n* (%)	215 (30.9)	112 (28.4)	327 (30.0)
Other/unknown, *n* (%)	61 (8.8)	45 (11.4)	106 (9.7)
Year of failure			
2002, *n* (%)	78 (11.2)	56 (14.2)	134 (12.3)
2003, *n* (%)	150 (21.6)	94 (23.8)	244 (22.4)
2004, *n* (%)	136 (19.6)	105 (26.6)	241 (22.1)
2005, *n* (%)	128 (18.4)	70 (17.7)	198 (18.2)
2006, *n* (%)	85 (12.2)	44 (11.1)	129 (11.8)
≥2007, *n* (%)	118 (17.0)	26 (6.6)	144 (13.2)
Use of cART prior to 2002, yes *n* (%)	533 (76.7)	337 (85.3)	870 (79.8)
Use of ART prior to regimen at VF, yes *n* (%)	599 (86.2)	360 (91.1)	959 (88.0)
Use of PIs prior to regimen at VF, yes *n* (%)	534 (76.8)	326 (82.5)	860 (78.9)
Use of NNRTIs prior to regimen at VF, yes *n* (%)	400 (57.6)	278 (70.4)	678 (62.2)
Resistance tests available >4 months preceding VF, yes *n* (%)	191 (27.5)	202 (51.1)	393 (36.1)
Followed for >1 year following VF, yes *n* (%)	563 (81.0)	365 (92.4)	928 (85.1)
Median age, years (IQR)	41.2 (36.4–47.4)	42.0 (37.9–48.3)	41.6 (37.0–47.5)
Median viral load at VF, $\log_{10}$ copies/ml (IQR)	4.2 (3.6–4.8)	4.2 (3.6–4.8)	4.2 (3.6–4.8)
Median $CD4^+$ T-cell count at the time of VF, cells/mm^3 (IQR)	307 (164–470)	256 (139–448)	289 (155–460)
Median number of ARVs used prior to regimen at VF (IQR)	8 (5–11)	9 (6–12)	8 (6–11)
Median number of cART regimens failed prior to regimen at VF (IQR)	2 (1–4)	2 (1–5)	2 (1–4)
Median time on regimen prior to VF, months (IQR)	8.6 (5.7–17.7)	7.5 (5.3–13.4)	8.1 (5.6–16.0)

ART, antiretroviral therapy; ARV, antiretroviral; cART, combination antiretroviral therapy; IDU, intravenous drug use; MSM, men who have sex with men; NNRTI, non-nucleoside reverse transcriptase inhibitor; PI, protease inhibitor; VF, virological failure.

of having a resistance test included region (OR 0.29, 95% CI 0.14–0.62 for Eastern Europe versus Northern Europe and OR 0.64, 95% CI 0.48–0.85 for Southern Europe versus Northern Europe; global P=0.0006), current calendar year (OR 0.45, 95% CI 0.30–0.68 for ≥2007 versus 2004; global P=0.003) and availability of a pre-VF resistance test (OR 2.20, 95% CI 1.77–2.75 for yes versus no; P<0.0001; Figure 1B).

Discussion

In our study of individuals with VF, the probability of a resistance being performed around the time of the VF or within 1 year afterwards was 36.2%, which is consistent with results from the UK CHIC study [13].

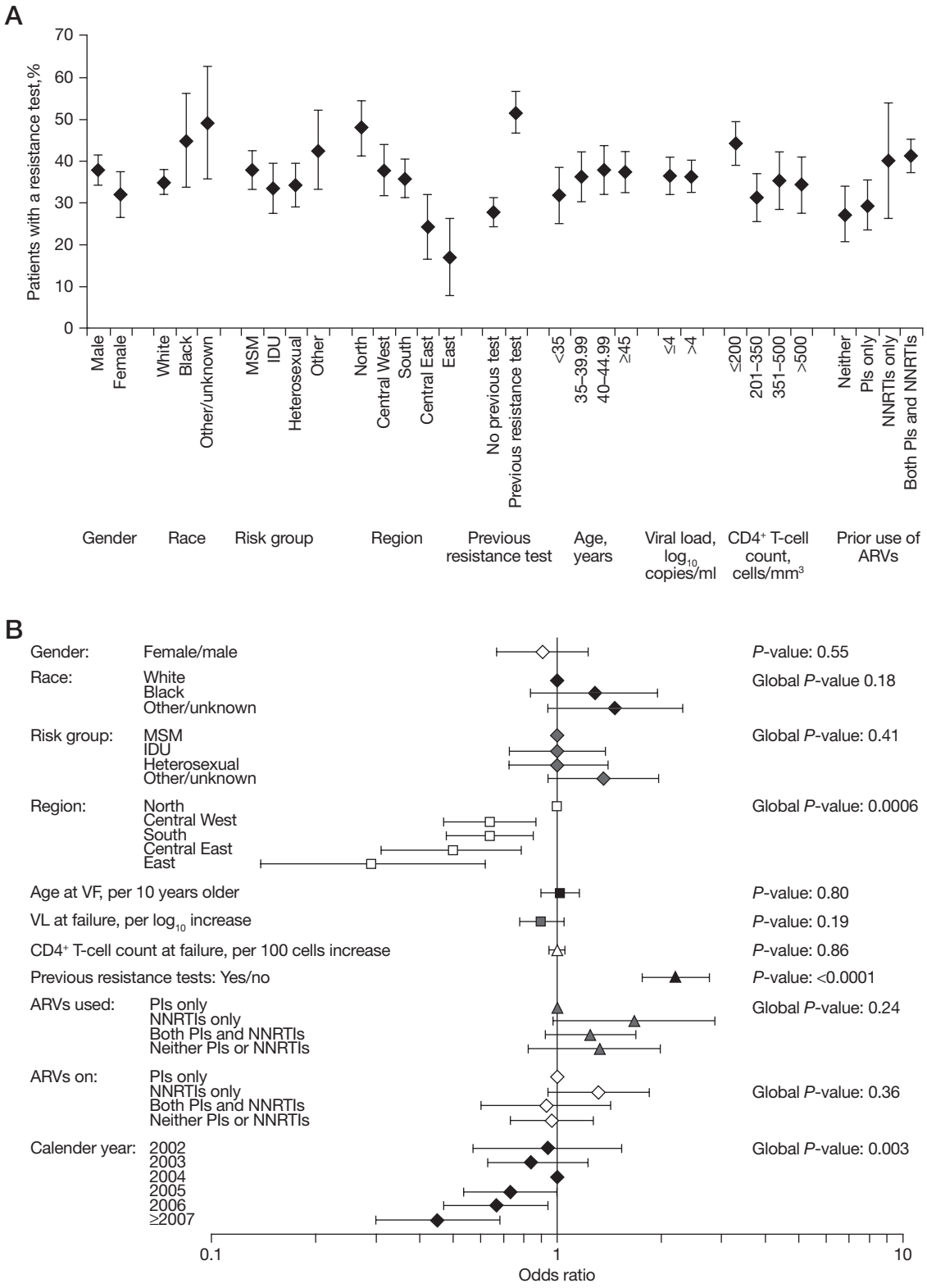
Some of the reasons why clinicians may not perform resistance tests following VF could be: if a patient has perceived poor adherence and is unlikely to show any mutations; if new drugs can be chosen in the absence of a resistance test or if a patient is likely to show extensive mutations and will have no drugs to switch to.

Of the patients without a resistance test, 47% did not change medication in the year following VF, illustrating that known non-compliance, unawareness of VL levels, unwillingness to change therapy, or unavailability of alternative options could explain why resistance tests were not performed. In this study, data come from clinical forms in which it is indicated whether a resistance test was done on-site; because we do not have genotypes for all patients studied here, we are unable to estimate treatment options at VF.

Other explanations for the low rates of resistance testing are variability in how the clinicians interpret guidelines and clinician inexperience. In addition, although we try to ensure comprehensive collection of data, we cannot rule out the possibility that data on resistance testing is not always complete.

Lack of available drugs and limited access to genotyping is likely to explain the low testing rates we

Figure 1. Resistance tests from 4 months before and up to 1 year after VF according to different risk factors



(A) Patients with a resistance test from 4 months prior to and 1 year after virological failure (VF). Error bars show 95% CI. **(B)** Logistic regression model exploring predictors of having a resistance test in the year following VF (results from a multivariable analysis). Error bars show 95% CI. ARV, antiretroviral; IDU, intravenous drug use; MSM, men who have sex with men; NNRTI, non-nucleoside reverse transcriptase inhibitor; PI, protease inhibitor; VF, viroloical failure; VL, viral load.

saw in Eastern Europe [12]. Considering the costs of resistance tests in combination with the specialized equipment/personnel needed and the fact that only 22% of adults in need of cART in Eastern Europe are receiving it [14], treatment expansion is likely to be more of a priority than resistance testing in Eastern Europe [15,16]. Treatment failure is common in Eastern Europe [12] and because increasing levels of resistance have been noted there [17,18] it is crucial to test for resistance before changing treatment.

Between 2005 and 2007, there was an increase in the number of drug possibilities (that is, with the introduction of darunavir/ritonavir, etravirine, maraviroc and raltegravir) and because these drugs are likely to be considered active irrespective of mutations that are present, this may have resulted in the decrease in resistance testing we see in later years.

Having a previous resistance test was a strong predictor of having one at the time of VF. This observation may suggest that availability of tests plays a significant role, or that patients who underwent timely testing may be perceived to be at greater risk of resistance

It is difficult to control for all confounders in the analysis because some are not known and others are not available. For example, there are no data on local availability of resistance tests or on regional variations on the implementation of guidelines. Furthermore, there are no data on the level of experience/expertise of providers by region.

In summary, these results indicate a relatively slow rate of resistance testing following VF. In the modern era, with the approval of new ARVs that are less compromised by resistance mutations, delays in testing may be less of an issue. However, it is likely to become an issue again once these new ARVs are used widely and for prolonged periods of time. In our analysis there was a marked lower utilization of resistance tests in Central East and Eastern Europe compared with Northern Europe, suggesting that different resistance testing benchmarks have been utilized across Europe. There is a need to better understand why geographical differences exist in resistance testing, not only within areas of Europe with less access to genotyping, but also between regions that are likely to have similarly good access to testing.

Disclosure statement

The authors declare no competing interests.

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