Factors associated with virological rebound in HIV-infected patients receiving protease inhibitor monotherapy in the PIVOT trial

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ABSTRACT

Objective: The PIVOT trial found that protease inhibitor (PI) monotherapy as a simplification strategy is safe in terms of drug resistance but less effective than combination therapy in suppressing HIV viral load (VL). We sought to identify factors associated with the risk of VL rebound in this trial.

Methods: PIVOT was a randomized controlled trial in HIV-positive adults with suppressed VL for ≥24 weeks on combination therapy comparing a strategy of physician-selected ritonavir-boosted PI monotherapy versus ongoing triple therapy. In participants receiving monotherapy, we analysed time to confirmed VL rebound and its predictors using flexible parametric survival models.

Results: Of 290 participants initiating PI monotherapy (80% darunavir, 14% lopinavir, 6% other), 93 developed VL rebound on monotherapy. The risk of VL rebound peaked at 9 months after starting monotherapy, and then declined to approximately 5 per 100 person-years from 18 months onwards. Independent predictors of VL rebound were duration of VL suppression prior to starting monotherapy (hazard ratio [HR] 0.81 per additional year <50 copies/mL; p<0.001), CD4 cell count (HR 0.73 per additional 100 cells/mm^3 for CD4 nadir; p=0.008); ethnicity (HR 1.87 for non-white versus white, p=0.025) but not the PI agent used (p=0.27). Patients whose VL was analysed with the Roche Taqman-2 assay had a 1.87-fold risk for VL rebound compared with Abbott RealTime assay (p=0.012).

Conclusions: A number of factors can identify patients at low risk of rebound with PI monotherapy and this may help to better target those who may benefit from this management strategy.

Keywords: HIV, clinical trial, virological rebound, protease inhibitor, monotherapy.

INTRODUCTION

The PIVOT trial showed that PI monotherapy, with regular HIV viral load (VL) monitoring and prompt reintroduction of combination therapy for VL rebound, was non-inferior to combination therapy in preserving future treatment options (i.e. drug resistance) and therefore is a safe and effective alternative for long-term clinical management of HIV infection [1].

Like several other monotherapy trials, the PIVOT trial demonstrated a much higher rate of VL rebound on PI monotherapy than with standard combination therapy (32% higher in PIVOT, 10 to 13% difference found in a previous systematic review) [2]. The VL rebound is generally of low level, easily and rapidly supressed by re-introduction of triple therapy and without apparent long term consequences [1]. Nevertheless, the ability to target patient selection to those at lower risk of VL rebound might increase the appeal of the strategy.

Several previous trials and observational studies have aimed to find characteristics associated with a lower risk of VL rebound. Lower nadir CD4 counts [3-6], a shorter duration of VL suppression [6-8] or treatment prior to monotherapy [9], detectable baseline HIV-VL [9, 10], hepatitis C virus (HCV) coinfection [10, 11] and non-adherence to treatment [4, 9] have all been identified as predictors, although not consistently. The impact of the type of VL assay does not appear to have been explored.

The aim of this study was to analyse the risk of VL rebound by duration of PI monotherapy and to identify clinical predictors of this risk.

METHODS

Study design and population

PIVOT was a randomised controlled trial, performed in 43 centres in the United Kingdom. The main inclusion criteria were being on ART comprising two nucleoside reverse transcriptase inhibitors (NRTI) and one non-NRTI or PI for at least 24 weeks, having a VL<50 copies/ml at screening and for at least 24 weeks beforehand (isolated blips < 200 copies/ml allowed, if followed by two VL<50 copies/ml) and having a CD4 count >100 cells/mm^3 at screening. The main exclusion criteria were the presence of known major PI resistance mutation(s) on a resistance test prior to study entry, previous ART change for unsatisfactory virological response, undergoing or planning to start treatment for HCV co-infection, and co-infection with hepatitis B virus (HBV) unless the patient has had a documented HBV DNA measurement of <1000 copies/ml taken whilst off HBV active drugs. The full eligibility criteria are published elsewhere [1].

Participants were randomly assigned 1:1 to maintain ongoing triple therapy or switch to a strategy of physician-selected ritonavir-boosted PI monotherapy with prompt return to combination therapy if VL rebound (see below) occurred. This analysis is limited to participants assigned to the PI monotherapy arm who commenced PI monotherapy. Blood samples were taken at screening, baseline, weeks 4, 8, 12, and every 12 weeks thereafter. VL was measured in the clinical laboratory at each centre, using a variety of assays determined by local preference.

The protocol was approved by the Cambridgeshire 4 Research Ethics Committee and the Medicines and Healthcare products Regulatory Agency, UK. All participants provided written informed consent.

Statistical methods

The endpoint of this analysis was VL rebound defined as 3 consecutive VL tests ≥50 copies/ml, with at least 4 weeks between the first and last test; one of these 3 tests could be a repeat of the same sample. Time from start of PI monotherapy to first VL rebound (date of 3rd consecutive VL ≥50 copies/ml) was analysed using time-to-event methods, censoring at the earlier of switch from PI monotherapy for any reason (stop of all antiretroviral drugs, or restart of combination ART) or last clinical follow-up if the outcome had not occurred. We ignored subsequent VL rebounds in patients who had more than one episode.

We used flexible parametric survival models to estimate the probability of not having VL rebound by duration of PI monotherapy, including extrapolation beyond the actual follow-up. In brief, these models extend standard parametric models using restricted cubic splines rather than linear functions for the underlying log cumulative hazard, and are implemented in Stata stpm2 [12]. We fitted models on the log cumulative hazard scale to parallel the hazard ratio (HR) calculated from the standard Cox model. Akaike Information Criteria was used to identify the best-fitting model, testing 1 to 6 degrees of freedom (df) of the underlying spline for the log cumulative hazard; the best fitting model had df=5.

Univariable and multivariable flexible parametric survival models were used to find predictors of VL rebound. We evaluated the following baseline factors: age, sex, ART at randomisation (NRTIs+NNRTI vs. NRTIs+PI), time since first consistently suppressed VL below 50 copies/ml, CD4 cell count nadir, baseline CD4 cell count, VL ≥50 copies/ml at baseline or in the previous 6 months, ethnicity (white, non-white), body mass index and HCV status (antibody positive). In addition, we evaluated several time-varying factors: 1) ART non-adherence (missed at least one dose since the last visit, self-report), 2) current PI (some patients changed PI during follow-up due to adverse events or other clinical decision), and 3) HIV-VL assay (some centres changed their local VL assay during the trial period). For illustration of the predictive utility in clinical practice, we predicted time without VL rebound

for hypothetical patients with selected values of duration of VL suppression, CD4 cell count nadir and VL assay adjusted for all factors with $p<0.1$ in the multivariable model; stpm2 achieves this by estimating a survival curve for each individual and then averaging these.

Stata software, version 14.0 (StataCorp), was used for all analyses.

RESULTS

Of the 587 participants in enrolled in PIVOT, 296 were randomised to PI monotherapy. Of these, 6 patients never started monotherapy and were excluded from this analysis. Baseline characteristics are summarised in Table 1. Initial ritonavir-boosted PI was darunavir in 233 (80%), lopinavir in 40 (14%), atazanavir in 16 (6%), and saquinavir in 1 (<1%). At study entry, the median time since first consistently suppressed VL <50 copies/ml was 3 years.

Rate and risk of VL rebound over time

The 290 participants included had a median trial follow-up time of 44 (range 3 to 59) months. Of these, 93 (Kaplan-Meier estimate 38.5%) had VL rebound whilst treated with PI monotherapy with a median peak viral load of 526 copies/ml (peak was <400 copies/ml in 39 [42%]). Follow-up was censored because of death (n=1), withdrawal/lost to follow-up (n=2), discontinuation of monotherapy for other reasons than VL rebound (n=40: 15 for toxicity, 9 detectable VL not meeting protocol criteria for VL rebound, 9 patient decision, 7 other or unknown reasons), and end of trial (n=154).

The cumulative risk of VL rebound estimated from the parametric model is shown in Figure 1 and was very close to the curve obtained using the Kaplan-Meier approach. The rate of VL rebound increased initially, peaking at around 9 months after the start of PI monotherapy. It declined subsequently until around 18 months and then remained comparatively stable (approximately 5 per 100 person-years).

Overall, the estimated proportion of participants without VL rebound was 77% (95% CI 73-82%), 68% (95% CI 62-73%) and 62% (95% CI 56-69%) by 1, 3 and 5 years after start of PI monotherapy. Extrapolated probabilities of not having VL rebound after 7 and 10 years on PI monotherapy, that is beyond the actual follow-up in PIVOT, were 58% (49-66%), and 53% (42-63%).

Predictors of VL rebound

Across all VL measurements, Abbott RealTime (40% of all measurements) and Roche Taqman-2 (35%) were the most commonly used VL assays; all other assays, including in-house assays (11%), Siemens Versant 3.0 bDNA (10%), Nuclisense 2.0 (2%) and various other assays (≤1% each, including Roche Amplicor) were combined in a third category for analysis. VL assay was changed during follow-up in 41/290 (14%) participants.

Results of univariable and multivariable models are shown in Table 2. Independent predictors of VL rebound were shorter time since first VL suppression (HR 0.81 per year longer suppressed; p<0.001), lower CD4 cell count nadir (HR 0.73 per additional 100 cells/mm^3; p=0.008), a higher baseline CD4 cell count (HR 1.13 per 100 cells/mm^3 increase; p=0.023), non-white ethnicity (HR 1.87 versus white, p=0.025), and testing with the Taqman-2 assay (HR 1.87 compared with Abbott RealTime assay, p=0.012). There also was a suggestion of a higher rate of VL rebound associated with lower adherence (p=0.069). There was no significant difference between lopinavir and darunavir monotherapy (p=0.13), and no significant association with any other factor tested.

The impact of the type of assay used is illustrated in Figure 2. The overall probability of no VL rebound by year 5 on PI monotherapy was 68% for Abbott RealTime, and 50% for the Taqman-2 assay.

For a hypothetical patient with a CD4 nadir of 350 cells/mm^3, 6 years' prior VL suppression with triple therapy, and follow-up testing using the Abbott RealTime assay, the model predicted that the probability of no VL rebound by 5 years would be 86% (88% at 3 years). For a patient with a CD4 nadir of 100 cells/mm^3, 1.5 years prior VL suppression on triple therapy and follow-up testing with the Abbott RealTime assay, the model predicted a probability of no VL rebound by 5 years of 51% (58% at 3 years).

DISCUSSION

In this analysis, we have provided further information on the timing of VL rebounds in patients taking PI monotherapy in the PIVOT trial. We have shown a high rate of early VL rebound which peaked around 9 months from switch. This early rate of confirmed VL rebound appears to be greater than that observed in previous trials of PI monotherapy [2] for reasons that are unclear (although differences in trial populations on some of the factors below, especially the method of VL testing, are likely to have played a role). Importantly, this high rate was short-lived: in the period after 18 months from the time of switch only about 5% of patients experienced VL rebound each year. Thus a sub-group of patients are likely to be able to remain on PI monotherapy in the longer term, highlighting the importance of identifying factors that predict a sustained response. The size of the PIVOT trial, the duration of follow-up, and the larger number of VL rebounds episodes observed means that this trial population is able to make a definitive contribution to the understanding of these factors.

We found an increasing risk of virological failure with shorter duration of VL suppression prior to switch to monotherapy, as has been found in other studies [6-8]. A related measure, shorter time of prior ART exposure, was found to be a risk factor in the MONOI trial on darunavir monotherapy [9]. Prolonged viral suppression could be a marker for better adherence to antiretroviral therapy or favourable genetic factors but might also be a surrogate marker of lower viral reservoir size [13, 14]. In the MONOI trial a baseline ultrasensitive VL >1 copy/mL and higher baseline HIV-1 DNA were predictive of virological rebound [9]. Similar to a study on combination ART [13], we did not find any ceiling on this effect – the longer that patients were supressed prior to switching (even beyond 2-3 years) the better they appeared to do on monotherapy. This suggests that PI monotherapy may be best positioned as a clinical management option for patients who have already demonstrated long-standing VL suppression rather than being considered as part of a planned stepwise reduction strategy in patients starting ART for the first time [15].

A lower CD4 cell count nadir was also associated with a higher risk for VL rebound, independently of time since HIV-RNA suppression, which was also seen in other monotherapy studies [3-6, 16]. Patients with low nadir CD4 appear to have deficits in immune function that recover only slowly and have a larger HIV reservoir despite prolonged VL suppression on ART [17-19]. Although effective ART is essential for control of VL replication in most patients, this does not negate a potentially important contribution of the immune system to maintaining VL suppression, and this may vary between individuals [20, 21]. A higher baseline CD4 count had no effect in the univariable model, but was associated with a higher risk of rebound in the multivariable analysis. This effect appeared only after adjusting for CD4 nadir and time since RNA suppression, and is therefore likely a statistical artefact introduced by over-adjustment for these factors, or possibly due to residual confounding.

We found a strong association between the type of VL assay and VL rebound, with Taqman-2 showing a nearly two-fold higher rate of rebound than the Abbott RealTime assay. This result is consistent with several studies that tested samples in parallel with these two assays and found that Taqman-2 has greater sensitivity for detecting low-level HIV RNA [22, 23]. Although not based on a direct comparison of assay methods in each patient, this finding has some important implications in the context of PI monotherapy. Firstly, for clinical management of patients on PI monotherapy it may be more appropriate to define higher thresholds of VL rebound for more sensitive assays [24]. The PIVOT protocol specified a conservative approach for re-initiating combination therapy and patients tested using the Taqman-2 assay may have reintroduced triple therapy prematurely (especially given that resistance resulting from the strategy was very rare). Secondly, it may explain in part the higher rate of rebound seen in PIVOT compared to some other PI monotherapy trials. The impact is hard to assess as a variety of VL assays have been used in previous trials [2] and

in many cases this was not reported. Thirdly, it will be important that this factor is assessed in any future studies or analyses of PI monotherapy data.

PIVOT was a strategic trial intended to resemble the use of PI monotherapy in clinical practice and hence we allowed clinicians to select the PI for use as monotherapy (although we recommended the use of darunavir or lopinavir). This allows us to compare several PIs within the same trial, whereas previous PI monotherapy trials mandated the use of a single PI. Most clinicians / patients selected darunavir although a sizeable minority (14%) used lopinavir. We did not see a difference between darunavir and lopinavir in virological rebound which is consistent with observational studies of PI monotherapy [6, 8, 25]. Two small trials have randomised patients to either lopinavir or darunavir monotherapy and reported no difference in virological failure [26, 27]. In the absence of clear evidence for differences in VL suppression, convenience, tolerability, and the potential for longer-term toxicity may be the most important factors in the selection between darunavir and lopinavir for use as monotherapy. Of note, only one participant in PIVOT developed clinically significant resistance to a protease inhibitor taken as monotherapy, and this was on atazanavir [1].

There was no difference in risk for VL rebound between participants on PI and those on NNRTI at enrolment. In contrast to other studies we did not see an association between HCV coinfection and virological rebound [10, 11], although the number of co-infected patients in PIVOT was relatively small (5%) and limited to those not on HCV treatment or with stable HCV disease. We found non-white patients had a greater risk for VL rebound. Ethnicity has not been examined in other PI monotherapy trials but there are similar findings in trials with combination ART. For example, a combined analysis of several HIV trials in the US found a 40% higher virological failure risk in blacks than in whites that was not fully explained by demographic, clinical, socioeconomic, or adherence factors and was consistent over a wide range of regimens. It was suggested that it may be driven by unmeasured social factors, although biological factors could not be ruled out [28].

Our analysis has some limitations. First, patients who participate in randomised trials may not be representative of the general clinic population. In particular, the very low rate of virological rebound observed in the OT arm suggests that PIVOT enrolled patients who are more adherent than average [1]. Second, as the predictive factors that we analysed (including PI used as monotherapy) were not randomly allocated, the observed associations with VL rebound are potentially (partly) explained by unmeasured confounders. Third, we have speculated that the effects of nadir CD4 count and duration of viral load suppression are due to the size of the HIV reservoir. It would have been informative to have examined this directly (e.g. quantification of HIV DNA in peripheral blood mononuclear cells) but the appropriate samples were not collected in PIVOT.

In spite of the apparent lack of harm associated with short-term VL rebound that was shown by the main analysis of the PIVOT trial the strategy is nevertheless likely to have greater appeal to clinicians and patients if selection criteria can identify those with lower probability of rebound. We showed that patients who had suppressed VL for several years prior to switch to monotherapy, and have a higher CD4 cell nadir would be most suitable. Our findings may help to reassure both patients and clinicians that a strategy of PI monotherapy (with prompt reintroduction of triple therapy in the event of VL rebound) is an acceptable alternative to the management of chronic HIV infection. For patients who have remained without rebound on PI monotherapy for about 18 months, our findings also provide reassurance that the good response is likely to be sustained in the longer term.

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NIP and DTD designed the PIVOT trial. AA-P, CO, AC, IW, MJ, NJB, and EW enrolled participants into the study. NIP, WS, AA-P, KS, and DTD coordinated and oversaw the trial. WS and DTD did the statistical analysis. All authors interpreted data. WS, DTD, and NIP drafted the report. All authors provided input into the report and approved the final version of the report.

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Table 1: Baseline characteristics

Demographic and clinical characteristics (n=290)	
Age (years)	44 (39-50)
Female	70 (24%)
Ethnicity White Black/Other	 193 (67%) 97 (33%)
Body mass index (kg/m^2)	25.3 (22.8-28.4)
Hepatitis C virus antibody positive	14 (5%)
HIV disease status	
CD4 cell count nadir (cells/mm^3)	173 (80-239)
HIV-VL ≥50 copies/ml (or in last 6 months) [a]	29 (10%)
CD4 cell count at recruitment (cells/mm^3)	520 (405-716)
Time since first HIV-VL suppression <50 copies/ml (years)	3 (2-5)
ART history	
Number of drugs ever received	4 (3-6)
On first ART combination	92 (32%)
NRTIs at entry – n (%) Any Tenofovir/emtricitabine Abacavir/lamivudine Other	 290 (100%) 177 (61%) 80 (28%) 33 (11%)
NNRTI at entry Any Efavirenz	 151 (52%) 112 (39%)

Nevirapine	36 (12%)
Etravirine	3 (1%)
PI at entry	
Any	139 (48%)
Atazanavir	59 (20%)
Lopinavir	49 (17%)
Saquinavir	15 (5%)
Darunavir	13 (4%)
Fosamprenavir	3 (1%)

Results as median (IQR) or frequency (%). [a] isolated blips <200 copies/ml in 6 months pre-baseline (n=13) or ≥ 50 copies/ml at baseline (n=12) or both (n=4)

Table 2: Predictors of VL rebound during PI monotherapy

		Univariable model				Multivariable model			
		HR	95%-CI	p-value	overall p	HR	95%-CI	p-value	overall p
Baseline Factors									
Sex:	male	ref.				ref.			
	female	1.46	0.93-2.29	0.104		0.92	0.53-1.61	0.766	
Age (per 10 years)		0.82	0.65-1.05	0.122		0.96	0.74-1.25	0.775	
Ethnicity:	white	ref.				ref.			
	non-white	1.55	0.45-2.66	**0.006**		1.87	1.08-3.23	**0.025**	
Years since first VL suppression		0.87	0.79-0.95	**0.002**		0.81	0.73-0.90	**<0.001**	
CD4 nadir (per 100 cells/mm^3)		0.90	0.74-1.08	0.259		0.73	0.58-0.92	**0.008**	
VL ≥50 copies/mL (or in last 6 months)		1.20	0.64-2.24	0.578		0.91	0.47-1.79	0.794	
CD4 cell count (per 100 cells/mm^3)		1.00	0.91-1.10	0. 956		1.13	1.02-1.26	**0.023**	
Body mass index (per kg/m^2)		1.00	0.96-1.05	0.922		0.97	0.92-1.02	0.261	
Hepatitis C virus antibody positive		1.05	0.39-2.86	0.922		1.16	0.41-3.28	0.783	
ART	(NNRTI vs. PI)	1.46	0.97-2.21	0.071		1.27	0.79-2.06	0.327	
Time-varying factors									
PI:	darunavir	ref.				ref.			
	lopinavir	0.68	0.33-1.40	0.296	0.545	0.53	0.23-1.20	0.130	0.273
	atazanavir/saquinavir	0.96	0.42-2.20	0.923		0.98	0.40-2.35	0.957	
VL assay	Abbott RealTime	ref.				ref.			
	Roche Taqman-2	2.12	1.34-3.35	**0.001**	**<0.001**	1.87	1.15-3.06	**0.012**	**<0.001**
	Other	0.73	0.38-1.41	0.353		0.66	0.34-1.29	0.221	
Non-adherence (any dose missed since last visit)		1.46	0.90-2.36	0.124		1.58	0.97-2.57	0.069	

Note: HR: Hazard ratio; ref.: reference category.

Figure 1: Overall probability and hazard of VL rebound

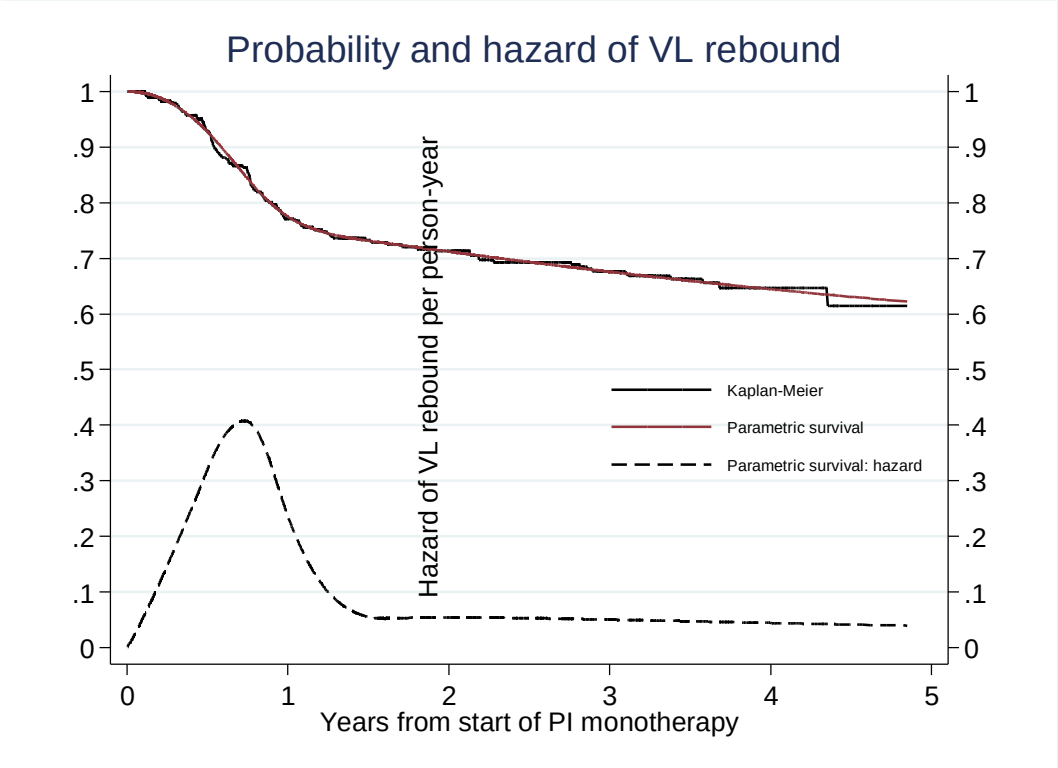
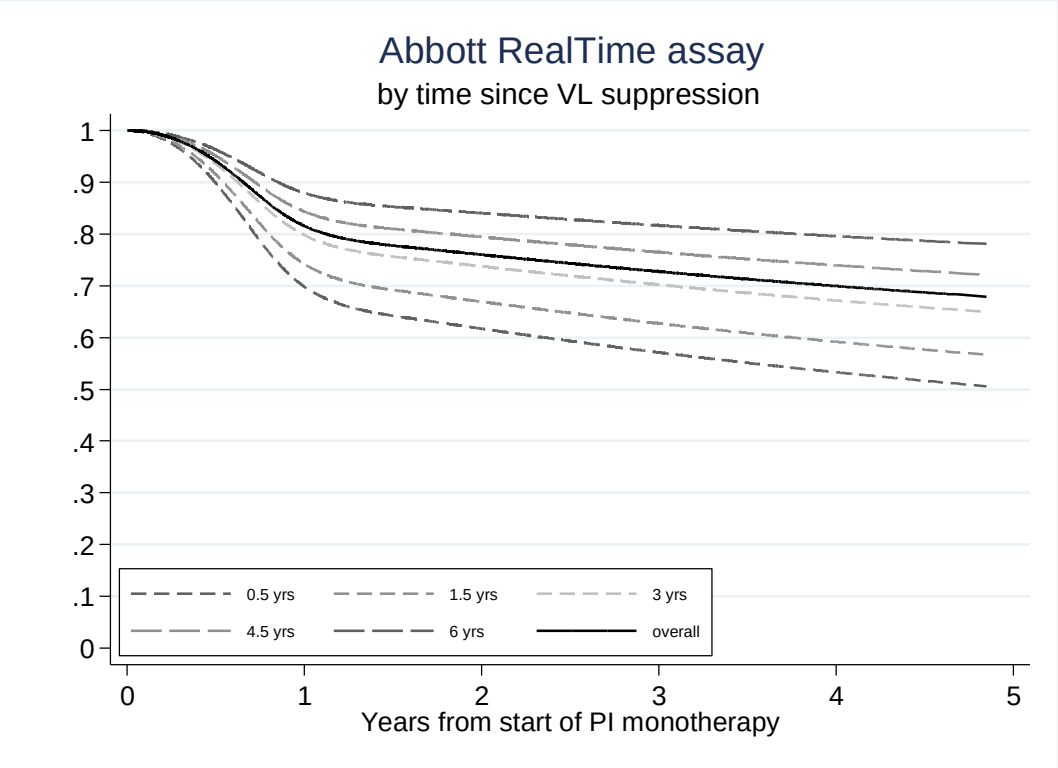


Figure 2: Predicted time to VL rebound, by VL assay and time since VL suppression

a) Abbott RealTime assay



b) Taqman-2 assay