Whatman FTA® cards versus plasma specimens for the quantitation of HIV-1 RNA using two Real-Time PCR assays

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Abstract

Background: Several studies have been conducted to compare the use DBS as alternative to plasma specimens, but mainly using Whatman 903® cards as filter paper. The aim of this study was to evaluate Whatman FTA® cards (FTA cards) specimens for HIV-1 viral load testing by comparing it to plasma specimens, using 2 real-Time PCR assays.

Methodology: A cross-sectional study was conducted between April 2017 and September 2017, in HIV-1 patients admitted at Yalgado Ouédraogo teaching hospital. Paired FTA cards and plasma specimens were collected and analyzed using Abbott RealTi*m*e HIV-1 assay (Abbott) and COBAS® AmpliPrep/COBAS® TaqMan v2.0 (Roche), following manufacturers' protocol.

Results: A total of 107 patients were included. No Statistical differences (p-value > 0.05) were observed between the mean viral loads obtained from FTA cards and plasma specimens with Roche and Abbott assays. Twenty-nine samples with Roche and 15 samples with Abbott assay showed discrepant results. At viral loads of ≤1000 copies/mL, the sensitivity and specificity of FTA cards were 78.6%, and 100% with Roche, and 92.3% and 95.9% with Abbott. Strong correlation was found between FTA cards and plasma specimens with both assays. With Roche, Bland-Altman analysis showed bias of -0.3 and 95% limits of agreement of -2.6 to 1.8 log10, with 97/99 cases (97.9%) within agreement limits. With Abbott, Bland-Altman analysis showed bias of -0.1 and 95% limits of agreement of -2.3 to 2.1 log10, with 96/99 cases (96.9%) within agreement limits.

Conclusion: Our study demonstrated the feasibility of using FTA cards filter paper for HIV-1 viral load testing. However, further studies are required for FTA cards filter paper validation in HIV-1 treatment monitoring.

Keywords: Whatman FTA® cards, dried blood spots, HIV-1, viral load, real-time PCR, Burkina Faso.

Introduction

Viral load (VL) testing is the gold standard for HIV treatment monitoring. Periodic VL tests are the most accurate way of determining whether antiretroviral therapy (ART) is working to suppress replication of the virus [1–3]. With ART rapidly expanding in resource-limited settings, VL is a fundamental and even crucial issue in scaling up antiretroviral treatment. However, many barriers exist to VL testing in resource-limited settings, including lack of basic essential equipment, storage and transport limitations for whole blood and plasma[4]. Due to the lability of viral RNA, whole blood in EDTA/K3 tubes cannot be stored more than 6 hours at 25°C[5,6]. Plasma storage and transport require that plasma is transported within 24 hours at 25°C in EDTA/K3 tubes, or within 5 days at 4°C for EDTA/K3 tubes, after centrifugation [7]. In low and middle-income countries, such restrictive guidance on whole blood and plasma transport greatly limits access to VL testing to only those in close proximity to national or regional laboratories. Therefore, a simple method is needed to allow access to HIV-1 VL testing for patients in rural areas.

Since June 2013, WHO has been recommending the use of dried blood spot (DBS) as alternative to plasma for collection, transport, and HIV-1 VL testing and genotyping drug resistance [8,9]. DBS are an inexpensive and practical alternative to plasma; samples are easy to transport, without the need for cold chains or complex equipment; a further benefit of DBS is the reduction in blood sample volume [10,11].

Numerous studies carried out in Burkina Faso and other countries have shown a strong correlation between DBS and plasma specimens for HIV-1 VL testing [12–16]. However, in most of these studies, Whatman 903® cards filter paper was the only filter paper that has been used for HIV-1 load [17]. Diversifying the type of filter papers available for HIV-1 treatment monitoring could reduce shortages risk, decrease costs through price competition and increase the availability of filter paper. In Burkina Faso, another type of paper is now also routinely used for sample collection during malaria vigilance programs and antimalarial drug trials: Whatman FTA® cards filter paper.

The aim of this study was to evaluate Whatman FTA® Cards (FTA cards) specimens for HIV-1 VL testing by comparing it to plasma specimens, using COBAS® AmpliPrep/COBAS® TaqMan v2.0 HIV-1 test (Roche) and Abbott RealTi*m*e HIV-1 assay (Abbott).

Methodology

Study site and design

A cross-sectional study was conducted between April and September 2017, at the National Reference Laboratory for HIV/AIDS and Sexually Transmitted Infections, located at Yalgado Ouédraogo teaching hospital (CHU-YO), 03 BP 7022, Ouaga 03, Ouagadougou, Burkina Faso. Socio-demographic, clinical, and laboratory data were obtained from the study subjects using a structured questionnaire and laboratory analysis blood samples.

Study population

HIV-1 patients admitted at CHU-YO were the study population. The inclusions criteria were: Patients infected with HIV-1, who consented, antiretroviral-naive patients or patients under antiretroviral treatment.

Sample collection and processing

Venous collection of whole blood was performed in 2 EDTA/K3 tubes of 4.7 mL or a single EDTA/K3 tube of 10 mL from patients during their routine visits to the CHU-YO. Before plasma separation, DBS were prepared by dispensing 50 µL and 100 µL of blood per spot (2 spots per card) onto FTA cards and dried at room temperature (25 ± 2 °C) for 18-24 hours. The FTA cards were stored in zip-lock plastic bags with 2 silica gel desiccants at room temperature upon receipt. Plasma was prepared by centrifugation of the whole blood, aliquoted, and stored at −70 °C until testing for HIV-1 viral load. FTA cards samples were analyzed not more than 14 days after being deposited. Processing of FTA cards for elution of RNA was done as per the protocol for HIV-1 RNA quantitation provided by the ROCHE and ABBOTT diagnostics to be used on their assays systems, COBAS® AmpliPrep/COBAS® TaqMan v2.0 HIV-1 test and m2000rt, respectively.

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Viral Load Quantification

VL was measured from the paired FTA cards and in plasma specimens with Roche and Abbott assays, following manufacturers' protocol. VL results obtained from FTA cards specimens were then compared with that of plasma specimens (gold standard).

Statistical analysis

The statistical analyzes were performed using RStudio (Version 0.99.903). Sensitivity, specificity predictive positive value and predictive negative value were estimated to determine the performances of FTA cards for quantification of HIV-1 VL at a viral load threshold of 1000 copies/ml, a decision point for therapeutic efficacy. Bland–Altman analysis was used to measure agreement in viral load values obtained from FTA cards and plasma specimens. Correlations between viral loads obtained from FTA cards and plasma specimens were assessed with the Pearson statistical test. All HIV-1 VL values were log10 transformed prior Bland–Altman and correlation analysis. The significance level was set at a p-value of 0.05.

Ethical considerations

An informed consent form was presented to each patient prior to blood collection and patients who gave verbal consent were enrolled. Additionally, in order to guarantee confidentiality, random anonymous identification numbers were assigned to each patient.

Results

Patient characteristics

A total of 107 patients were included. The mean age of the patients was 42.0 ± 13.4 years (ranging from 1 days to 77 years). Most patients were female (sex ratio =0.39).

Sample collection and bioanalysis

Whole blood was collected from all the patients and the paired FTA cards and plasma specimens collected were analyzed with Abbott and Roche assays for HIV-1 RNA VL. Of all 107 paired FTA cards and plasma specimens tested, 8 FTA cards specimens gave an invalid result with the Abbott assay and were thus excluded from further analysis. Thus, out of 107 paired FTA cards and plasma samples collected, 99 were analysis.

Comparison between FTA cards and plasma specimens in HIV-1 RNA quantitation

With Roche assay

No statistical differences (p-value = 0.1704) were observed between the mean VL obtained from FTA cards (1.75 log10) and plasma (1.37 log10) specimens **(Figure 1A).**

A total of 29 samples showed discrepant results. Eight (17.0%) samples tested not detected on FTA cards but were moderate positive (n=7; 14.9%) and high positive (n=1; 2.1%) on plasma specimens. Twenty-one (70.0%) samples tested moderate positive on FTA cards specimens but gave not detected (n=16, 53.3%) and high positive (n=5; 16.7%) results on plasma specimens (**Table 1**).

With Abbott assay

No statistical differences (p-value = 0.72) were observed between the mean VL obtained from FTA cards (1.50 log10) and plasma (1.38 log10) specimens **(Figure 1B).**

A total of 15 samples showed discrepant results. Twelve (16.7%) samples tested not detected on FTA cards specimens but were moderate positive (n=7; 14.9%) and high positive (n=2; 2.8%) on plasma specimens. Three (11.1%) samples tested high positive on FTA cards specimens but gave not detected (n=1; 3.7%) and moderate positive (n=2; 7.4%) results on plasma specimens (**Table 2**).

Performance of FTA cards for HIV-1 RNA quantitation

With Roche assay

The sensitivity and specificity of FTA cards at VL of ≤1000 copies/mL were 78.6%, and 100%, respectively (**Table 3**).

***With Abbott* assay**

The sensitivity and specificity of FTA cards at VL of ≤1000 copies/mL were 92.3% and 95.9%, respectively (**Table 3**).

Correlation and agreement between FTA cards and plasma specimens in HIV-1 RNA quantitation

With Roche assay

There was a strong correlation (R^2 = 0.790; p-value < 2.2e-16) between FTA cards and plasma specimens values **(Figure 2A)**. Bland-Altman analysis showed bias of -0.3 and 95% limits of agreement of -2.6 to 1.8 log10; total number of cases within agreement limits in this study was 97/99 (97.9%) **(Figure 3A).**

With Abbott assay

The correlation between VL values obtained from FTA cards and plasma specimens tested was strong (R^2 = 0.764; p-value < 2.2e-16) **(Figure 2B)**. Bland-Altman analysis showed bias of -0.1 and 95% limits of agreement of -2.3 to 2.1 log10; total number of cases within agreement limits in this study was 96/99 (96.9%) **(Figure 3B).**

Discussion

Several studies have been conducted to compare the use DBS as alternative to plasma specimens, but mainly using Whatman 903® as filter paper [17]. In Burkina Faso, another type of paper (FTA cards) is now also routinely used for sample collection during malaria vigilance programs and antimalarial drug trials. In this study, FTA cards was evaluated as an alternative sample collection method to plasma for HIV-1 RNA quantitation using commercial Roche and Abbott assays. To the best of our knowledge, this was the first study to evaluate and compare the use of FTA cards filter paper (for DBS) to plasma specimens for VL testing using both Roche and Abbott assays.

In our study, no statistical differences (p-value > 0.05) were observed between the mean VL obtained from FTA cards and plasma specimens using Roche and Abbott assay. These findings are similar to those of previous reports obtained using Whatman 903® cards [18–20]. However, in this study, 17.0% samples tested not detected on FTA cards, were positive on plasma specimens, with 2.1% high positive. This observation of discrepant results is consistent with findings from other studies using Whatman 903® cards [15,21–23]. The reasons of these discrepant results were well documented in the literature[15,17,21–23]. In a systematic review published in

2014, Smit et al. [17] have indicated that the most important reasons DBS is not, and may never be, as sensitive as plasma is because of the differences in sample volume between DBS and plasma. In the current study, the sample volume used on FTA cards was 50 µL and 100 µL, with the Roche and Abbott assays, respectively. Hematocrit has been suggested to recalculate DBS VL to plasma VL copies/mL by applying the difference between plasma and DBS sample volume [24,25]. To make the calculation, hematocrit values can be obtained to adjust DBS VL results by calculating the amount of plasma in a DBS sample. The current study did not used hematocrit adjustment. Indeed, according to the manufacturer's protocol, a hematocrit adjustment is not required for the calculation of VL obtained from DBS with the Roche and Abbott assays.

An over estimation of HIV-1 RNA levels in FTA cards specimens with low-level viremia (below 1000 copies/mL) was observed in this study; this overestimation was also highlighted in the Bland-Altman analysis (means difference of −0.3 and – 0.1 log10, with Roche and Abbott assays, respectively). This observation is consistent with findings by others with DBS [9,17,19,21,23]. A possible and most advanced explanation of this repeated finding could be the contribution of intracellular HIV-1 DNA and RNA which is present in the DBS but not in the plasma counterpart [26,27]. Vidya et al. [18] suggested that the contribution of intracellular HIV-1 DNA and RNA could be more relevant to specimens with low or undetectable levels viremia than to specimens containing higher levels of extracellular HIV-1 RNA.

At the clinical threshold of 1000 copies/mL, sensibility of FTA cards in this study was seen at 78.6% with Roche, which was slightly less than that observed with Abbott assay (Se = 92.3%) and in already most available literature using Whatman 903® cards [15,21–24,28]. Inversely, our results were higher than that observed in Vietnam using Whatman 903® cards with Roche assay [29]. Additionally, the contribution of HIV cell-associated DNA and RNA could be the reason for slightly lower sensibility for Roche assay. Another possible explanation of the sensitivity observed with Roche could be the elution protocol used in the current study. Following manufacturers' instructions, time to incubation of DBS is 10 minutes in thermomixer at 1000 rpm, 56°C.

Both Roche and Abbott assays in this study showed a good correlation and agreement between FTA cards and plasma values which is similar to other studies

comparing DBS (using Whatman 903® cards) to plasma specimens with Roche and Abbott assays [12,14,15,20].

Our study has some limitations. First, the sample size was restricted. Second, FTA cards were not blotted via finger-prick blood. Third, our study was a laboratory-based study, so the impact of FTA cards sample transport conditions was not evaluated. This study gives preliminary elements to investigations for a longitudinally designed study with a stronger power, incorporating additional factors, such as transport and storage under local conditions, to further evaluate FTA cards specimens for HIV-1 VL testing.

Conclusion:

In summary, we demonstrated the feasibility of using FTA cards for HIV-1 VL testing. FTA cards was found to be a sensitive and specific alternative to plasma testing for HIV-1 VL testing using Abbott assay. Both Roche and Abbott assays showed a good correlation and agreement between FTA cards and plasma values. This information is relevant when considering how to improve access to VL testing by diversifying the type of filter papers available in resource-limited settings.

A study which will increase the testing population size and compare the use of Whatman FTA® to Whatman 903® cards specimens for VL testing using both Roche and Abbott assays is planned for the future.

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Conflict of interest

The authors declare that they have no competing interest.

Authors Contributions

L.S. conceived and designed the experiments; A.Y. actively participated to the specimen collection and the study design; A.Y., M.C, G.K.D., H.S., D.C., K.O.

280 performed the experiments. A.Y. analyzed the data; A.Y. and L.S. wrote the
281 manuscript. All authors read and approved the final manuscript.
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283 **References**

284 1. 1. World Health Organization: Antiretroviral therapy for HIV infection in adults
285 and adolescents recommendations for a public health approach: 2010 revision -
286 Available on: https://www.google.com/search?client=firefox-b-
287 d&q=1.%09World+Health+Organization%3A+Antiretroviral+therapy+for+HIV+inf
288 ection+in+adults+and+adolescents+recommendations+for+a+public+health+app
289 roach%3A+2010+revision. Accessed August 26, 2019

290 2. O'Connor J, Vjecha MJ, Phillips AN, Angus B, Cooper D, Grinsztejn B, et al.
291 Effect of immediate initiation of antiretroviral therapy on risk of severe bacterial
292 infections in HIV-positive people with CD4 cell counts of more than 500 cells per
293 µL: secondary outcome results from a randomised controlled trial. Lancet HIV.
294 2017;4(3):e105‑12.

295 3. Hamers RL, Wallis CL, Kityo C, Siwale M, Mandaliya K, Conradie F, et al. HIV-1
296 drug resistance in antiretroviral-naive individuals in sub-Saharan Africa after
297 rollout of antiretroviral therapy: a multicentre observational study. Lancet Infect
298 Dis. oct 2011;11(10):750‑9.

299 4. Barnabas RV, Revill P, Tan N, Phillips A. Cost-effectiveness of routine viral load
300 monitoring in low- and middle-income countries: a systematic review. J Int AIDS
301 Soc. 2017;20 Suppl 7.

302 5. Ginocchio CC, Wang XP, Kaplan MH, Mulligan G, Witt D, Romano JW, et al.
303 Effects of specimen collection, processing, and storage conditions on stability of
304 human immunodeficiency virus type 1 RNA levels in plasma. J Clin Microbiol.
305 nov 1997;35(11):2886‑93.

306 6. Bonner K, Siemieniuk RA, Boozary A, Roberts T, Fajardo E, Cohn J. Expanding
307 Access to HIV Viral Load Testing: A Systematic Review of RNA Stability in
308 EDTA Tubes and PPT beyond Current Time and Temperature Thresholds.
309 PLOS ONE. 1 déc 2014;9(12):e113813.

310 7. Hardie D, Korsman S, Ameer S, Vojnov L, Hsiao N-Y. Reliability of plasma HIV
311 viral load testing beyond 24 hours: Insights gained from a study in a routine
312 diagnostic laboratory. PloS One. 2019;14(7):e0219381.

313 8. WHO | Consolidated guidelines on the use of antiretroviral drugs for treating and
314 preventing HIV infection. Available on:
315 https://www.who.int/hiv/pub/guidelines/arv2013/download/en/. Accessed August
316 26, 2019

317 9. WHO | Consolidated guidelines on the use of antiretroviral drugs for treating and
318 preventing HIV infection. [cité 26 août 2019]. Available on:
319 https://www.who.int/hiv/pub/arv/arv-2016/en/. Accessed August 26, 2019

10. Singh D, Dhummakupt A, Siems L, Persaud D. Alternative Sample Types for HIV-1 Antiretroviral Drug Resistance Testing. J Infect Dis. 01 2017;216(suppl_9):S834‑7.

11. Zhang G, DeVos J, Medina-Moreno S, Wagar N, Diallo K, Beard RS, et al. Utilization of dried blood spot specimens can expedite nationwide surveillance of HIV drug resistance in resource-limited settings. Plos One. 7 sept 2018;13(9):e0203296.

12. Viljoen J, Gampini S, Danaviah S, Valéa D, Pillay S, Kania D, et al. Dried Blood Spot HIV-1 RNA Quantification Using Open Real-Time Systems in South Africa and Burkina Faso. JAIDS J Acquir Immune Defic Syndr. nov 2010;55(3):290.

13. Reigadas S, Schrive MH, Aurillac-Lavignolle V, Fleury HJ. Quantitation of HIV-1 RNA in dried blood and plasma spots. J Virol Methods. 1 oct 2009;161(1):177‑80.

14. Arredondo M, Garrido C, Parkin N, Zahonero N, Bertagnolio S, Soriano V, et al. Comparison of HIV-1 RNA Measurements Obtained by Using Plasma and Dried Blood Spots in the Automated Abbott Real-Time Viral Load Assay. J Clin Microbiol. 1 mars 2012;50(3):569‑72.

15. Zeh C, Ndiege K, Inzaule S, Achieng R, Williamson J, Chang JC-W, et al. Evaluation of the performance of Abbott m2000 and Roche COBAS Ampliprep/COBAS Taqman assays for HIV-1 viral load determination using dried blood spots and dried plasma spots in Kenya. PLOS ONE. 16 juin 2017;12(6):e0179316.

16. Rutstein SE, Hosseinipour MC, Kamwendo D, Soko A, Mkandawire M, Biddle AK, et al. Dried Blood Spots for Viral Load Monitoring in Malawi: Feasible and Effective. PLOS ONE. 21 avr 2015;10(4):e0124748.

17. Smit PW, Sollis KA, Fiscus S, Ford N, Vitoria M, Essajee S, et al. Systematic Review of the Use of Dried Blood Spots for Monitoring HIV Viral Load and for Early Infant Diagnosis. PLOS ONE. 6 mars 2014;9(3):e86461.

18. Vidya M, Saravanan S, Rifkin S, Solomon SS, Waldrop G, Mayer KH, et al. Dried blood spots versus plasma for the quantitation of HIV-1 RNA using a real-Time PCR, m2000rt assay. J Virol Methods. 1 mai 2012;181(2):177‑81.

19. Mbida AD, Sosso S, Flori P, Saoudin H, Lawrence P, Monny-Lobé M, et al. Measure of Viral Load by Using the Abbott Real-Time HIV-1 Assay on Dried Blood and Plasma Spot Specimens Collected in 2 Rural Dispensaries in Cameroon. JAIDS J Acquir Immune Defic Syndr. sept 2009;52(1):9.

20. Ouma KN, Basavaraju SV, Okonji JA, Williamson J, Thomas TK, Mills LA, et al. Evaluation of Quantification of HIV-1 RNA Viral Load in Plasma and Dried Blood Spots by Use of the Semiautomated Cobas Amplicor Assay and the Fully Automated Cobas Ampliprep/TaqMan Assay, Version 2.0, in Kisumu, Kenya. J Clin Microbiol. 1 avr 2013;51(4):1208‑18.

21. Waters L, Kambugu A, Tibenderana H, Meya D, John L, Mandalia S, et al. Evaluation of Filter Paper Transfer of Whole-Blood and Plasma Samples for Quantifying HIV RNA in Subjects on Antiretroviral Therapy in Uganda. JAIDS J Acquir Immune Defic Syndr. déc 2007;46(5):590.

22. Monleau M, Montavon C, Laurent C, Segondy M, Montes B, Delaporte E, et al. Evaluation of Different RNA Extraction Methods and Storage Conditions of Dried Plasma or Blood Spots for Human Immunodeficiency Virus Type 1 RNA Quantification and PCR Amplification for Drug Resistance Testing. J Clin Microbiol. 1 avr 2009;47(4):1107‑18.

23. Marconi A, Balestrieri M, Comastri G, Pulvirenti FR, Gennari W, Tagliazucchi S, et al. Evaluation of the Abbott Real-Time HIV-1 quantitative assay with dried blood spot specimens. Clin Microbiol Infect. 1 janv 2009;15(1):93‑7.

24. Leelawiwat W, Young NL, Chaowanachan T, Ou CY, Culnane M, Vanprapa N, et al. Dried blood spots for the diagnosis and quantitation of HIV-1: stability studies and evaluation of sensitivity and specificity for the diagnosis of infant HIV-1 infection in Thailand. J Virol Methods. févr 2009;155(2):109‑17.

25. Nyagupe C, Shewade HD, Ade S, Timire C, Tweya H, Vere N, et al. HIV Viral Load Estimation Using Hematocrit Corrected Dried Blood Spot Results on a BioMerieux NucliSENS® Platform. Diagn Basel Switz. 30 juill 2019;9(3).

26. Guichet E, Serrano L, Laurent C, Eymard-Duvernay S, Kuaban C, Vidal L, et al. Comparison of different nucleic acid preparation methods to improve specific HIV-1 RNA isolation for viral load testing on dried blood spots. J Virol Methods. janv 2018;251:75‑9.

27. Seu L, Mwape I, Guffey MB. Single genome amplification of proviral HIV-1 DNA from dried blood spot specimens collected during early infant screening programs in Lusaka, Zambia. J Virol Methods. juill 2014;203:97‑101.

28. Andreotti M, Pirillo M, Guidotti G, Ceffa S, Paturzo G, Germano P, et al. Correlation between HIV-1 viral load quantification in plasma, dried blood spots, and dried plasma spots using the Roche COBAS Taqman assay. J Clin Virol. 1 janv 2010;47(1):4‑7.

29. Taieb F, Tram TH, Ho HT, Pham VA, Nguyen HL, Pham BH, et al. Evaluation of Two Techniques for Viral Load Monitoring Using Dried Blood Spot in Routine Practice in Vietnam (French National Agency for AIDS and Hepatitis Research 12338). Open Forum Infect Dis [Internet]. 1 mai 2016 [cité 26 août 2019];3(3). Disponible sur: https://academic.oup.com/ofid/article/3/3/ofw142/2593284

Figures and tables legends

Figure 1: Comparison between Whatman FTA® cards and plasma specimens in HIV-1 RNA. The boxplot to the left (**A**): using Roche assay; the boxplot to the right (**B**): using Abbott assay. The black points in the boxplot indicates the means values.

Figure 2: Correlation between FTA cards and plasma specimens in HIV-1 RNA quantitation. **A**: using Roche assay; **B**: using Abbott assay.

Figure 2: Bland Altman analysis between FTA cards and plasma specimens in HIV-1 RNA quantitation. **A**: using Roche assay; **B**: using Abbott assay.

Table 1: HIV-1 viral load using Whatman FTA® cards and plasma specimens with Roche assay

Table 2: HIV-1 viral load using Whatman FTA® cards and plasma specimens with Abbott assay

Table 3: Sensitivity, specificity, positive predictive value, and negative predictive value of Whatman FTA® cards compared with paired plasma specimen for HIV-1 viral load testing at a 1000 copies/mL medical decision point

Table 1: HIV-1 viral load using Whatman FTA® cards and plasma specimens with Roche assay

FTA cards specimens using Roche	Plasma specimens using Roche			Total	p value
	Not detected	Moderate	High		
Not detected	39 (83.1)	7 (14.9)	1 (2.1)	47 (100.0)	$< 2.2\ 10^{-16}$
Moderate	16 (53.3)	9 (30.0)	5 (16.7)	30 (100.0)	
High	0	0	22 (28.3)	22 (100.0)	

Table 2: HIV-1 viral load using Whatman FTA® cards and plasma specimens with Abbott assay

FTA cards specimens using Abbott	Plasma specimens using Abbott			Total	p value
	Not detected	Moderate	High		
Not detected	60 (83.3)	10 (13.9)	2 (2.8)	72 (100.0)	$< 2.2\ 10^{-16}$
High	1 (3.7)	2 (7.4)	24 (88.9)	27 (100.0)	

Table 3: Sensitivity, specificity, positive predictive value, and negative predictive value of Whatman FTA® cards compared with paired plasma specimen for HIV-1 viral load testing at a 1000 copies/mL medical decision point

FTA cards (copies/mL)		Plasma (copies/mL)		Total	Se, Sp, PPV, and NPV
		≤ 1000	≥ 1000		
ROCHE	**≤ 1000**	22	0	22	Se = 78.6%; Sp = 100.0%, PPV = 100.0%; NPV = 92.2%
	≥ 1000	6	71	77	
	Total	28	71	99	
Abbott	**≤ 1000**	24	3	27	Se = 92.3%; Sp = 95.9%; PPV = 88.9%; NPV = 97.2%
	≥ 1000	2	70	72	
	Total	26	73	99	

Figures

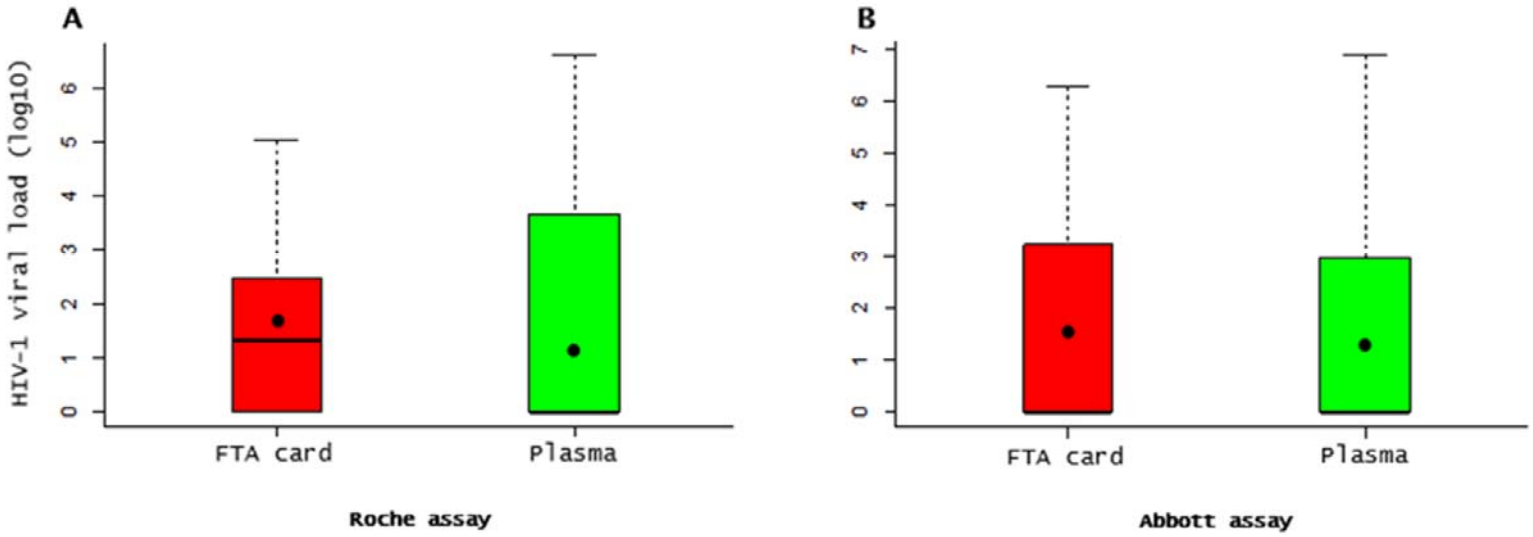

Figure 1: Comparison between Whatman FTA® cards and plasma specimens in HIV-1 RNA. The boxplot to the left (**A**): using Roche assay; the boxplot to the right (**B**): using Abbott assay. The black points in the boxplot indicates the means values.

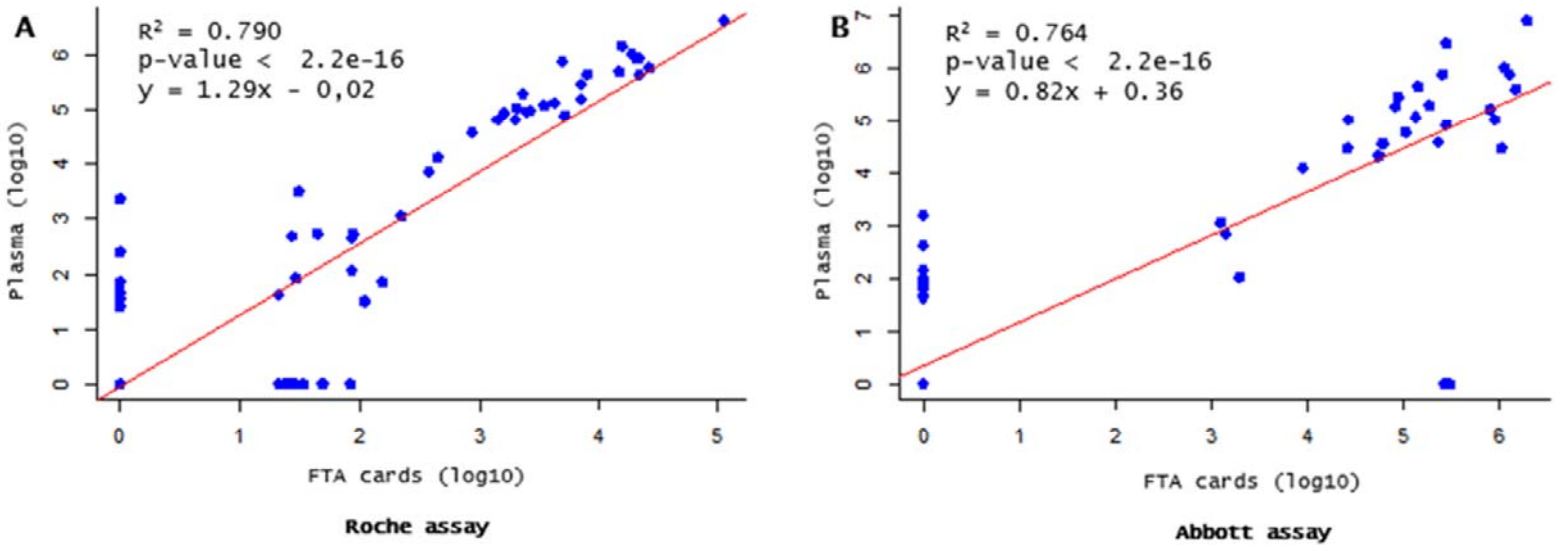

Figure 2: Correlation between FTA cards and plasma specimens in HIV-1 RNA quantitation. **A**: using Roche assay; **B**: using Abbott assay.

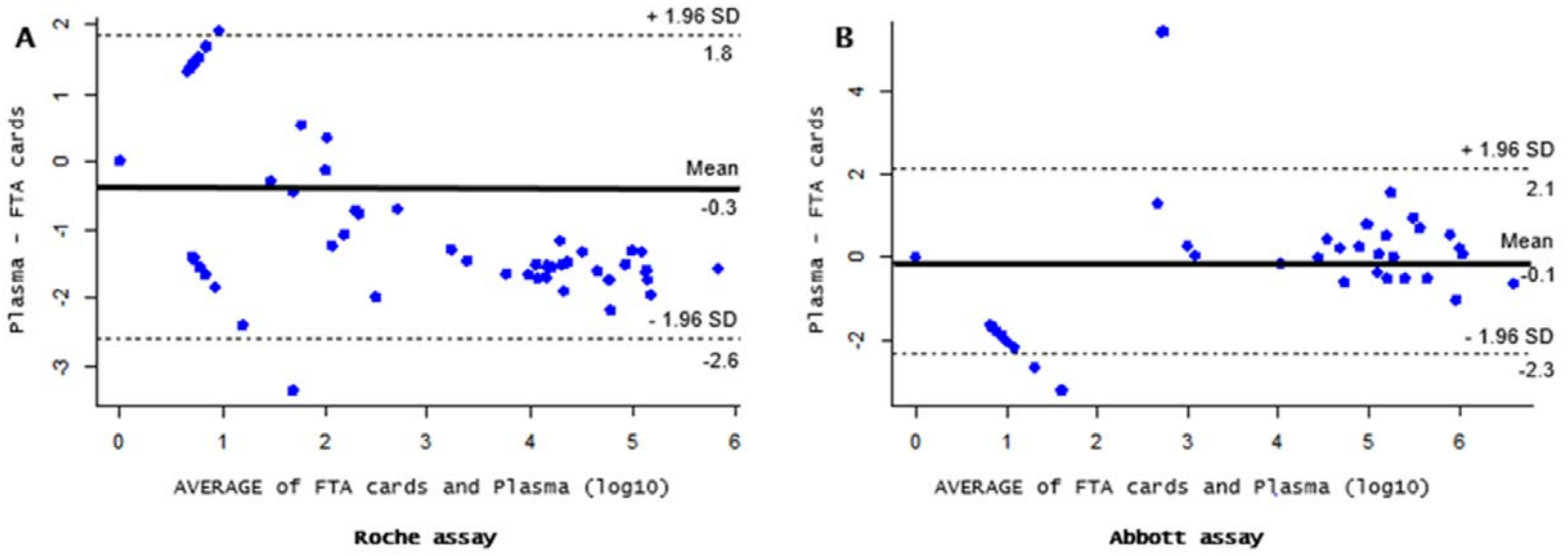

Figure 3: Bland Altman analysis between FTA cards and plasma specimens in HIV-1 RNA quantitation. **A**: using Roche assay; **B**: using Abbott assay.