University of Cape Town

Division of Clinical Haematology, Department of Medicine

The intersection of the HIV epidemic and blood donation in South Africa.

Karin van den Berg

MBChB, PG Dip Trans Med (UFS), MMedSci (UFS)

VBRKAR007

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Supervisor: Professor Vernon J Louw

Co-supervisor: Emeritus Professor Gary Maartens

Co-supervisor: Emeritus Professor Edward L Murphy

Co-supervisor: Doctor Shana Hughes

DECLARATIONS

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ABSTRACT

BACKGROUND

South Africa's large population of people living with HIV (PLWH) affects the local blood transfusion services (BTS) in multiple ways, including the recruitment of safe donors, the demand for blood and the development of blood safety policies. The latter includes the deferral of persons at risk of recently acquired HIV and sensitive testing for HIV antibodies and RNA. Estimating HIV incidence in blood donors is a key measure of successful prevention strategies. Blood donation by PLWH on antiretroviral therapy (ART) was identified as an emerging risk to blood safety as early ART initiation may result in delayed seroconversion, seroreversion, and prolonged suppression of viral replication which may escape detection by HIV antibody and nucleic acid amplification testing (NAT). My PhD research used epidemiologic, incidence modelling and mixed-method qualitative research techniques to assess the impact of undisclosed ART use among blood donors on the safety of the country's blood supply.

AIMS AND METHODS

1. Use summary statistics and multivariable logistic regression to determine the prevalence and determinants of undisclosed antiretroviral (ARV) use among HIV+ donors (HIV+/ARV+) in South Africa by performing ARV testing on stored samples from HIV-positive donors who donated during 2017.
2. Use mixed-methods research techniques to explore the motivations of HIV+/ARV+ blood donors. Specifically investigate how HIV+/ARV+ donors frame potentially conflicting understandings of blood donation, HIV treatment and transmissibility in their decision to donate. Explore what role social, community and policy factors play in this context.
3. Construct recent infection testing algorithms to explore the benefits and limitations of each in differentiating between recent and longstanding HIV infections and determine the extent to which undisclosed ARV use influences HIV incidence estimation and modelling.

RESULTS

ARV-use was demonstrable among 9.8% of HIV-positive blood donors and was independently associated with older age and first-time donation. Most of the HIV+/ARV+ donors were motivated altruism which included both a general wish to help others and wanting to donate specifically for other PLWH. A lack of privacy prevented some donors from disclosing their status. Among blood donors routinely tested with individual donation NAT, the addition of viral load (VL) and ARV testing yielded only marginal improvements in identifying true recent HIV cases with marginal improvements in incidence estimates.

CONCLUSION

Existing programs and policies to limit the risk of transfusion-transmitted (TT) HIV failed to prevent blood donation by HIV+/ARV+ persons. Additional interventions, including targeted communication strategies and procedures to assist those who feel unable to opt out of donation due to peer pressure and privacy concerns, must be considered. While complex, recency testing and incidence modelling adapted to local circumstances may be effective in monitoring the success of such risk mitigation strategies. Overall, BTS must be aware of and actively monitor for undisclosed ARV use among blood donors.

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This thesis is dedicated to my children, Zania and Divan Ingram, who not only provided me with love and support during the times I felt overwhelmed, but who also had to contend with sharing their mother with both her high-stress work at the South African National Blood Service (SANBS) as well as with sacrificing the many weekends and holidays required to complete this body of work. I am eternally grateful for being forgiven for missing so many family events and commitments but believe that my children will learn to value the power of an excellent education.

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- Ravi Reddy, SANBS CEO, and the rest of the SANBS Executive and Board for their deep commitment and support of ongoing learning and research.
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- The staff in the various SANBS laboratories whose daily work not only support the safety of the country's blood supply but also enables research such as what is presented in this thesis.
- All the blood donors, especially the 25 donors who bravely shared their very personal, often traumatic stories with us for no other reason but to support this project.

ABBREVIATIONS

ACASI:	Audio computer-assisted self-interview
aOR:	Adjusted odds ratio
ART:	Antiretroviral therapy
ARV:	Antiretroviral drugs
BTS:	Blood transfusion service
CEPHIA:	Consortium for the Evaluation and Performance of HIV Incidence Assays
CI:	Confidence interval
DHQ:	Donor history questionnaire
EC:	Elite controllers
FRR:	False recency rate
HBV:	Hepatitis B virus
HCV:	Hepatitis C virus
HREC:	Human Research Ethics Committee
HIV+/ARV+:	Denotes undisclosed HIV-positive status and ARV use
HPTN:	HIV Prevention Trials Network
ID-NAT:	Individual donation nucleic acid amplification testing
IQR:	Interquartile range
LAg:	Limiting antigen avidity
MDRI:	Mean duration of recent infection
MSM:	Men who have sex with men
NAT:	Nucleic acid amplification testing
NHLBI:	National Heart, Lung and Blood Institute
NIH:	National Institutes of Health
ODn:	Normalised optical density
OR:	Odds ratio
pa:	Per annum
PEP:	Post-exposure prophylaxis
PLWH:	People living with HIV
PrEP:	Pre-exposure prophylaxis
REDS-III:	Recipient Epidemiology and Donor Evaluation Study-III
RITA:	Recent infection testing algorithms
RSE:	Relative standard error
SANBS:	South African National Blood Service
SEM:	Social Ecological Model

SQDs	Study qualifying donations
TT:	Transfusion transmitted
TTI:	Transfusion transmitted infections
UCSF:	University of California, San Francisco
U=U:	Undetectable = untransmissible
VL:	Viral load
WHO:	World Health Organisation

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PREFACE

The journey towards the completion of this PhD thesis started almost 15 years ago shortly after joining the SANBS when the then Medical Director of SANBS, the late Dr Sam Gulube, sent me to attend the "Training in Clinical Research" course hosted by colleagues from the USA. This was my first introduction to any form of clinical research, but also to Professors Ed Murphy, Brian Custer and Mike Busch who would play a pivotal role in including SANBS in the international arm of the NHLBI funded REDS-III program. I was the in-country lead investigator for several of the REDS-III South Africa sub-projects and principal investigator for the study investigating the association between HIV and blood transfusion in peri-partum women in the Eastern Cape[1] as well as for the Monitoring and Acute Treatment of HIV Study[2]. It is through these projects that I developed my passion for research, and particularly the impact of HIV on the blood transfusion landscape in South Africa. It is also during these projects that we identified a previously unknown phenomenon of PLWH already on ARV donating blood without disclosing their HIV status and ARV history which would later become the focus of my PhD.

While working on the REDS-III projects, I completed a Postgraduate Diploma in Transfusion Medicine at the University of the Free State, where I first met Prof Vernon Louw, who would later be my supervisor for my Master's degree which focused on the impact of HIV on the blood demand in South Africa.[3] The knowledge and experience gained through the REDS-III collaborations and my work with Ed and Vernon directly contributed to my appointment as the national Lead Consultant Translational Research at SANBS and the drive to register for my PhD in 2018. Two events contributed to the delay in completing my thesis beyond what I initially planned. The first was the COVID-19 pandemic which had a major impact on blood transfusion services globally. SANBS, too, had to respond rapidly to ensure we were able to meet the country's blood demand, but also to ensure that we contributed to the international body of knowledge to help manage the pandemic. I authored and co-authored multiple COVID-19 publications during this time.[4-8] Most notably was the clinical trial, for which I was the principal investigator, on the use of convalescent plasma in the treatment of patients hospitalised with COVID[9] as well as co-authorship on the seminal publication that demonstrated that SARS-CoV-2 501Y.V2 escaped neutralization by COVID-19 convalescent plasma collected from donors with presumed wild type virus[10]. The second event was my appointment as the Medical Director of SANBS, a position that required much of my attention over the past two and a half years, but which also allowed me to understand the risk and impact of the HIV epidemic on blood transfusion in South Africa even better.

The purpose of my PhD was to better understand the (at the time) newly identified phenomenon of donors donating blood knowing that they are HIV positive and on treatment. Nationally and internationally this discovery caused significant concern as ARV use may affect the efficacy of current testing strategies to identify and interdict HIV-positive donation.[11-14] This project was arranged around 3 questions (see CHAPTERS 2-4).

First, I wanted to establish the prevalence of this behaviour and potentially identify associated factors. For this part of the project, we tested all available 2017 HIV-positive donations for ARVs. The results of the testing were used to perform prevalence calculations, bivariate as well as multivariable logistic regression analysis.[15]

Second, I wanted to better understand the motivation for this behaviour as it is only through a thorough understanding that we could develop interventions to address this challenge.[16] For this part of the study, we used the results of the ARV testing performed for the first part of the project to identify those donors who were on ARV at the time of donation. In preparation for the project, I solicited the assistance of an international expert on performing qualitative research in the field of HIV and in particular among HIV-positive blood donors. With Dr Shana Hughes' assistance, we trained our local staff (all of whom were trained HIV counsellors) to perform the recruitment, enrolment, consenting and interviewing of these donors. Dr Hughes also mentored me in qualitative data analysis, an underrated skill I am most proud of having acquired.

Third, I wanted to establish what the impact of the failure to disclose HIV status and ARV use had on HIV recency assays and incidence modelling. It is known that recency assays are prone to "false recent" results in PLWH who are on ARV treatment, that is, classifying people with longstanding HIV infections who are on treatment as having recently acquired infections. For this part of the study, we constructed progressively more comprehensive recent infection testing algorithms which were populated with the results of the various tests performed on approximately 500 of the HIV-positive donors who donated during 2017. The outcome of these algorithms was used to perform incidence estimates and the impact the algorithms on the incidence estimates were then quantified and compared. This manuscript was submitted to Vox Sanguinis and is under review.

Various BTS have in recent years raised concerns regarding the risk of undisclosed HIV and ARV use on the safety of the blood supply.[17-19] In fact, a case report by Nishiya *et al.*[11], clearly demonstrated the altered kinetics of post-blood donation HIV testing in what is assumed to be a pre-exposure prophylaxis (PrEP) "breakthrough" infection, underlining the potential for blood transfusion services to fail to identify such donations, thereby further confirming the continued relevance of this work, especially in a country that plans to roll-out PrEP in significant numbers to a key demographic blood donor group.

Dr Karin van den Berg
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CHAPTER 1: INTRODUCTION

1.1 RATIONALE FOR THIS PROJECT

A safe, sustainable blood supply remains an essential pillar of medical care, even in the current era of restrictive transfusion practices and patient blood management.[1] Although fewer than 1% of South Africans receive transfusions each year, and the blood donor base falls short of the World Health Organisation's (WHO) recommended 1% of the population, fulfilling the local need for blood products continues to be challenging.[2-4] Limited resources hamper implementation of programs to recruit and retain low-risk blood donors, to identify donors infected with blood-borne infections, and to develop systems to record, track and notify infected individuals over time. Even where such systems are in place, changes in donor behaviour can affect blood safety in unexpected ways.

South Africa's HIV epidemic, the largest in the world by number of people infected,[5,6] remains the most important public health concern in South Africa,[7,8] and, specifically in this context, affects the ability to identify sufficient low-risk donors to provide for the country's blood demand. Blood donor selection programs and pre-donation risk screening by questionnaire resulted in HIV prevalence among first-time donors of only ~1% while >15% of the general population is HIV positive.[9,10] Despite this relative success, the continued high residual risk of HIV transmission prompted implementation of highly sensitive individual donation nucleic acid amplification testing (ID-NAT) for HIV, Hepatitis C RNA and Hepatitis B DNA in 2005.[11] The implementation of ID NAT has allowed the early detection of donors with acute HIV infections (RNA-positive but still seronegative) and HIV elite controllers (EC), i.e. those who are RNA-negative but seropositive (assumed to be due to inherent immunologic control of the virus).

While enrolling participants into a study of HIV EC, we received reports from our field workers that some of these donors reported that they were aware of their HIV diagnosis and were taking concurrent ARV at the time they presented to donate blood. In response, we performed liquid chromatography/mass spectrometry testing for ARV of stored plasma samples collected between 2010 and 2016, which revealed high rates of ARV use among 226 (67%) presumed EC which were further confirmed in extended testing of historical presumed EC samples.[12] In addition, testing of stored plasma from 135 donors with recent and chronic HIV infection, selected for low levels of viraemia (<400 copies/mL, selected to enrich the sample for HIV-positive donors on ARV but not virally suppressed) also showed a high prevalence (33%) of ARV detection. A virologically suppressed HIV-positive donor on ARV or a true EC could have viremia too low to be detected by current donation screening technology but could still be infectious to blood recipients.[13,14] Furthermore, studies confirmed that prolonged ARV use in prevalent HIV infections or early ARV use in acute infections affects tests aimed at determining HIV recency.[15-18] Undisclosed ARV use may also affect interpretation of these tests, thereby contributing to incorrect HIV incidence calculations.[19-21]

HIV+/ARV+ blood donors pose a new and growing threat to the safety of the country's blood supply.[22] The overarching goal of this study is to better understand this phenomenon, the motivation for this behaviour and its impact on testing algorithms and incidence estimates and modelling.

1.2 BACKGROUND AND LITERATURE REVIEW

The South African HIV Epidemic and its Impact on Blood Services.

South Africa's large and growing HIV-positive population intersects with the local blood transfusion services, SANBS and the Western Cape Blood Service, at multiple levels. In addition to hampering the recruitment and retention of safe donors, it also affects blood demand.[23] While reported HIV incidence rates in South Africa are declining, large-scale roll-out of ART is impacting survival, resulting in increased numbers of PLWH,[24-27] and has contributed to creating the largest treated HIV population in the world.[5,6] At the same time, the high prevalence of anaemia and thrombocytopaenia among HIV-infected individuals places an added burden on the demand for blood products in the country.[23,28] In South Africa and other high HIV burden countries, HIV/AIDS has been the main driver of blood safety policies. [29] Even in well-resourced countries with low HIV-prevalence, concerns regarding even remote potential of TT HIV continues to influence donor deferral and blood screening programs.[30-32]

Despite implementing ID-NAT, the estimated residual risk of receiving a transfusion with a unit of blood donated by a donor in the HIV window period remains high when compared to that of high development index countries. The HIV window period refers to the period between HIV acquisition and the time such an infected person tests positive for HIV. The duration of the HIV window period is determined by viral dynamics, host response and the sensitivity of the test. The risk of HIV transmission through a blood transfusion is determined by the risk of an HIV-infected person who is still in this window period of the infection making a donation that is then missed by available screening programs. In South Africa in 2016, this residual risk using the Weusten model[33] with an infectious dose of 1 virion, was estimated as being 1:53 890.[34,35] In contrast, the HIV residual risk in the USA (in 2016) was approximately 1 in 1.1 million donations and 1 in 21.4 million donation in Canada (in 2012-2014).[32,36] While testing will likely interdict donations made by EC or donors with acute HIV infection, the risk of administrative errors leading to the release of infected units remains possible. In addition, large scale roll-out of "test and treat" programs as well as the launch of PrEP for at risk populations may result in persons with recently acquired HIV-infection accessing ART with the potential for seroreversion[15,16], which may limit the identification of such units using current testing methods.[18]

The phenomenon of undisclosed ARV use by blood donors poses a major concern to blood safety in countries with high HIV prevalence and good access to ART. However, this behavior may occur in any setting where social

stigma related to HIV status remains and where blood centers are known to perform high-quality testing which may inform HIV management and treatment options. Historically, blood centers experienced HIV test seeking behaviour after sensitive HIV donation testing programs were implemented.[37] Detailed analyses of such behaviour resulted in the implementation of several interventions aimed at minimizing it. The newly identified phenomenon of HIV+/ARV+ persons donating blood, may stem from similar patterns of behavior and beliefs, and may demand new tailored interventions. Although real-time testing for ARV as a mitigation strategy is attractive in theory, major constraints related to throughput and cost render this currently unfeasible. A more practical approach might involve both educational campaigns and operational interventions. To maximise impact, the former would need to target multiple sectors, including: the community at large, healthcare providers treating HIV-infected persons, blood center staff and prospective donors. Operational interventions may include developing additional or revised donor selection strategies for prospective donors. The findings from the study proposed here could provide crucial evidence base for such interventions.

Current South African Blood Donor Testing and Counseling.

South Africa has legally mandated screening of all donated blood and blood products. Currently, the testing performed at SANBS includes molecular and serologic testing for HIV, Hepatitis B (HBV), Hepatitis C (HCV), as well as serologic testing for syphilis. The testing aims to interdict donations from donors with such infections. South Africa is one of the few low- and middle-income countries in the world that performs ID-NAT for HIV RNA, HCV RNA, and HBV DNA. ID-NAT enables the detection of HIV infected donations prior to seroconversion, i.e. donations from donors with recently acquired HIV who are still in the serology window period. Current SANBS testing algorithms efficiently identifies longstanding infections. As a result, these donations pose limited risk of transmission and is therefore of less concern when estimating the risk of transfusion-transmitted HIV. In contrast, donors with recently acquired HIV pose a much larger risk. In response, blood services employ behavioural risk questionnaires and one-on-one assessments to limit donation by donors at risk of recently acquired HIV. Identification of donors with recently acquired HIV presents a failure of these behavioural screening strategies and is used to calculate the residual risk of HIV transmission, given that there is still an infectious window period, even when employing ID-NAT testing.

The ID-NAT assay for HIV used in SANBS has a 95% and 50% limit of detection of 18.4 (12-29) and 2.7 (1.9 – 4.0) copies/mL respectively which translates to an estimated window period of 2.9 days.[38] In addition to standard HIV serologic testing, SANBS performed limiting antigen avidity (LAg) testing between January 2012 and January 2018. The LAg test is one of several testing methods which were developed to assist in identifying persons with recently acquired HIV-infections versus those with longstanding infections. It is used to calculate HIV incidence at population levels where repeat testing of individuals to confirm seroconversion is not

available.[20,39,40] This testing strategy enables the detection and categorisation of four subsets of HIV-infected blood donations, namely acute (pre-seroconversion), recent, longstanding and (presumed) EC (**Table 1**).

Table 1. Sub-classification of HIV positive blood donations at the South African National Blood Service.

HIV Sub-classification	HIV RNA	HIV Ab	LAg test	Blood service nomenclature
Acute*	Positive	Negative	Not applicable	NAT yield
Recent*	Positive	Positive	Recent	LAg recent
Longstanding	Positive	Positive	Longstanding	LAg longstanding
Elite controller**	Negative	Positive	Not applicable	Serology yield

* Incident infections includes both Acute and Recent infections; ** Presumed EC until ARV use is excluded.

Routine blood donor testing in SANBS identifies ~1,500 HIV-positive blood donors per annum.[10,41] As described earlier, ID-NAT testing in parallel with HIV serology testing allows for the identification of donors with acute, recent and longstanding HIV infections, as well as potential EC. HIV EC maintain HIV viral loads at very low levels (<50 copies/mL) for a period of at least two years without taking ART, presumably due to inherent protective immune responses.[42] All donors who test both HIV RNA and serology positive are routinely recalled for confirmatory testing, using HIV rapid testing, and counselling. Those who test either RNA only or serology only positive (NAT- and serology yields) are initially fully retested and counselled upon confirmation of their HIV status. These extensive testing strategies combined with donor recall for counselling play a pivotal role in maintaining the safety of the country's blood supply, but, importantly, also creates opportunities for novel research and research collaborations.

One such program, the South African component of REDS-III, was a collaboration between the University of California, San Francisco (UCSF), Vitalant Research Institute (previously Blood Systems Research Institute) and SANBS sponsored by the USA NHLBI. It included a portfolio of clinical and epidemiological research protocols approved by the NHLBI and each institution's Human Research Ethics Committee (HREC), including a case-control study of risk factors for recent HIV and HBV infections as well as a cohort study to assess early ART initiation on HIV disease progression in donors with acute and recently acquired HIV. For these studies, concordant HIV positive donors (RNA+/Ab+) were further classified as recent or longstanding cases. All acute and recently infected HIV-positive donors as well as presumed EC were approached for enrolment in the REDS-III studies. All HIV-positive donors were referred for further management and care following post-donation counselling. For those donors enrolled in the REDS-III early treatment monitoring study, linkage to care was done urgently, included referral to a clinician experienced in HIV-management, immediate initiation on therapy and ongoing monitoring for a three-year period.

During the enrollment and subsequent counselling of donors for these studies we became aware that some of these donors had been diagnosed with HIV and initiated on ART prior to their index donation. Concurrently, we also noted a year-on-year increase in the proportion of our HIV-positive donors who presented as EC. Further investigation confirmed that a large (and growing) proportion of these donors who initially presented as EC, were in fact already taking ARV.[12] This thesis was developed around the concerns raised from these early findings and further built on the work performed as part of the REDS-III South Africa program.

Undisclosed ARV Use – An International Phenomenon

Failure by presumed EC to disclose ART use raised broader research questions regarding the prevalence among blood donors of persons previously diagnosed with HIV and taking ART. It was of interest to us that this behavior is not limited to the blood donation setting. Kim *et al*. found substantial non-disclosure among respondents to the 2012 Kenyan AIDS indicator survey, which led to underestimates of both HIV diagnosis and treatment in the country.[43] Unexpected numbers of virally suppressed participants also led researchers from the HIV Prevention Trials Network 052 (HPTN 052) study to assess ARV use on enrolment samples of 209 HIV-infected index participants.[44] Overall, 23% of participants had at least one drug detected while 47% of virally suppressed participants showed evidence of undisclosed ARV use. The Ugandan Rakai Community Cohort study also demonstrated failure to disclose ARV use among 11% of 557 HIV-positive participants surveyed between September and December 2011, with non-disclosure more common among younger persons.[45] Since the development of this thesis, multiple researchers in various settings have confirmed similar findings of undisclosed ARV use, including among patients attending primary healthcare facilities[46,47], those participating in population census programs[48,49], at risk populations such as men who have sex with men[50], and among blood donors in the USA[22].

Motivations for non-disclosure of ARV use (or any other healthcare information) are not well understood. Some researchers in the field of HIV treatment suggested that access to better healthcare through the research programs and failure to previously disclose HIV status to sexual partners may in part explain this behavior. This may well be true, as this behavior appears to be more prevalent in Africa as opposed to Asia and the Americas where access to care may be better.[44] In addition, despite concerted efforts globally, HIV remains highly stigmatised and may contribute significantly to the failure to disclose HIV status and ARV use even in settings such as prevention of mother-to-child transmission of HIV.[51] Interestingly, in the field of blood transfusion, American researchers have found that donors understood the purpose of a donor questionnaire to be that of assessing the safety of their (the donor's) blood. This understanding resulted in donors responding to the questionnaire in a way that reflects their personal assessment of their perceived risk to the blood supply as opposed to an accurate recording of the factually correct response.[52]

The Impact of Undisclosed ART Use on HIV Testing and Incidence Calculations.

Early initiation of ART (current public health policy in SA) may reduce HIV RNA and antibody levels.[15,16] While this is of clear benefit for the person taking ARV, there are other unintended consequences such as rendering the laboratory assays used to identify HIV-positive donations more prone to false negative results, as has recently been demonstrated.[18] The effect of early initiation of ARV has also been investigated elsewhere. The HIV Prevention Trials Network 052 group assessed the impact of early ART on the performance of HIV rapid tests and HIV incidence assays. [19] Almost 5% of participants who had been virally suppressed for ≥4 years had a non- or weakly reactive rapid test and 18.9% had a false-recent incidence assay result. Similar findings of misclassification as recent infections after ongoing ART use were noted in the Multicenter AIDS Cohort Study and the AIDS Linked to the IntraVenous Experience cohorts.[53] A systematic evaluation of 5 different HIV incidence assays, comprising the LAg[54], BED[55], less sensitive/detuned Vitros[56], Vitros avidity[56] and BioRad avidity[57] assays, by the Consortium for the Evaluation and Performance of HIV Incidence Assays (CEPHIA) demonstrated consistently high false-recent rates (40.4 to 83.6%) across all five tests when performed on HIV-positive persons on uninterrupted treatment for at least 3 months.[39] Further work by this group suggests the inclusion of a minimum viral load of 75 copies/mL improve the false-recent rates among virally suppressed treated persons, but does not eliminate nonzero FRR among other sub-populations and requires consistent estimation of the MDRI relative to the applicable RITA .[58] Increasing the viral load cut-off to 1,000 copies/mL further decreases the false-recent rate, but then also decreases the estimated mean duration of recent infection significantly.

Given the limits of blood donation testing sensitivity, potential seroreversion with early ARV initiation and the possibility of human error, all of which could lead to transfusion-transmitted infection (TTI), there is urgency to understand the extent of blood donation by this population and investigate interventions to reduce it. The range of settings in which ARV use denial occurs is unknown. However, with the impact of ARV use on HIV antibody tests, it may well have implications for any HIV research reliant on antibody-based incidence testing. Our study findings may become more widely relevant as the scope of non-disclosure and its effect on recency testing and incidence estimates are ascertained.

Significance to Transfusion Medicine.

With the continued progress in HIV treatment, potential cure research and prevention interventions (including pre-exposure prophylaxis and prophylactic and treatment vaccinations), the traditional virologic and serologic profile of HIV-infected individuals may change.[18,59] This could compromise the ability of current blood screening methods to accurately identify infected donations. In fact, the effect of undisclosed PrEP on the kinetics of current blood donation testing was clearly demonstrated in Brazil. [18] South Africa, due to the size of its HIV epidemic, large ART program, and the presence of a technically sophisticated blood transfusion service,

presents a unique opportunity to rigorously study the extent of undisclosed ARV use as relevant to the field of Transfusion Medicine and public health.

Although similar denial of HIV status and ART use has previously been reported in at-risk populations and in HIV prevalence surveys[43,46,48,50], this had, at the time of developing this thesis, not been described in blood donors. We will use this opportunity to quantify the prevalence of HIV+/ARV+ individuals among blood donors, characterise the profile of such donors, and explore their motivations and analyze its impact on HIV recency testing algorithms and incidence estimates.

Our research will provide blood services, public health authorities, and other researchers with data on HIV+/ARV+ donors and the determinants of such donations which may inform the development of measures to mitigate such noncompliant donations, while providing information on the impact of undisclosed ARV use on recent infection testing algorithms and estimating HIV incidence. Collectively, the results from this study may help identify potential strategies to reduce donation by HIV+/ARV+ individuals and mitigate risk for blood recipients. Furthermore, increased awareness of this phenomenon may impact blood safety strategies globally, particularly in other high HIV prevalence countries with large-scale ART availability. In addition, it may identify methodological and/or data interpretation issues for other HIV research fields reliant on incidence testing and ARV use disclosure.

1.3 SPECIFIC AIMS, QUALITATIVE RESEARCH QUESTIONS AND HYPOTHESES

Aims and objectives:

There are three broad aims to this research project:

1. Determine the prevalence and determinants of undisclosed ARV use among HIV+/ARV+ donors in South Africa
    a. Using stored samples in the SANBS Biorepository of HIV-positive donors who donated during 2017, perform ARV testing on all available samples to identify cases of undisclosed ARV use.
    b. Perform summary statistics and multivariable logistic regression to determine the association between age, sex, race, donor type (first time, repeat or returned donors), clinic type (fixed and mobile clinics) and region with HIV+/ARV+ donations.
2. Describe the demographic profile and explore the motivations of persons previously diagnosed with HIV, and on ARV, who donate blood.
    a. Develop mixed-methods research knowledge and build capacity to perform this aim as this is lacking in both the researcher and SANBS.
    b. Recruit and enrol HIV+/ARV+ donors identified in Aim 1 to complete an audio computer-assisted self-interview (ACASI) survey.

i. Following the Social Ecological Model (SEM), potential inducements to donation at multiple levels will be examined, including: understanding of HIV disease progression, overestimation of SANBS's ability to detect HIV in donated blood (individual level); economic need and influence of donation incentives, potential peer influence on donation (social level); perception of HIV-related stigma (community level); and access to HIV treatment monitoring in the public healthcare system in South Africa (policy level).

c. Conduct semi-structured one-on-one qualitative interviews with HIV+/ARV+ donors to explore their motivations for donating blood.

3. Determine the extent to which undisclosed ARV use by HIV+/ARV+ donors contribute to false recent results in HIV recent infection testing algorithms and how it influences HIV incidence estimation and modelling.
    a. Using the samples and test results from Aim 1, including limited VL testing, construct progressively more comprehensive recent infection testing algorithms to explore the benefits and limitations of each in accurately differentiating between recent and longstanding HIV infections.
    b. Apply the outcomes of each algorithm to a publicly available HIV incidence model using context-specific mean durations of recent infection (MDRI) and false recency rates (FRR) and compare the results with those derived when using previously published MDRI and FRR for the South African setting.

<u>Hypotheses and Research Questions:</u>

Aim 1:

1. The prevalence of undisclosed ARV use will be: i) lower in recent and longstanding HIV cases compared to potential EC; and ii) higher in those with low compared to high HIV viral load.
2. Undisclosed ARV use will be associated with i) male sex; ii) younger age; and iii) donation at mobile blood drives (due to infrastructure not always allowing for privacy)

Aim 2:

1. How do HIV+/ARV+ donors frame potentially conflicting understandings of blood donation, HIV treatment and transmissibility in their decision to donate?
2. What role do social, community and policy factors play in this context?

Aim 3:

1. "False recency" due to undisclosed ARV use will account for ~10% of all LAg avidity recent cases;
2. The inclusion of ARV testing in HIV recent infection testing algorithms will reduce incidence estimations and improve the precision of the incidence modelling.

I derived these research questions for this PhD through my lived experience as the, at the time, Lead Consultant Translational Research of SANBS, who was responsible for monitoring risks to the country's blood supply. Later, as the Medical Director of SANBS, developing a detailed understanding of the risks associated with HIV+/ARV+ blood donation, became imperative for me to execute my legislated duty to protect the country's blood supply.

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CHAPTER 2: UNDISCLOSED HIV STATUS AND ANTIRETROVIRAL THERAPY USE AMONG SOUTH AFRICAN BLOOD DONORS

Karin van den Berg[1,2,3]; Marion Vermeulen[3,4]; Vernon J Louw[2]; *Edward L Murphy[5,6]; *Gary Maartens[7]

1. Translational Research Department, Medical Division, South African National Blood Service, Roodepoort, South Africa
2. Division of Clinical Haematology, Department of Medicine, Faculty of Health Sciences, University of Cape Town, South Africa
3. Division of Clinical Haematology, University of the Free State, Bloemfontein, South Africa
4. Operations Testing Department, Operations Division, South African National Blood Service, Roodepoort, 1715, South Africa
5. Departments of Laboratory Medicine and Epidemiology/Biostatistics, University of California San Francisco, USA
6. Affiliate Investigator, Vitalant Research Institute, San Francisco, California, USA
7. Division of Clinical Pharmacology, Department of Medicine, University of Cape Town, South Africa

* Edward L Murphy and Gary Maartens contributed equally as senior authors to this manuscript

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Chapter Overview:

Non-disclosure of HIV status and ARV use was first described by our group in 2019. At the time, all HIV-positive donors who tested HIV RNA negative, but serology positive, were presumed to be elite controllers. We noted an increase in the proportion of such donors over time. In addition, our HIV counsellors reported that some HIV-positive donors indicated they were aware of their HIV status and already on ARV treatment. This triggered a study investigating the presence of ARVs in the plasma of these donors which found that approximately two-third of these donors where already on treatment at the time of their blood donation. Questions were then raised whether undisclosed ARV use were also prevalent among other HIV-positive donors and if so, what factors may be associated with undisclosed HIV status and ARV use.

To address these questions, we performed recency testing using the Sedia® HIV-1 LAg-Avidity EIA test (Sedia Biosciences Corporation, Portland, Oregon) as well as testing for ARV using a validated, high-performance liquid chromatography tandem mass spectrometry assay on all available stored plasma samples from HIV-positive donors who donated in 2017. We found that nearly 10% of all HIV-positive donors were already on treatment at the time of donation. Multivariable logistic regression models confirmed that increasing age, first-time donors and donation in provinces with a high HIV burden were independently associated with undisclosed ARV use. To our knowledge, this is the largest study outside of the USA to systematically assess undisclosed ARV use among HIV-positive blood donors, and, at the time, the only such study in a setting of predominant heterosexual HIV transmission. Our findings confirmed that blood services, especially those in high HIV-prevalence settings, should implement measures to actively discourage and manage donation by PLWH on ARV treatment.

I presented this work at the 29th Regional Congress of the International Society of Blood Transfusion in Basel, Switzerland in June 2019 and at the 35th South African National Blood Transfusion Congress in Sun City, South Africa in August 2019 and at the 23rd International AIDS Conference (Virtual) in July 2020. I was also an invited speaker at the 34th Regional Congress of the International Society of Blood Transfusion in Cape Town, South Africa in November 2023 where my topic, Non-disclosure of ARV therapy by blood donors – implications for blood transfusion services, included some of the work in this chapter.

2.1 ABSTRACT

Background

Undisclosed ARV use among blood donors who tested HIV antibody positive, but RNA negative, was previously described by our group. Undisclosed ARV use represents a risk to blood transfusion safety. We assessed the prevalence of and associations with undisclosed ARV use among HIV-positive donors who donated during 2017.

Study design and methods:

SANBS blood donors are screened by self-administered donor health questionnaire, one-on-one interview, and ID-NAT and serologic testing for HIV. Stored samples from HIV-positive donations were tested for ARV and characterized as recent/longstanding using LAg avidity testing.

Results:

Of the 1462 HIV-positive donations in 2017, 1250 had plasma availability for testing of which 122 (9.8%) tested positive for ARV. Undisclosed ARV use did not differ by gender ($p = 0.205$) or ethnicity ($p = 0.505$) but did differ by age category ($p < 0.0001$), donor ($p < 0.0001$), clinic type ($p = 0.012$), home province ($p = 0.01$), and recency ($p < 0.0001$). Multivariable logistic regression found older age (adjusted odds ratio [aOR] 3.73, 95% confidence interval [CI] 1.98–7.04 for donors >40 compared with those <21), first-time donation (aOR 5.24; 95% CI 2.48–11.11), and donation in a high HIV-prevalence province (aOR 9.10; 95% CI 2.70–30.72) compared with Northern Rural provinces to be independently associated with undisclosed ARV use.

Discussion:

Almost 1 in 10 HIV-positive blood donors neglected to disclose their HIV status and ARV use. Demographic characteristics of donors with undisclosed ARV use differed from those noted in other study. Underlying motivations for nondisclosure among blood donors remain unclear and may differ from those in other populations with significant undisclosed ARV use.

KEYWORDS: HIV; anti-retroviral agents; disclosure; prevalence; blood donors; South Africa;

2.2 INTRODUCTION

A safe, sustainable blood supply remains an essential pillar of medical care.[1] Although only about 0.6% of the South African population receives transfusions annually,[2] the blood donor population is less than the WHO's recommended 1%[3]; therefore, meeting the local demand for blood products is challenging. Furthermore, South Africa has the largest HIV epidemic in the world,[4] which hampers the recruitment of sufficient low-risk blood donors to provide for the country's blood demand. Blood donor selection programs and pre-donation risk screening by questionnaire resulted in HIV prevalence among first-time donors of 1.14% even though >18% of the population aged 15–49 years, the target age group for blood donors, is HIV positive.[5,6] Despite this relative success, the continued high residual risk of HIV transmission prompted implementation of highly sensitive ID-NAT for HIV and HCV RNA as well as HBV DNA in 2005.[7] The implementation of ID-NAT has allowed the recognition of acute HIV infections (RNA+/HIV antibody-) and presumed HIV elite controllers (EC), namely those who are antibody+ but RNA- due to inherent immunologic control of the virus. Such categorization of HIV infections is important for estimation of incidence and valuable for research.

While enrolling participants into a study of HIV EC, we received anecdotal reports that some of these donors were previously diagnosed as HIV-positive, taking concurrent ARV and presented to donate blood without disclosing their HIV status or ARV use. We conducted a study of ARV drug assays on stored plasma and found a high proportion (66.4%) of ARV use among presumed EC.[8] This study suggests that HIV+/ARV+ blood donors presenting to donate blood may pose a new and growing threat to the safety of the country's blood supply as early initiation of ARV (current public health policy in South Africa) may reduce HIV RNA and antibody levels.[9–11] Although the likelihood of occurrence and potential number of such instances are currently small, the impact of the community trust in the blood supply may be significant.

The failure by presumed EC to disclose ARV use raises broader research questions regarding the prevalence among the entire blood donor pool of persons previously diagnosed with HIV and taking ARV. Given the limits of blood donation testing sensitivity, potential seroreversion with early ART initiation and the possibility of human error, all of which could lead to TTI, there is urgency to understand the extent of blood donation by this population and investigate interventions to reduce it. In this study, we assessed the prevalence of and demographic (age, sex, ethnicity, donor type, and geographical location) correlates with undisclosed ARV use among HIV-positive donors who donated at SANBS during 2017. We hypothesized that undisclosed ARV use will be lower in males, younger age, and in recent and longstanding HIV cases compared to potential EC.

2.3 MATERIALS AND METHODS

Study design, setting and participants

We performed a cross-sectional study of the prevalence of undisclosed ARV use among donors who tested HIV positive after donating at a SANBS donor centre during 2017. At the time, SANBS collected about 830,000 whole blood donations from voluntary, nonremunerated donors in 8 of the 9 South African provinces per annum. Blood is collected at both fixed site and mobile blood drives; the latter includes high school blood drives as the minimum age for donation in South Africa is 16 years. SANBS donors complete a self-administered questionnaire, which includes questions on HIV status and ARV use. Trained staff review donor responses; donors who disclose HIV-risk factors and/or positive HIV status and/or ARV use are excluded from donation. Donor demographic and donation history data are captured and stored in a database on a secure server. Two specimens are collected from the donor at the time of donation for HIV serologic and ID-NAT, which are performed on all blood donations. Frozen plasma aliquots of donors who test HIV positive are stored at -20°C. All HIV-positive donors who donated during 2017 and for whom plasma aliquots were available for ARV assays were included in the study.

Laboratory assays

Serological assays for HIV antibodies were performed in parallel with multimarker ID-NAT. The Abbott Prism HIV 1/2® (Abbott Diagnostics, Delkenheim, Germany) was used to screen for anti-HIV 1/2 antibodies; specimens with reactive test results were retested in duplicate using the same assay and serology was deemed repeat reactive if either one of the replicates was reactive. In parallel, the Ultrio Elite® multimarker probe assay (Grifols Diagnostics, Barcelona, Spain) on the Procleix Panther® platform was used to screen for HIV RNA, HCV RNA, and HBV DNA. Specimens with reactive test results were repeated in duplicate using the Ultrio Elite® multimarker assay as well as individually with the discriminatory probe assays for HIV, HBV, and HCV to determine which virus was reacting. ID-NAT was deemed repeat reactive if any of the repeat tests were positive.

Donations were classified as HIV positive if HIV ID-NAT and/or serology and immunoblot tests were positive. Based on the combination of their NAT and serology result, HIV-positive donations were classified as RNA+/antibody-, RNA-/antibody+, or RNA+ /antibody+. The Bio-Rad Geenius® HIV 1/2 immunoblot assay is used as a confirmatory test for both RNA+/Ab- as well as RNA-/Ab+ donation. Donations from donors for whom the HIV NAT was reactive and serology negative (RNA+/Ab-), were requested to provide a follow-up sample to check for seroconversion. Where no follow-up occurred, the results of multiple replicate tests from the original plasma bag were used to classify the donation as an acute HIV infection. Donations that were RNA+/antibody+ was classified as concordant-positive HIV infections.

All donations found to be antibody+ (RNA+/antibody+; RNA-/antibody+) and for which plasma was available for testing, were further classified as being a recent or longstanding HIV infection using the Sedia® HIV-1 LAg-

Avidity EIA test (Sedia Biosciences Corporation, Portland, Oregon), a limiting antigen avidity (LAg) test. This assay has a mean duration of recency of 195 (95% confidence interval [CI]: 168–222) days at a normalised optical density (ODn) <1.5 recency/longstanding threshold.[12]

Stored plasma aliquots of HIV-positive donations were sent for batch ARV testing at the Division of Clinical Pharmacology Laboratory, University of Cape Town. A validated, high-performance liquid chromatography tandem mass spectrometry assay was used for the qualitative determination of nevirapine, efavirenz, lopinavir, and atazanavir; this drug combination was selected to detect the vast majority of ARV regimens in use in South Africa during the study period.[13,14] The samples were processed with a protein precipitation extraction method. Deuterated internal standards were used for each analyte. The extraction procedure was followed by liquid chromatographic separation using an Atlantis T3® (3 µm, 2.1 mm x 100 mm) analytical column. An AB Sciex API 4000® mass spectrometer at unit resolution in the multiple reaction-monitoring mode was used to monitor the transition of the protonated precursor ions *m/z* 705.6, 316.0, 629.6, and 267.1 to the product ions *m/z* 168.2, 243.9, 447.3, and 226.0 for atazanavir, efavirenz, lopinavir, and nevirapine, respectively. Electron spray ionization was used for ion production. The validated cutoff concentration for all four analytes was 0.02 µg/mL.

Definitions

Donors are classified as follows: first time if they have never donated a unit of blood before, repeat if they have donated at least one unit in the 12 months preceding the index donation, and lapsed if they donated previously but not in the 12 months preceding the index donation. Recency category “unknown” refers to donors for whom no LAg test result is available.

Statistical analysis

We estimated that stored plasma samples of 1 350 HIV-positive donors would be available for ARV testing. Given this sample size, we would have adequate precision to detect ARV prevalence ranging from 5% (95% CI: 4.0, 6.4%) to 20% (95% CI: 17.9, 22.3%). Donor demographic and donation history characteristics were summarized using descriptive statistics. Chi-square tests were used to assess the differences in the frequencies of undisclosed ARV use by donor demographic and donation history factors. Multivariable logistic regression models for factors associated with undisclosed ARV used were then developed. Variables were first evaluated separately in unadjusted models. After assessing the data for multicollinearity, a variable-inclusive approach was used to develop adjusted models. All variables with a significance level $p<0.1$ in the bivariate analysis were included in the model. All statistical analyses were performed using STATA SE® software (StataCorp. 2019. Stata Statistical Software: Release 16. College Station, TX: StataCorp LLC).

Ethical considerations

All blood donors consented to HIV testing and the use of their blood and data for research aimed at improving the safety of the country's blood supply at the time of donation. Ethics approval for this study was obtained from the SANBS and University of Cape Town HREC as well as the University of California San Francisco Institutional Review Board, which included a waiver for individual informed consent.

2.4 RESULTS

Of the 1 007 580 presentations for blood donation at SANBS donor centres during 2017, 3% resulted in deferral for high HIV risk exposures including self-reported ARV use (**Figure 1)**.

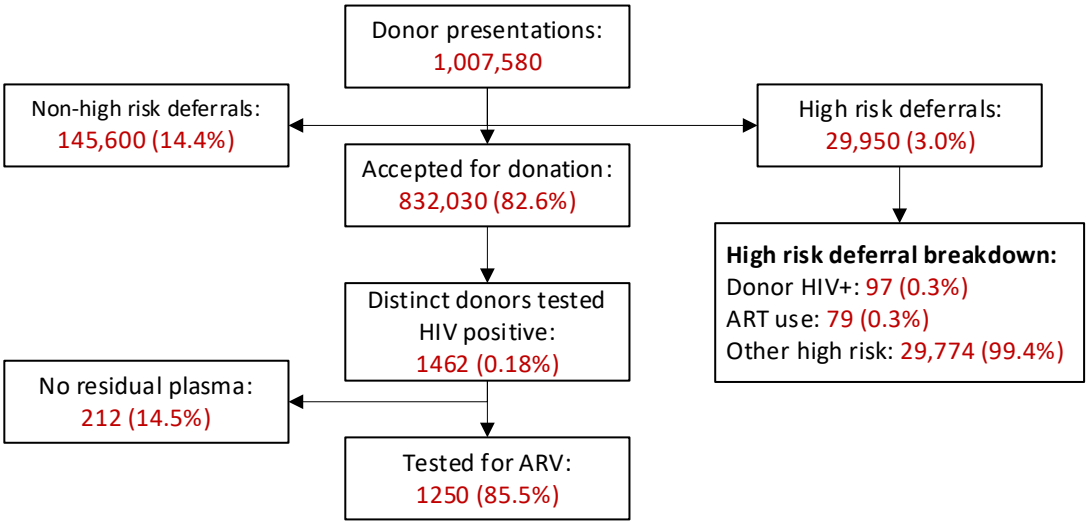

Figure 1: Schematic illustration of donor presentations, deferrals, HIV status and sample availability for HIV testing at SANBS during 2017.

Of the 1 462 (0.18%) donations that tested HIV positive (all from unique donors), 1 250 (85.5%) had frozen aliquots of plasma available for ARV testing. The demographic distribution of donors for whom no frozen aliquots were available did not differ significantly from those for whom aliquots were available. The baseline characteristics of the 1 250 donors included in the analysis are shown in **Table 2.**

Table 2. Baseline characteristics of the 1250 HIV-positive donors by ARV status.

	ARV Negative		ARV Positive		Total	P-Value
	N	%	N	%		
Total	1128	90.2	122	9.8	1250	
Gender						0.205
Female	808	89.6	94	10.4	902	
Male	320	92.0	28	8.0	348	
Ethnicity						0.505
Asian/Indian	10	90.9	1	9.1	11	
Black	1021	90.2	111	9.8	1132	

Coloured	30	85.7	5	14.3	35	
Unknown	29	87.9	4	12.1	33	
White	38	97.4	1	2.6	39	
Age Cat						**<0.0001**
<21	290	93.2	21	6.8	311	
21 - 30	479	93.2	35	6.8	514	
31 - 40	226	85.3	39	14.7	265	
>40	133	83.1	27	16.9	160	
Donor Type						**<0.0001**
First-time	605	85.7	101	14.3	706	
Lapsed*	250	95.1	13	4.9	263	
Repeat	273	97.2	8	2.8	281	
Clinic Type						**0.012**
Fixed	238	94.4	14	5.6	252	
Mobile	890	89.2	108	10.8	998	
Home Province						**0.010**
Eastern Cape	100	91.7	9	8.3	109	
Free State	94	89.5	11	10.5	105	
Gauteng	406	91.2	39	8.8	445	
KwaZulu Natal	199	85.0	35	15.0	234	
Limpopo	65	97.0	2	3.0	67	
Mpumalanga	196	88.7	25	11.3	221	
North West	58	100.0	0	0.0	58	
Northern Cape	10	90.9	1	9.1	11	

* Lapsed donors: previous donation was more than 12-months before the index donation.

ARVs were detected in 122 (9.8%) of tested HIV positive donations. Among the 122 donors with undisclosed ARV use, most (94.3%) tested positive for efavirenz while none tested positive for zidovudine. No participant tested positive for more than one drug (data not shown). There was no difference in undisclosed ARV use by gender or ethnicity (**Table 2**). ARV prevalence increased significantly (p<0.0001) with increasing age, was highest among first-time donors (14.3%, p<0.0001) and those donating at mobile blood drives (10.8%, p<0.0001). The distribution of undisclosed ARV use differed significantly (p=0.010) by home province of the donors, with the highest prevalence in KwaZulu Natal, Mpumalanga and the Free State. ARV use differed significantly by HIV diagnostic category (p <0.0001), with the highest prevalence of 68/80, (85.0%) among the RNA-/Ab+ donors and no ARV detected in the RNA+/Ab- group (**Table 3)**. Among the 1188 HIV Ab+ donors (both RNA- and RNA+ groups), 1153 (97.1%) had samples available for LAg testing of whom 347 (30.1%) tested LAg recent. Undisclosed ARV use prevalence was similar among those who tested LAg Recent (9.8%) and those who tested LAg longstanding (9.2%).

Table 3. HIV testing characteristics of the 1250 participant by ARV status

	ARV Negative		**ARV Positive**		**Total**	
	N	**%**	**N**	**%**		**P-Value**
Total	1128	90.2	122	9.8	1250	
Diagnostic category						<0.0001
RNA+/Ab-	62	100	0	0	62	
RNA-/Ab+	12	15.0	68	85.0	80	

RNA+/Ab+:	1054	95.1	54	4.9	1108	
Recency category*	1066	89.7	122	10.3	1188	
Longstanding**	732	90.8	74	9.2	806	<0.0001
Recent~	313	90.2	34	9.8	347	
Not Tested	21	60.0	14	40.0	35	

* LAg testing performed on Ab+ samples only; ** Estimated MDRI of ≤195 days; ~ Estimated MDRI of > 195 days

A multivariable logistic regression model of factors associated with undisclosed ARV use is shown in **Table 4**. Factors independently associated with undisclosed ARV use were increasing age (aOR 3.73; 95%CI 1.98-7.04) in donors >40 compared to donors younger than 21, first-time donors (compared to repeat donors; aOR 5.24; 95%CI 2.48-11.11) and provinces with a high HIV burden (highest odds in KwaZulu Natal; aOR 9.10; 95%CI 2.70-30.72). There was a non-significant trend of higher undisclosed ARV use among donors donating at mobile blood drives compared to those who donated at fixed sites (OR: 1.75; 95%CI 0.95 – 3.21).

Table 4. Multivariable model of factors associated with undisclosed ARV use among HIV-positive blood donors.

	Bivariate Analysis			Multivariable Model		
Reference Group	OR*	95% CI**		aOR[#]	95% CI	
Gender						
Female	1.33	0.86	2.07	-		
Male	Reference					
Ethnicity						
Asian/Indian	0.92	0.12	7.25	-		
Black	Reference					
Coloured	1.53	0.58	4.03			
Unknown	1.27	0.44	3.68			
White	0.24	0.03	1.78			
Age Category						
<21	Reference			Reference		
21-30	1.01	0.58	1.767	1.52	0.85	2.73
31-40	**2.38**	**1.37**	**4.17**	**3.22**	**1.80**	**5.77**
>40	**2.80**	**1.53**	**5.14**	**3.73**	**1.98**	**7.04**
Donor Type						
Repeat	Reference			Reference		
First-time	5.70	2.74		**5.24**	**2.48**	**11.11**
Lapsed~	1.77	0.72		1.49	0.60	3.70
Clinic Type						
Fixed	Reference			Reference		
Mobile	**2.06**	**1.16**	**3.67**	1.75	0.95	3.21
Home Province						
Northern Rural[##]	Reference			Reference		
Eastern Cape	**3.99**	**1.05**	**15.12**	**4.63**	**1.19**	**17.95**
Free State	**5.19**	**1.41**	**19.11**	**6.44**	**1.71**	**24.29**
Gauteng	**4.26**	**1.30**	**14.01**	**4.12**	**1.24**	**13.73**
KwaZulu Natal	**7.80**	**2.35**	**25.87**	**9.10**	**2.70**	**30.72**
Mpumalanga	**5.66**	**1.67**	**19.11**	**5.48**	**1.60**	**18.80**

*Odds ratio; **Confidence interval; [#]Adjusted odds ratio; [##]Limpopo, North West, Northern Cape; ~ Lapsed donors: previous donation was more than 12-months before the index donation.

2.5 DISCUSSION

We confirmed that a substantial proportion of HIV-positive, prospective South African blood donors had undisclosed ARV use. The overall prevalence of undisclosed ARV use was 9.8% with no significant difference by gender or ethnicity. Undisclosed ARV use was high among donors found to be HIV RNA-/Ab+, a phenotype likely associated with their ARV intake and previously described as "false EC".[8] Factors independently associated with undisclosed ARV use were increasing age, first-time donor status, donation in provinces in South Africa with the highest HIV burden[15].

Failure to disclose ARV use within clinical or research settings has been previously demonstrated.[16-18] Failure to disclose health information, even under direct questioning is common, with 10.4% to 15.5% of patients not disclosing medication use for a variety of conditions to their attending doctor.[19] Several national and community household surveys in Africa reported varying rates of undisclosed ARV use among both those who self-disclose as well as among those who deny HIV-positive health status.[16,20-24] In addition, studies among high-risk populations, such as men-who-have-sex-with-men (MSM) communities[18] and those enrolled in the HPTN 052 study[17], demonstrated high rates of undisclosed ARV. Undisclosed ARV use ranged from 2.2% (South Africa) to 44.4% (Malawi) among persons enrolled in HPTN 052, to as high as 49% among HIV-positive MSM who self-reported an HIV-negative health status.

We found an association between undisclosed ARV use and increasing age, first-time donation status, and residence in South African provinces with highest HIV prevalence. Studies investigating determinants of undisclosed ARV use have been conducted in a variety of settings, including household surveys, HIV vaccine trials, and high-risk populations, and have disparate results in relation to age, education or wealth levels, but consistently found no association with gender, which we also found.[8,16-18,20-22] Neither HPTN 052[17] nor HPTN 075[25] demonstrated associations with age, but did show local as well as between-country regional differences. Household and community surveys generally reported younger age to be associated with greater odds of undisclosed ARV use, but had disparate results in terms of wealth and education.[16,20,21,23,24] By contrast, undisclosed ARV use and HIV status were associated with older age among American MSM.[18] These disparate results may suggest differing behavioural motivations for undisclosed ARV use among different populations.

Our study confirmed the high proportion of undisclosed ARV use among RNA-/Ab+ donors noted by Sykes *et al.*[8] The proportion undisclosed ARV use among RNA-/Ab+ donors in our study period (2017) was 85% compared to the 76.1% reported by Sykes *et al*. for 2016; and may be related to the increasing ARV coverage in South Africa.[26,27] Over this study period, Sykes *et al*. found a temporal increase in the proportion of HIV-positive blood donors who were RNA-/Ab+ and within this group, undisclosed ARV use increased from 38.5% in 2010 to 76.1% in 2016. Similar high rates of undisclosed ARV use have been reported among presumed ARV

naïve HIV-positive persons who had suppressed viral loads, particularly in settings involving HIV treatment studies.[17] A US study investigating the factors associated with patient nondisclosure of medically relevant information found that concerns about being judged, lectured or embarrassed to be independently associated with nondisclosure.[19] In relation to blood donors, feelings of being judged or embarrassed or potentially believing that HIV-positive donors with undetectable viral loads should be able to donate may similarly be reasons for not disclosing their HIV status and ARV use. The higher odds of undisclosed ARV use at mobile blood drives may be indicative of a relative lack of privacy at these settings which potentially creates a barrier to disclosed HIV status and ARV use. Potential test seeking behaviour either to confirm HIV status or efficacy of treatment in controlling the virus may also be motivating non-disclosure.

Despite similar prevalence of undisclosed ARV use among HIV-positive LAg recent (9.8%) and LAg longstanding donor, these data may conceal "false" recency test due to prolonged ARV use. Studies have shown that prolonged ARV use affects most tests, including the LAg avidity test, aimed at determining recency of HIV infection, resulting in over-estimation of recent infections.[28-30] The false-recent rate of these tests range from 50% to 76% among persons on ARV, which more likely explains the high proportion (34 of 347) of undisclosed ARV use among donors classified as having "recent infection".[28] Failed PrEP and early ART initiation are associated with delayed seroconversion and in some instances with seroreversion.[9,31] Rapid initiation of ART in persons with recently acquired HIV could therefore result in failure to identify donors with suppressed VL and delayed or reversed seroconversion with current blood banking testing algorithms .[32] Vermeulen *et al.* reported a 2% probability of TT HIV infection from a red blood cell product donated by donors who are RNA-/Ab+. This finding would suggest that blood products donated by HIV-positive donors who seroreverted or who failed to seroconvert due to very early ART initiation, would likely have a similar probability of transmitting HIV to recipients. While the absolute risk of transmission of HIV through a blood transfusion remains relatively small, the effect on the public's trust may be significant. The impact of the international outcry of what was considered the failure of blood services to protect blood transfusion recipients from HIV and HCV infections during the 1980's[33], still affects existing and potential blood donor perception of the risks involved in donating blood, with >5% of both groups citing risk of "contamination" as a deterrent to blood donation.[34] Furthermore, the perceived incidence of transfusion transmissible infections was 13.5% among patients surveyed in an academic hospital in Germany with 38% of those surveyed having a significant risk perception about possible transfusion transmissible infections.[35]

The 9.8% prevalence of undisclosed ARV use among South African blood donors is alarming and has implications for blood transfusion safety. The higher prevalence of non-disclosure among first-time donors was not unexpected since they had not been previously exposed to the blood service's behavioural screening. However, the non-disclosure among repeat and lapsed donors is more alarming because it indicates a failure

of repeated exposures to the SANBS donor self-assessment questionnaire and one-on-one interview procedure to identify HIV-positive donors on ARV. Of interest is the similar high proportion of undisclosed ARV use among HIV-positive blood donors in the USA. Custer *et al* found that 15% of HIV-positive blood donors and 0.6% of all first-time male blood donors did not disclose their ARV use.[36]

Our study has limitations. First, no samples were available for ARV testing from 14.5% of HIV-positive donations. While some plasma bags were destroyed due to leakage and other operational factors, the majority of these was discarded following failure of a freezer at one of the two SANBS testing sites, which may affect some of the geographic sub-analysis as most of the discarded units were from two geographical areas. Second, due to the short half-life of some ARV drugs our testing algorithm may have missed donors on ARV who had interrupted therapy in the days prior to donation, which may result in underestimation of undisclosed ARV use. We only tested for four of the most commonly prescribed ARV in South Africa (nevirapine, efavirenz, lopinavir and zidovudine). However, at the time, efavirenz was standard in the three-drug fixed dose combination therapy prescribed as first-line therapy for the majority of adults diagnosed with HIV in South Africa and zidovudine was a key component of second-line therapy.[13] Third, we also did not test for tenofovir, which may have missed donors on PrEP. At the time, though, PrEP was not yet widely available in South Africa and unlikely to have materially affected the overall results of this investigation.

2.6 CONCLUSION

We confirmed that almost 1 in 10 HIV-positive blood donors in South Africa did not disclose their known HIV status and ARV use, likely an unintended consequence of the massive national ART rollout. Potential for seroreversion and non-seroconversion with early ART or delayed seroconversion with PrEP will confer increased potential risk to blood safety. While the absolute number of cases is currently still small, the impact of even one case of TT HIV by a blood donor who failed to disclose their HIV status and ARV use, may have a significant impact on the trust of both the donor and recipient communities. While we identified a number of factors associated with undisclosed ARV use among blood donors, the underlying cause and motivation remains unclear and further research is required to better understand this behaviour. Our research may help increase awareness of this phenomenon and help inform blood safety strategies globally, in particular in other high HIV prevalence countries with large-scale ARV availability.

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CHAPTER 3 MOTIVATION FOR BLOOD DONATION BY HIV-POSITIVE INDIVIDUALS ON ANTIRETROVIRALS IN SOUTH AFRICA: A QUALITATIVE STUDY

Karin van den Berg[1,2,3], Edward L Murphy[4,5], Vernon J Louw[2], Gary Maartens[6], Shana D Hughes[5]

1. Medical Division, South African National Blood Service, Roodepoort, South Africa
2. Division of Clinical Haematology, Department of Medicine, University of Cape Town, Cape Town, South Africa
3. Division of Clinical Haematology, University of the Free State, Bloemfontein, South Africa
4. Departments of Laboratory Medicine and Epidemiology/Biostatistics, University of California, San Francisco, California, USA
5. Vitalant Research Institute, San Francisco, California, USA
6. Division of Clinical Pharmacology, Department of Medicine, University of Cape Town, Cape Town, South Africa

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Chapter Overview:

For decades, the risk of TTI and especially HIV infection has been one of the biggest driver of interventions aimed at improving the safety of the blood supply. Significant research investment was expended to determine the risk factors associated with recent HIV infection to enable blood services to develop risk mitigation strategies and interventions aimed at preventing potential donors at risk of recently acquired HIV infection from donating blood. These interventions generally focused on firstly dissuading potential donors from using blood donation centres as an easy access point for high quality HIV testing and secondly educating donors on the risk of window period donations and developing systems for identifying donors at risk of window period infections. The underlying premise of these interventions was that donors were unaware of their potential positive HIV status. However, the discovery that PLWH already on ARV were donating and failing to disclose their status required a complete rethink of previously employed strategies and interventions. Developing strategies to prevent HIV+/ARV+ persons from donating blood would require a detailed understanding of the motivations driving this behaviour.

I performed a mixed-methods study, consisting of semi-structure interviews (qualitative component) and surveys (quantitative component) to explore motivations associated with HIV+/ARV+ donation, with the aim of identifying potential strategies to reduce this behaviour and mitigate risk for blood recipients. The Social Ecological Model (SEM) provided the conceptual framework for this study. Previously identified eligible, consenting HIV+/ARV+ donors were invited to complete an ACASI-based survey and participate in individual, in-depth qualitative interviews. We identified two distinct motivational themes, with altruism the most commonly reported. Two sub-themes in this group included a general desire to help others, but also a previously undescribed motivation of donating specifically for other PLWH. The second major theme was a lack of privacy and fear of others becoming aware of their status. These findings suggest that blood services should develop targeted communication strategies to increase knowledge among PLWH of their deferral from

blood donation and do so without increasing stigma. In addition, they should develop procedures to assist those who feel unable to opt-out of donation due to peer pressure and privacy concerns. To the best of my knowledge this is the first study in the international literature to explore the motivations of the donation behaviour of HIV+/ARV+ blood donors.

I presented this work at the 13th Annual Haematology Oncology Symposium in Cape Town, South Africa in July 2020; 37th International Congress of the International Society of Blood Transfusion (Virtual) in June 2022 as well as at the 36th South African National Blood Transfusion Congress in Durban, South Africa in August 2022. I was also an invited speaker at the 34th Regional Congress of the International Society of Blood Transfusion in Cape Town, South Africa in November 2023 where my topic, Non-disclosure of ARV therapy by blood donors – implications for blood transfusion services, included some of the work in this chapter.

3.1 ABSTRACT

OBJECTIVES

We performed a mixed-methods study to explore the motivations associated with blood donation by HIV+/ARV+ donors seeking potential strategies to reduce such donations and mitigate risk for blood recipients. Here we report predominantly the qualitative component.

BACKGROUND

A safe and sustainable blood supply is dependent in part, on effective pre-donation donor assessment. We previously described failure by HIV+/ARV+ blood donors to disclose their status. Such donations may lead to TT HIV.

METHODS

The SEM provided the conceptual framework for this study. Previously identified HIV+/ARV+ donors were invited to complete a survey (including a validated stigma scale) and qualitative interview, which underwent inductive and deductive thematic analysis.

RESULTS

We uncovered two primary motivational paths to HIV+/ARV+ blood donations: privacy and altruism. The latter included a motivation not previously reported in the literature: donating specifically for other PLWH. The other primary factor was a lack of privacy. These accounts often included donors encountering donation opportunities when accompanied by people to whom they had not and did not plan to disclose their HIV status. Most were highly confident their donations would be identified as HIV-positive and discarded.

CONCLUSION

We demonstrated a complex interaction between individual, social, cultural, and structural/policy factors in blood donations by PLWH who take ARV. Recommendations to limit HIV+ARV+ donations include: 1) Targeted communication strategies to increase knowledge among PLWH of their deferral from blood donation—without increasing stigma, and 2) development of procedures to assist those who feel unable to opt-out of donation due to privacy concerns.

KEYWORDS: Blood donation; motivations; HIV; anti-retroviral agents; health status disclosure;

3.2 INTRODUCTION

The WHO recently reaffirmed the global need for a safe, sustainable blood supply to support the effective delivery of health services and programs.[1] A key safety component is the reduction of the risk of TTI such as HIV, Hepatitis B and C.[2] To achieve this, blood transfusion services employ a multi-pronged approach. While laboratory screening is conducted on all donated blood, donors must pass a combination of pre-donation education, donor assessment via a Donor History Questionnaire (DHQ) and potential deferral. Deferral is disqualification from donation, whether indefinitely (e.g. people living with certain infections, including HIV) or for a specified period (e.g. iron deficiency).[3] These efforts have been remarkably successful, decreasing the number of transfusion-related HIV transmissions in the USA from thousands in the early 1980s[4] to an estimated risk of <1 in 1.6 million donations in 2021.[5] Even in South Africa, with its generalized and growing HIV epidemic of more than 8 million people[6] of whom an estimated 70% are on treatment[7], improved screening strategies reduced the risk of HIV transmission from an estimated 22 per million transfusions in 1994[8] to less than 13 per million transfusions in 2015.[9]

The effectiveness of pre-donation donor assessment and deferral, as a blood safety strategy, depends both on asking the right questions (those that address behaviors and health conditions that truly pose risk) and the willingness of would-be donors to disclose such personal information.[10-12] This can pose challenges. Studies in Europe[11,13], North America[14], Australia[15] and India[16] confirmed higher rates of nondisclosure of risk factors for HIV TTI among donors who tested positive for these than among donors who tested negative. Most studies investigating non-disclosure among blood donors focus on *risk behaviors* for HIV and other TTI. Non-disclosure of *known* HIV status or ARV use among blood donors has been less frequently explored.

Donations from donors with undisclosed, but known HIV-positive status and/or ARV use (HIV+/ARV+), even with undetectable viral loads, may pose a risk to the safety of the blood supply, especially in high HIV prevalence and ARV uptake settings such as South Africa. This is because when PLWH are not deferred prior to donation, detection of virus in their blood is dependent on serologic and molecular HIV assays, the efficacy of which may be compromised in persons with early initiation of ARV or those with pre-exposure prophylaxes breakthrough infections.[17-19] It should be noted that while an undetectable viral load is largely protective for sexual transmission of HIV, this is not necessarily true for transfusion associated transmission, as demonstrated by modelling.[20] South Africa implemented a universal “test and treat” strategy in September 2016,[21] so early ARV initiation should now be the norm. With a continued HIV incidence rate greater than 1%, the number of people treated (early) for HIV in the country grows annually[6], and the risk of non-compliant blood donation resulting in HIV transmission to a blood recipient grows along with it.

Failure to disclose HIV+/ARV+ has recently been quantitatively described in both South Africa[22,23] and the USA.[24] Our group at SANBS became aware of anecdotal reports of undisclosed HIV+/ARV+ among South African blood donors. Subsequent investigation revealed detectable ARV in two-thirds of donors who tested HIV antibody positive but negative by individual donation nucleic acid amplification testing, a result that suggests viral suppression from antiretroviral therapy in an HIV-positive person.[22] We found that almost 10% of all HIV-positive donors who donated at SANBS had demonstrable levels of ARV.[23] In the USA, Custer *et al.*[24] demonstrated undisclosed ARV use among 15% of HIV-positive blood donors and in 0.6% of all first-time male blood donors. To our knowledge, qualitative studies of this phenomenon are, as yet, non-existent. To further explore the phenomenon of blood donation by HIV+/ARV+ donors, we designed a mixed-methods study to explore the motivations associated with this behavior. Here we report predominantly the findings of the qualitative component.

3.3 METHODS

Institutional review board approval was obtained from both SANBS and the University of Cape Town. This mixed-methods study was conducted at SANBS, a blood service that serves 8 of the 9 provinces in South Africa, and collects approximately 900,000 units of blood from ~450 000 donors. From February to April 2019, eligible, consenting HIV+/ARV+ donors were invited to complete a survey, administered through ACASI technology, and an individual, in-depth qualitative interview ("interview"). The SEM provided the conceptual framework for this study. The SEM is frequently used in health research[25-27] and posits a complex interplay between multiple levels of influences, human behaviour and health outcomes. A version of the SEM adapted specifically to deal with HIV risk[28] guided the development of data collection instruments (**Figure 2**). Specifically, survey items and interview questions addressed influences at individual, social, cultural, and policy levels, as well as potential donor motivations entertained by the research team during study conception (e.g., desire for incentives offered for donation, CD4 or viral load test seeking, donor belief they have been cured of HIV).

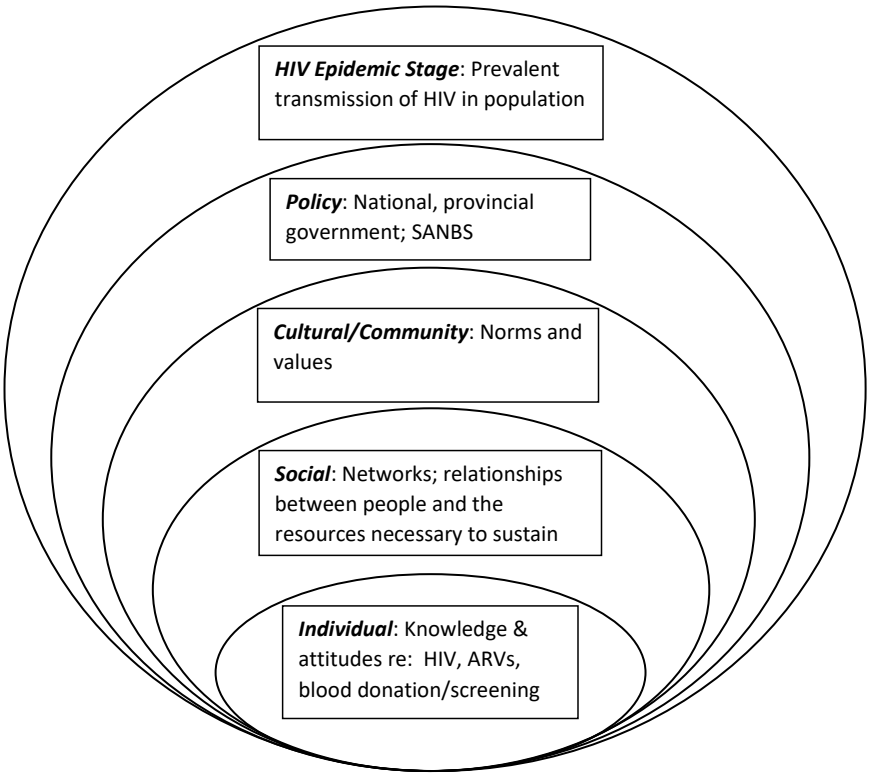

Figure 2: SEM showing levels and potential influences on decision by HIV+/ARV+ individuals to donate blood (adapted from Baral *et al*. 2013[28]).

Sampling and Recruitment

The 122 HIV-positive donors who tested positive for ARV as part of the HIV+/ARV+ prevalence study[23] were eligible for enrolment. Eligibility criteria included: being aged 18 years or older, conversant in English, residing in the accessible areas of the Gauteng, KwaZulu Natal, Mpumalanga or Eastern Cape Provinces, able to complete data collection procedures and provide consent. Initial outreach followed routine SANBS procedures but was performed by trained research staff with prior experience in HIV counseling and study recruitment. All eligible participants were approached for enrolment and invited to complete the ACASI and interview. Participants had the option to consent to either the ACASI or interview or to both procedures.

Data Collection

The ACASI and interviews were administered in person, at a location mutually acceptable to study participants and staff. The survey instrument included, among other elements, a validated 12-item stigma scale.[29] (**Supplementary Table 1 (Annexure 7))** The scale had four domains, each with three items: 1) personalised stigma, 2) disclosure concerns, 3) concerns with public attitudes and 4) negative self-image. All items were answered using a 1-4 point Likert scale, such that higher numbers indicated greater stigma.

The interviews occurred immediately after survey completion, lasted 45-90 minutes, and were audio-recorded with participant consent. These interviews examined the trajectory of HIV testing, diagnosis, and treatment initiation; the impact of HIV; perceptions of HIV transmission risk; perceptions of blood donation and the healthcare system; timing and context of donation; and motivation to donate. Interview questions were open-ended and designed to produce detailed narratives through a story-telling approach, with interviewers probing as needed for full understanding.

Data Analysis

Survey data were extracted to Excel. Responses from stigma subscales two to four were summed to produce an overall score (subscale 1, which focuses on personalised stigma was of questionable utility within this sample. As noted by the scale's developers, high levels of secrecy surrounding serostatus, like those present in our sample, may negatively affect the reliability of questions on personalized stigma [30]). Using sub-scales two to four, the lowest possible score was 12 and the highest 36. Scores were averaged (19.1), and then dichotomized into high (above sample mean) or low (below sample mean) stigma categories. These categories were used to segment qualitative data and search for any patterns in motivation related to stigma.

De-identified verbatim transcripts from all interviews were uploaded to Dedoose ® (a cross-platform application for analysing qualitative and mixed methods data, including text and spreadsheet data)[31] and subjected to thematic analysis.[32] This included inductive, line-by-line analysis to identify themes that emerge from close reading of the text, as well as deductive analysis, which focused on pre-identified themes drawn from the interview guide and relevant academic literature. [33,34]. A codebook was created following accepted procedures.[35] Data were compared across participant gender and stigma score.[34] In addition, we applied narrative analysis to interviewees' accounts of their study qualifying donations (SQDs) to better grasp their experience as a whole.[36]

3.4 RESULTS

Contact details were available for 120 of the 122 potential participants, 62 (52%) of whom were unreachable (either due to incorrect details or not responding to calls); 12 (10%) were ineligible (11 were not conversant in English); 20 (17%) directly or indirectly refused participation; 1 participant enrolled only for the ACASI and was excluded from this manuscript. Hence, a survey and interview were collected from the 25 (21%) consenting participants. (**Table 5**) Participants were predominantly Black African, female, in their 30s, residents of Gauteng province, and had donated at a mobile site. Eight donors (32%) had donated at least one previous donation. Only a third of participants had ever disclosed their HIV status to more than three people. Eleven were classified as perceiving high stigma. Two interviewees (1101M and 2202M) insisted they had not knowingly donated while HIV+/ARV+.

Table 5. Participant demographics, disclosure practices and stigma scores

	N/Median	%/(IQR)*
Total	25	100
Ethnicity		
Black African	23	92
Coloured	2	8
Gender		
Female	18	72
Male	7	28
Donor Type		
First Time	17	68
Lapsed	5	20
Repeat	3	12
Age		
Median	32	(24-39)
Province		
Eastern Cape	2	8
Free State	4	16
Gauteng	10	40
KwaZulu Natal	5	20
Mpumalanga	4	16
Clinic Type		
Mobile	22	88
Fixed Site	3	12
Disclosure Practic**		
Extremely restricted	3	12
Very Highly restricted	5	20
Highly restricted	8	32
Moderately restricted	8	32
Least restricted	1	4
Stigma Scores		
Overall	22	(16-29)
Disclosure Concerns	6	(4-9)
Public Attitude	7	(3-10)
Self-Image	4	(3-6)

*Interquartile range; **Extremely restricted = Disclosed to no one; Very Highly Restricted = Disclosed to one person; Highly Restricted = disclosed to 2-3 people; Moderately restricted = Disclosed to >3 people, but not outside family and friends; Least restricted = Disclosed to >3 people, including beyond family and friends.

Reviewing interviewees' reported motivations for their SQDs, we grouped responses into three themes: 1) altruism, expressed both as a general wish to "save lives", and the specific intention of donating so that blood could be given to other PLWH; 2) a lack of privacy at the donation location, associated with a fear of status disclosure; and 3) other reasons. The latter category included disparate but largely secondary motivations, such as donation to manage a perceived superabundance of blood (i.e. having more than enough blood that could therefore be shared with others), or as a way to confirm HIV status. Here we focus on the first two

themes, as they heavily predominated among interviewees' responses. Notably, very few accounts suggested any kind of test-seeking, only one mentioned incentives and none provided evidence of interviewees believing they had been cured of HIV.

After stratifying interviewees by stigma scale score, and considering the narratives in their entirety, we did not find clear differences in reported motivations. For example, interviewees in both low and high stigma-perceiving categories mentioned: altruistic motivations, privacy-related motivations (including highly restricted serostatus disclosure practices outside of the donation context), and donating as a blood management practice. However, when segmenting interviewees by motivation, those who reported general altruism or privacy concerns as the predominant factor in their donation were evenly split in their stigma perceptions (2 high vs. 2 low; 5 high vs. 5 low, respectively); while those who reported donating blood for other PLWH were more likely to be classified as perceiving low stigma (6 low vs. 2 high). While the overall average stigma score in this data set was 19.2, the averages by motivation were 20.4 for donors concerned with privacy and 18 for those citing altruism. Given the small sample size, the above results are offered in a purely descriptive vein.

As the different stories told by these participants unfold largely along the lines of reported motivations, the following sections segment interviewees by their concern with altruism or privacy to explore the thematic findings in depth.

Altruism Motivated Donors

Overall, altruism was the most commonly reported motivation, mentioned by nearly half of the interviewees (N=12). Frequently framed as the desire to "save lives," this was reported by interviewees from Gauteng, Free State and Mpumalanga provinces, men and women (6 and 6, respectively), and from both stigma categories.

We identified two distinct sub-themes within these accounts. The first was a general wish to help others (N = 4). Some interviewees talked about friends or family members having previously needed or received a transfusion; others reported awareness of the general need for blood. For instance, one man explained, "I wanted to donate blood because I knew my blood type was the most wanted one" and that traffic accidents had caused "a need for blood" (2202M). A woman shared her wish to donate "because my mother was sick and they donated blood for her. So, I thought if I could donate maybe I could help someone else just like they helped my mother" (4402F).

The wish to engage in a more specific form of altruism was expressed by eight interviewees. They were motivated to donate blood so that it could be given to a recipient also living with HIV. Three other interviewees

(1103F, 4402F and 4403F) explicitly raised the possibility that a PLWH might donate for other PLWH, although that was not the primary motivation for their SQD. Talk of donating for other PLWH often drew on notions of "matching," seeming to equate serostatus matching with the matching of blood types required for transfusion. For instance, a male interviewee discussed donating to help "someone else who also has HIV and our blood codes are the same" (4406M). Some interviewees spoke of themselves as being particularly suitable donors for other PLWH, attributing this to their overall health, serological indicators, and/or medication adherence. One interviewee, on ARVs since 2012, said of her blood, "I think it is better than the people who have just found out that they are HIV positive…. because I am drinking the medications regularly and then my health is fine. It will help the people who are HIV positive, especially the ones with low CD4 count" (3306F).

Interviewees' certainty about the feasibility of donating for another PLWH varied, though most were remarkably confident. Eleven of these 12 interviewees reported having been unaware of the deferral of PLWH at the time they donated and spoke of feeling confusion or remorse when they learned of their ineligibility. One interviewee had been explicitly told he was not eligible but continued to believe his donations could help other PLWH. He explained his reasoning: "Blood is blood, whether infected or not. I still believe it can help other people in need" (3305M). The idea that PLWH would be ineligible to donate rarely surfaced in interviewees' accounts of the decision-making that led to their donation, and donating was sometimes framed as a duty: "If someone who is HIV-positive needs blood, … I have to [donate] so that I can help" (3302M).

Privacy-Motivated Donors

The other primary factor raised by interviewees as playing a meaningful role in their decision to donate was a lack of privacy. This was described by over a third of interviewees (n =10), across all provinces, and both stigma categories. Nine were female, one male. There was a mix of eligibility beliefs: some were aware of the deferral for HIV; others were uncertain or seemed not to have considered this possibility. In most cases, interviewees reported that when they encountered the opportunity to donate blood, frequently at school or workplace blood drives, they were with other people (co-workers, classmates, friends, romantic partners) to whom they had not planned to disclose their HIV status. The accounts featured a series of decision points, all experienced as threatening to interviewees' privacy. The first involved *presenting for donation*. Interviewees generally felt unable to opt-out of attempting to donate without prompting questions and raising suspicion. The second revolved around *discussing HIV status or donation eligibility* with SANBS staff. Most interviewees reported there was no private place to have such a conversation (though some had gone to the donation site with precisely this intent), a description which rings true as, especially mobile sites, often lack the infrastructure to ensure private and confidential discussions. The third decision point involved *answering the DHQ*. Interviewees felt unable to disclose their HIV status on the DHQ, either due to the proximity of co-workers and friends, or because they believed the confidentiality of their answers might be compromised. Overall, in

comparison to narratives shared by altruism-motivated donors, these stories were more rooted in the donation context itself. They framed donation as the only safe way out and knowledge of donation eligibility was, in a practical sense, immaterial.

This is well-illustrated by 1106F's account. She explained that SANBS ran a blood drive at her workplace and "everyone, most of the people in the office, they were going to donate…and yah, so I didn't have much of an excuse as to why I shouldn't go" (*presenting for donation*). She thought that "when I get there, I will [be] able to speak, maybe it will be in private ...but it was in the boardroom and you know they had the beds and stuff so we all just filled in the forms in one table" (*discussing eligibility*). Regarding the screening questions, she "didn't answer them truthfully" because her co-workers were close at hand (*answering the DHQ*). In addition, she observed that being deferred from donation attracted undue attention to her as the donor, which was exactly what she was trying to avoid.: "Everyone was looking at each other … it was like a joke because even those who had iron problems and [were] turned away… people were, like, talking like, 'Oh why have [they] been turned away?'" Similar stories were shared by interviewees who had donated with classmates at school, a female donor who had donated with a boyfriend, and a domestic worker taken to donate by her employer.

Though a more private environment might have allowed some of these donors to reveal their HIV status to SANBS staff, for others, additional perceived risks would likely still have precluded disclosure. 1107M noted that even having a question about HIV status on the DHQ, "is like, a violation of my privacy." He elaborated, "If [the form] fall on the wrong hands….my name is there, my ID is there, confidentiality is not there." Thus, in these narratives, fear of inadvertent HIV status disclosure surfaced in multiple ways, and no interviewee reported disclosing their status on the DHQ. Most were untroubled by having withheld this information because they generally reported confidence that testing done by SANBS would identify their donation as being HIV-positive. One female repeat donor explained, "I know how it works...the blood they are taking, it is going to go through these tests… You know hundred percent sure that this blood that I am giving ....is not going to go anywhere, it is not going to be given to anyone" (1105F).

3.5 DISCUSSION

Qualitative research with South African HIV+/ARV+ blood donors revealed that their primary reported motivations were altruism and privacy concerns, with no discernable difference by stigma scores. Among those motivated by altruism, we identified both a general wish to "save lives" and a motivation not previously reported in the literature: the specific desire to donate for other PLWH. Donors motivated by privacy concerns shared quite different accounts, highlighting various elements in the donation process they felt threatened the confidentiality of their HIV status. Far from offering multiple opportunities to exit the process, the experience was framed as a series of decision points in which, in each instance, donation was the only safe option. In this

Discussion we slot our research findings into an adapted version of the SEM that grounded our study, discuss the utility of the model, and offer recommendations for reducing the likelihood of future HIV+/ARV+ donation.

Despite the similar way altruism and privacy were described by interviewees (i.e., as "the reason" for their donation), they are different. Altruistic motivations led interviewees to feel *drawn* to donate, whereas privacy concerns led interviewees to feel *pushed* to donate. This difference led us to locate these factors in different levels of the SEM.

We categorized altruistic motivations as an individual-level influence because they were always constructed as an expression of personal morality. We recognize, however, that all levels of the model are interconnected (hence the dotted lines around them in **Figure 3**). For example, individual altruistic impulses arise within, but are shaped by a cultural context that frames altruism as morally good. Similarly, individual, altruistic motivations may be influenced by blood service policies. For example, blood services implement a policy of minimizing the likelihood of TTIs through multiple strategies. One strategy employed by SANBS is educating potential donors that the blood service should not be used as an HIV testing center, yet to date, however, SANBS has not explicitly disseminated messaging that PLWH are permanently deferred from donation. Considering the U=U (undetectable = untransmissible) campaigns[37] in the media, and successful kidney donations from HIV-positive persons with undetectable HIV viral loads to recipients living with HIV,[38] it should not be surprising that PLWH might consider the same to be possible for blood donation.

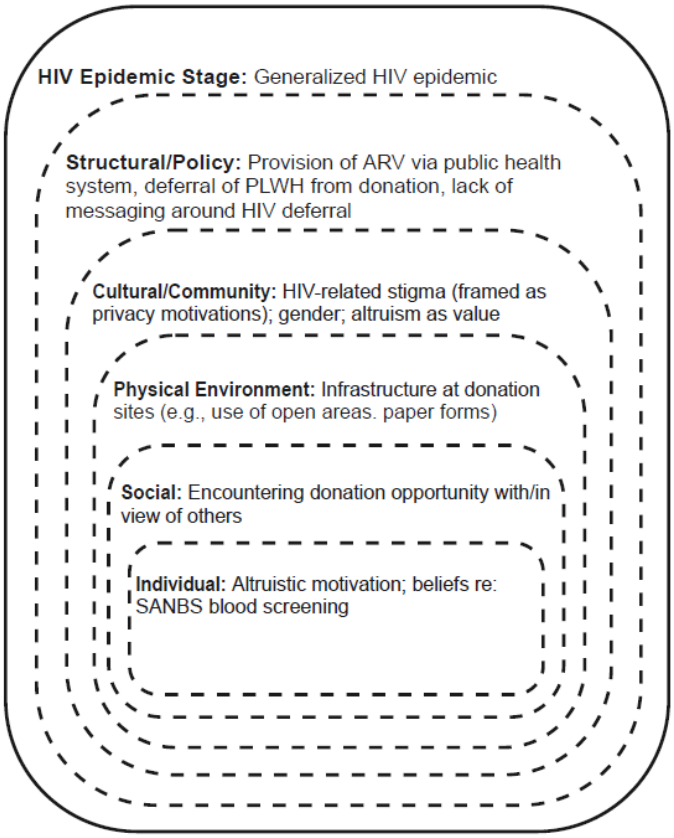

Figure 3: SEM showing levels and influences on decision by HIV+/ARV+ individuals to donate blood (Adapted from Baral *et al*. 2013[28])

In contrast to individual-level altruism, we classified privacy motivations as a cultural factor. This was because interviewees only raised privacy concerns about their HIV status (vs. other personal information), and linked them to fear of unwanted disclosure given the reported lack of privacy and confidentiality encountered while donating. Privacy concerns are therefore best understood as indexing something beyond individually-varying comfort levels: they are reactions to still-pervasive HIV-related stigma in donor communities. Supporting this, the privacy-motivated group had higher average stigma scores than those donating for altruistic reasons (20.4 vs 18.0), though high and low scores were present in both groups. Furthermore, the overwhelming majority of donors with privacy concerns were female. This is of particular importance in a strongly patriarchal South Africa with its predominantly heterosexual epidemic, which disproportionally affects younger, economically vulnerable, females.[39,40]

The cultural factor of stigma (expressed as privacy concerns) intersected with a social-level influence on interviewees' decision-making: HIV+/ARV+ donors often encountered donation opportunities in the presence of other people to whom they had not disclosed and did not wish to disclose, their status. These companions exerted pressure on interviewees' decision-making beyond the cultural influence of stigma because the

imagined potential losses that status disclosure might cause were more concrete and had their own social implications (e.g., loss of a romantic partner, colleagues' esteem).

A further factor added to this complexity: the physical environment and infrastructure at donation locations, especially mobile blood drives. Many interviewees noted that elements such as the use of paper forms and open spaces lacking privacy constituted an obstacle to disclosure. We created a separate level in the model for such considerations (as done in other ecological work in which the physical environment played an important role[41,42]). Since interviewees reported experiencing them as highly proximate, but more fixed than social interactions, we placed this level between social and cultural levels. It is worth noting that SANBS policy actually stipulates that screening, even at mobile drives, be conducted in a private environment. Thus, the lack of privacy, in this case, is a question of *implementation* rather than policy per se.

Policy does influence HIV+/ARV+ blood donation, even if not explicit in the interview data. As mentioned, SANBS's education efforts, aimed at reducing TTI risk, may influence eligibility perceptions and motivations among donors. At a national level, health policies are catalysts for both the deferral of PLWH from donation and the wide availability of ARVs in South Africa. All of the factors discussed heretofore operate within policy structures that grapple with the generalized South African HIV epidemic, which leaves significant proportions of the potential donor pool affected by HIV.[39] The need to balance supply and safety means deferrals of sub-populations perceived to be at-risk for TTI (e.g., MSM, as had been done elsewhere), is simply not possible in the South African context.

We accommodated factors this study found relevant for understanding HIV+/ARV+ blood donation in South Africa within the Social Ecological Model, adapting as necessary (**Figure 3**). We offer this as a heuristic approach upon which we will build (e.g., by incorporating findings related to eligibility beliefs and screening experiences), and that others may find useful for considering HIV+/ARV+ donation in other contexts.

This version of the SEM reveals a complex interaction between individual, social, cultural, and structural/policy factors and multiple pathways to donations by PLWH who take ARVs. Indeed, the nuances of the decision-making in interviewees' narratives cannot be adequately grasped, or responded to, without a multi-level model. In particular, the SEM is helpful in understanding that addressing factors related to HIV+/ARV+ donation at one level, such as individual motivation to help others, may not eliminate the behavior, as influences at other levels (HIV-related stigma and privacy concerns) will still be operant if measures are not taken to mitigate them. For example, someone who was initially motivated to donate for other PLWH might learn this is not possible and no longer wish to donate, but still feel compelled to if encountering a blood drive at their workplace. Thus, an "altruism-motivated" donor could "transform" into a "privacy-motivated" donor if blood

collection infrastructure/procedures and HIV-related stigma have not changed. Furthermore, a more private environment and screening experience might have made a meaningful difference in the comfort with status disclosure for some HIV+/ARV+ donors, but others clearly stated little could be done to mitigate the perceived threat posed by potential status revelation in a broader context of HIV-related stigma

The foregoing notwithstanding, we must mention a major conclusion of our analysis and note that reaching it required us to think beyond the adapted SEM that grounded this study. Adapting a model is typically seen as an appropriate way to attend to research context.[43,44] In this research, adapting the SEM allowed us to focus on HIV risk, which was both helpful and somewhat obfuscatory. This tailoring allowed us, for example, to consider the nature of South Africa's HIV epidemic, but it also led us to implicitly conceptualize "HIV+/ARV+ donors" as unique, rather than prompting us to ask what they might share with other donors, or how their behavior might be similar to that exhibited in other contexts. Despite this, commonalities emerged. For example, a large group of our interviewees reported altruistic motivations for donation. Activators for altruism were varied and included donating because of loved ones, knowledge of blood shortages, and a moral duty to donate, including specifically for other PLWH. Though donating for other PLWH is, as far as we know, a novel finding, the other reasons are indistinguishable from those offered by many donors *not* living with HIV, both in South Africa[45,46], and elsewhere[47]. We came to realize that the expectation that HIV+/ARV+ donors' motivations *would* be different derived from assumptions that, in some cases, were not supported by data (e.g., HIV+/ARV+ donors know they are ineligible to donate; such donors would not consider their blood helpful to others). For those donors who donated prior to HIV acquisition, it makes little sense to expect their donation motivations, post-HIV diagnosis and treatment initiation, to be different than they had been historically, especially given the messaging around "U=U" and HIV being just "another chronic disease".[37,48]

In addition, though reports of HIV+/ARV+ blood donations were initially surprising, looking beyond the specific context of blood donation suggests perhaps they should not have been. Non-disclosure of health information, including HIV status and ARV use, in other medical settings, is well described. Failure to disclose general medication use, even upon direct questioning by their clinicians, was reported in up to 15% of patients in the USA.[49] Furthermore, non-disclosure of known HIV status and ARV use have been confirmed in several African household surveys[50,51] and in HIV and ARV research programs.[52-54] ARV denial was reported in as many as one in three participants in a study validating self-reported ARV use in rural South Africa.[55] These trends should have led us to *expect* status disclosure in a semi-public setting to be problematic, even though other HIV+/ARV+ donors have disclosed their status.[56,57] What may warrant more investigation is the conditions under which some would-be donors living with HIV do disclose their status.

From the discussions with the participants and the main themes identified in this study, certain recommendations to limit HIV+ARV+ donations emerged. Specifically, these include: 1) Improve the likelihood that PLWH are aware of their permanent deferral from blood donation, as a way to reduce the potency of altruism as a motivator. Historical blood donor messaging relating to HIV centered on the risk of donation during the HIV "window period" and donation sites not being used as HIV-testing sites. This should be augmented with clear communication on the ineligibility of PLWH as blood donors. It is crucial that this be conveyed in a manner that will not further stigmatize those living with the virus. Crafting effective messaging and identifying appropriate channels for dissemination should be done in collaboration with PLWH. 2) Develop procedures to assist those who feel unable to opt-out of donation due to peer pressure and privacy concerns. These could include systems for donors to confidentially withdraw their donations directly after donation or providing donors with a "palatable" option to explain their potential deferral to those observing their donation process. The latter would still require improved privacy infrastructure conducive to confidential discussion at donation sites. Enforcement and compliance monitoring of existing privacy policies, especially at often-used facilities need to be further strengthened.

Our study had several limitations, including the potential for selection bias. We recruited participants from a relatively small pool of HIV+/ARV+, English conversant, South African blood donors. Those who were not interviewed may have had meaningfully different experiences that are not represented here. Furthermore, we tried to reduce potential social desirability bias through assurance of anonymity, personal safety and the use of open-ended questions. While these measures might not have been entirely successful (two interviewees refused to acknowledge awareness of HIV+/ARV+ status at the time of donation), interviewees did recount behavior often considered socially undesirable, suggesting they felt sufficiently comfortable at some level to share such responses.

To our knowledge, this is the first attempt at investigating the motivations driving donations by HIV+/ARV+ donors. As appropriate for highly exploratory, qualitative research, we make no claims of exhaustiveness and instead offer the significant convergence of themes we found around altruism and privacy as a starting point on which to build. Researchers and professionals should critically consider how these findings may apply in different contexts (national, cultural, and types of epidemics). We believe our findings may well have utility in other settings, as HIV is a relatively stigmatized infection globally, blood donation requires fundamentally similar processes regardless of national context, and our findings dovetail with those from other research on disclosure of healthcare information in general.[49-53]

3.6 CONCLUSION

The phenomenon of blood donation by HIV+/ARV+ has been documented in two contexts.[22,24] Though its global prevalence is unknown, there is little reason to assume that it is not occurring more widely. Our research uncovered complex, diverse motivations related to privacy and altruism leading to HIV+/ARV+ blood donations. The growing HIV+/ARV+ populations both in South Africa and elsewhere and the increasing uptake of pre-exposure HIV prophylaxis may well result in increasing numbers of such donations unless actively managed. To reduce such donations, we need a better understanding of why they are occurring. Here we have reported only on the main donor motivations associated with these non-compliant donations, which is but a small component of this complex phenomenon. We urge other scholars to assess the occurrence of this phenomenon in other settings and further elucidate the motivations and contexts leading or contributing thereto.

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CHAPTER 4: THE IMPACT OF NON-DISCLOSURE OF HIV STATUS AND ANTIRETROVIRAL THERAPY ON HIV RECENCY TESTING AND INCIDENCE ALGORITHMS

Karin van den Berg[1,2,3], Edward L Murphy[4,5], Gary Maartens[6], *Vernon J Louw[2], *Eduard Grebe[5]

1. Medical Division, South African National Blood Service, Roodepoort, South Africa
2. Division of Clinical Haematology, Department of Medicine, University of Cape Town and Groote Schuur Hospital, Cape Town, South Africa
3. Division of Clinical Haematology, University of the Free State, Bloemfontein, South Africa
4. Departments of Laboratory Medicine and Epidemiology/Biostatistics, University of California, San Francisco, California, USA
5. Vitalant Research Institute, San Francisco, California, USA
6. Division of Clinical Pharmacology, Department of Medicine, University of Cape Town, Cape Town, South Africa

* Joint senior authors

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Chapter Overview:

As mentioned in the previous chapter, interventions aimed at reducing the risk of transfusion transmitted infections (TTI), especially HIV, have for decades been a key focus area of blood transfusion services (BTS) globally and even more so in South Africa with our generalised HIV epidemic. Early in the evolution of the HIV epidemic, BTS monitored the seroprevalence among blood donors as a measure of the success of such interventions. With improved testing methods, BTS were less concerned with prevalent infections, which would be interdicted by the robust testing, and became more concerned with incident cases. Traditional monitoring of incidence is significantly problematic in the blood donation setting and by implication limits such assessments to repeat donors while the greater TTI risk lies with first time donors. Various methods were developed to assess the residual risk of transfusion transmitted (TT) HIV, many of which rely on accurately identifying recently acquired HIV cases. However, such identification is heavily impacted by EC state as well as importantly ARV use, especially if such ARV use is undisclosed.

Given my findings of significant undisclosed ARV use among HIV-positive donors, as discussed in chapters two and three, there was a need to establish how HIV+/ARV+ donors affect recency testing and incidence modelling. I developed a novel approach constructing four progressively more comprehensive resent infection testing algorithm (RITA) and applying test results from our 2017 HIV-positive donor cohort to each RITA. Separately, I calculated context-specific mean duration of recent infection (MDRI) and false recent rate (FRR) for each RITA. The outcomes of the four RITA were then used to run an open access incidence model and the results compared. As MDRI and FRR estimations are per se also complex, we also compared the results obtained from the above method to that when using previously published MDRI and FRR (from a similar population). I confirmed significant misclassification of recent cases when using only current blood donation

testing algorithms. ARV testing in addition to VL testing did not materially improve either the identification of recent cases or on incidence estimates. Compared to the published MDRI and FRR, the models using context-specific MDRI and FRR resulted in progressively lower incidence estimates, albeit not statistically significant. This work is, to the best of my knowledge, the first time this approach was used to evaluate recency testing and incidence modelling.

I presented this work at the 2023 Association for the Advancement of Blood & Biotherapies meeting in Oct 2023 in Nashville, Tennessee, USA, where it was selected as one of five plenary oral presentations. I was also an invited speaker at the 34th Regional Congress of the International Society of Blood Transfusion in Cape Town, South Africa in November 2023 where my topic, Non-disclosure of ARV therapy by blood donors – implications for blood transfusion services, included some of the work in this chapter.

4.1 ABSTRACT

Background:

Accurate HIV incidence estimates among blood donors are necessary to assess the effectiveness of programs aimed at limiting TT HIV. We assessed the impact of undisclosed HIV status and ARV use on HIV recency and incidence estimates using increasingly comprehensive recent infection testing algorithms.

Materials and Methods:

Using 2017 donation data from first-time and lapsed donors, we populated four HIV recency algorithms: 1) serology and limiting-antigen avidity testing, 2) with ID-NAT added to Algorithm 1, 3) with viral load added to Algorithm 2 and 4) with ARV testing added to Algorithm 3. MDRI and FRR were calculated for each algorithm and used to derive and compare incidence estimates.

Results:

Each increasingly comprehensive algorithm misclassified fewer donors as recent when compared to Algorithm 4: Algorithm 1: 61 (12.1%) misclassifications; Algorithm 2: 14 (2.8%) misclassifications; Algorithm 3: 3 (0.6%) misclassifications. Incidence estimates were marginally lower for each progressive algorithm: Algorithm 1: 0.19% per annum (pa) (95% CI: 0.13-0.26%); Algorithm 2: 0.18% pa (95% CI: 0.13-0.22%); Algorithm 3: 0.17% pa (95% CI: 0.13-0.22%); Algorithm 4: 0.17% pa (95% CI: 0.13 - 0.21%).

Conclusion:

We confirmed significant misclassification of recent HIV cases when not including viral load and ARV testing. Context-specific MDRI and FRR resulted in progressively lower incidence estimates but did not fully account for the context-specific variability in incidence modelling. The inclusion of ARV testing, in addition to VL and ID-NAT testing, did not have a significant impact on incidence estimates.

Keywords: Blood donation; HIV disclosure; anti-retroviral agents; HIV recency algorithms; HIV incidence estimation.

Highlights:

- Undisclosed positive HIV status and ARV use among blood donors impact recent infection testing algorithms
- Context-specific MDRI and FRR improve accuracy of incidence estimates
- The inclusion of ARV testing, in addition to VL and ID-NAT testing, did not have a significant impact on incidence estimates.

4.2 INTRODUCTION

Forty years after the world became aware of HIV, the prevention of TT HIV infections remains a key focus for blood services. Early in the epidemic, the success of such mitigation strategies could be evaluated by monitoring the prevalence of HIV-positive donations. However, in response to increasingly sensitive HIV testing assays, new methods for assessing HIV transmission risk and the success of prevention strategies were developed including models to calculate the residual risk of HIV transmission.[1]

In most instances, these models were based on identifying incident cases which might escape serologic detection during the "window period". Initial models focused on repeat donors only.[2] Subsequently, residual risk models were developed that included both repeat and first-time donors with recently acquired/incident HIV infections.[3] Such incident cases included those that were viremic but HIV-1 seronegative as well as those classified as 'recent' using a sensitive/less-sensitive HIV-1 enzyme immunoassays.[4] This marked the beginning of contemporary recent infection testing algorithms (RITA). Simultaneously, such RITA were used in progressively more complex population-level incidence models.[5,6]

Multiple assays for detecting recent versus non-recent infections were developed and evaluated in various settings and populations.[7] For a recency assay to be useful in modelling incidence estimates, it must have a sufficiently long MDRI and a low FRR.[7] Factors such as variable immune response and HIV subtypes complicate establishing a 'universal' MDRI with FRR significantly affected by elite controller status and ARV use.[8] Until recently, blood donation by persons living with HIV and already on ARV therapy was not recognized. As a result, little consideration was given to the potential impact of undisclosed HIV status and ARV use on RITA.[9] The identification of 'false elite controllers' and the substantial prevalence of undisclosed HIV status and ARV use among blood donors in South Africa[10,11] and elsewhere[12] means that blood services may need to consider more robust RITA to maximise the precision of incidence estimates.

The requirements for accurate and precise HIV incidence estimation models are onerous, especially in resource-constrained settings. Calibrating context-specific MDRI and FRR requires sophisticated statistical knowledge and detailed, recent HIV epidemiologic information. Recency assays e.g., the Limiting-Antigen Avidity Enzyme Immunoassay (LAg)[13], viral load (VL) assays and ARV testing are expensive and may require specialised laboratory testing. The degree to which uncertainty for each of these factors impacts incidence estimations, especially in the blood donation setting, is unknown. We therefore assessed and compared HIV recency and incidence estimates using increasingly comprehensive RITA.

4.3 METHODS

Study setting, design and population

In South Africa, strategies to minimize the risk of TT HIV include pre-donation education, the completion of a lifestyle and health questionnaire, one-on-one interviews and testing for HIV antibody by serology and for RNA by ID-NAT.

Utilising stored samples from our previous study[9], we performed additional testing to populate increasingly comprehensive RITA. All first-time and lapsed blood donors (those whose last donation was more than 12 months ago) who presented during the 2017 calendar year were included. Ethics approval was obtained from the SANBS and the University of Cape Town Institutional Review Boards.

Sampling

The SANBS Biorepository routinely stores plasma aliquots of HIV-positive blood donations at -30°C. For this study, samples from first-time and lapsed donors who donated during 2017 and who tested HIV-positive (RNA, antibody, or both) were included in the analysis. During 2017, LAg and ARV testing was performed on all HIV positive donations as part of an earlier study.[11] Due to budgetary constraints, HIV VL testing was performed on a limited subset of 441 specimens, including all HIV RNA positive, seronegative (23) as well as randomly selected RNA positive, seropositive donations (418).

Testing

SANBS routinely performs serological assays and ID-NAT for HIV, Hepatitis B and C in parallel. HIV serology testing was performed using the Abbott Prism HIV1/2® (Abbott Diagnostics, Delkenheim, Germany). The Ultrio Elite® multimarker probe assay (Grifols Diagnostics, Barcelona, Spain) on the Procleix Panther® platform was used for the ID-NAT testing. All HIV-seropositive donations were subjected to recency testing using the Sedia® HIV-1 LAg-Avidity EIA test (Sedia Biosciences Corporation, Portland, Oregon). HIV-positive donations with sufficient residual plasma underwent ARV testing using a validated, high-performance liquid chromatography tandem mass spectrometry assay performed at the Division of Clinical Pharmacology Laboratory, University of Cape Town. The details of these tests have been described previously.[11] VL quantification, using the Abbott RealTime HIV-1® VL assay on the m2000 system (Abbott Molecular Inc., Des Plaines, Illinois), was performed on the 441 specimens noted above. HIV-seropositive but ID-NAT negative donations were assumed to have VL below the lower limit of quantification of the Abbott RealTime HIV-1® VL assay.[14,15]

Donations were classified as follows: antibody-positive (Ab+) if the sample was serology positive; ID-NAT positive (NAT+) if HIV RNA was detected; LAg recent if the sample had a normalised optical density (ODn) <1.5; ARV positive if it tested positive for one or more ARV. Donations were dichotomized as either high or low VL using a 75 copies/mL cut-off. A relatively low VL cut-off, such as 75 copies/mL used by Kassanjee *et al.*[16],

ensures a sufficiently long MDRI with little impact on the FRR and allows for comparison across different settings.

Model and model parameters

We constructed four, progressively more comprehensive recency algorithms. Algorithm 1 included only HIV serology and LAg results. Subsequent algorithms added new test results cumulatively: for Algorithm 2 we included ID-NAT which allowed for the classification of NAT+/Ab- donations as recent and NAT-/Ab+ donations as non-recent; Algorithm 3 included a VL cut-off of 75 copies/mL with donations with a VL of <75 copies/mL classified as non-recent. Algorithm 4 included ARV testing with donations with detectable ARV classified as non-recent.

We employed version three of the South African Centre for Epidemiological Modelling and Analysis Assay-Based Incidence Estimation (ABIE_V3) toolbox[6] to perform our incidence estimates. The model requires inputs for the post-infection time cut-off *T*, the estimated MDRI and the estimated FRR. The MDRI, given a specific time cutoff *T*, is defined as the average time an HIV-infected person spent both alive and "recently" infected.[6] The FRR refers to the proportion of HIV-infected persons infected longer than *T* who are classified as recently infected by the RITA.[5,6,17] The 2022 WHO technical guidance on HIV recency assays[8] recommends a 2-year period for time cutoff T and that context-specific MDRI and FRR values be derived when employing assay-based incidence estimation.

We therefore estimated context-specific (for South African blood donors during 2017) FRR and MDRI values for each of the previously described algorithms. MDRI was estimated using HIV-1 subtype C calibration data from the CEPHIA consortium and a web-based tool from UNAIDS and WHO (https://worldhealthorg.shinyapps.io/recency_test_properties/). (**Table 6**) MDRI estimates accounted for the window period associated with the HIV screening strategy employed in each algorithm (ID-NAT and/or serology), viral load threshold, the average delay between HIV infection and diagnosis in the population and the proportion of HIV-infected donors who are receiving ART. FRR estimates accounted for ART coverage and the proportion of HIV-infected donors who are virally suppressed. ART coverage and viral suppression in the donor population were estimated using ARV testing and quantitative viral load data. In addition, we constructed a "reference" model for each algorithm using the context-specific FRR (1.3%) and MDRI (177 days) values published by Kassanjee *et al.*[10] for the subset of South African samples included in their study which excluded patients on ARV and elite controllers. Finally, we used the calculated FRR and MDRI values for each of these algorithms and the number of donations classified as "recent" using the methods described above to calculate incidence estimates for each algorithm.

Table 6. Model parameters used in the incidence calculations

Parameters:	**Kassanjee Reference Algorithm**	**Algorithm 1**	**Algorithm 2**	**Algorithm 3**	**Algorithm 4**
Post-infection time cut-off T	730	730	730	730	730
HIV screening	Western blot	Ab	Ab & NAT	Ab & NAT	Ab & NAT
Screening adjustment	Base	-17	-3	-3	-3
Recency test	LAg	LAg	LAg	LAg	LAg
LAg Odn	1.5	1.5	1.5	1.5	1.5
VL	Yes	No	No	Yes	Yes
VL threshold	75	None	None	75	75
ARV testing	No	No	No	No	Yes
Adjustment for treatment	No	No	Yes	Yes	Yes
Mean time to treatment	No	N/A	4.9 years	4.9 years	4.9 years
HIV Subtype	100% C	100% C	100% C	100% C	100% C
MDRI (unadjusted for screening method) (days)	N/A	204.7	204.7	192.3	192.3
MDRI (adjusted) (days)	177	188.7	201.7	188.3	188.3
MDRI RSE	0.05	0.074	0.069	0.069	0.069
FRR Untreated*	1.3	1.2	1.1	1.1	1.1
FRR Treated*	Excluded	56.6	N/A	N/A	0
FRR Treated and Suppressed*	Excluded	N/A	0	0	N/A
FRR Treated and Unsuppressed*	Excluded	N/A	56.6	56.6	0
FRR weighted*	1.3	6.6	3.5	1.6	1.0
FRR RSE*	20	50**	50**	50**	50**

Ab: Antibody; NAT: individual nucleic acid amplification test; LAg: limiting antigen avidity test; ODn: normalised optical density; VL: viral load; MDRI: mean duration of recent infection; RSE: relative standard error; FRR: false recency rate.
* Expressed as percentage; ** Assumed RSE, approximately correct.

Model assumptions:

The underlying assumptions applied in this analysis included: 1) The majority of people living with HIV in South Africa are diagnosed and take up treatment later than the specific time cutoff *T*; therefore testing positive for ARV denoted a longstanding infection; 2) Viremic control (VL <75 copies/mL), whether due to natural immune responses or ARV use, would only occur after the specific time cutoff *T* and therefore denotes longstanding infection; 4) For Algorithm 3, we computed a viral suppression rate based on VL <75 copies/mL among the ARV treated group and 5) for Algorithm 4 we assume ARV testing "perfectly" detects treatment.

4.4 RESULTS

A total of 207,768 first-time and lapsed donors were accepted for blood donation during 2017 of whom 969 (0.60%) were confirmed as HIV-positive (**Figure 4**). The LAg, VL and ARV results of the 506 HIV-positive donors for whom a complete set of results were available, are shown in **Figure 5.** Among the 65 Ab+/NAT- donors, 24 (36.9%) tested LAg recent and 56 (86.2%) had detectable ARV levels. A fifth (87/418) of the Ab+/NAT+ donations tested recent by LAg of whom 11 (12.6% of 87) had VL <75 copies/mL. Seven of these 11 donations had detectable ARV concentrations compared with three of the 76 (4.0%) donors with VL >75 copies/mL. In the Ab-/NAT+ group, four (17.4%) of the 23 donors had VL <75 copies/mL and none tested positive for ARV.

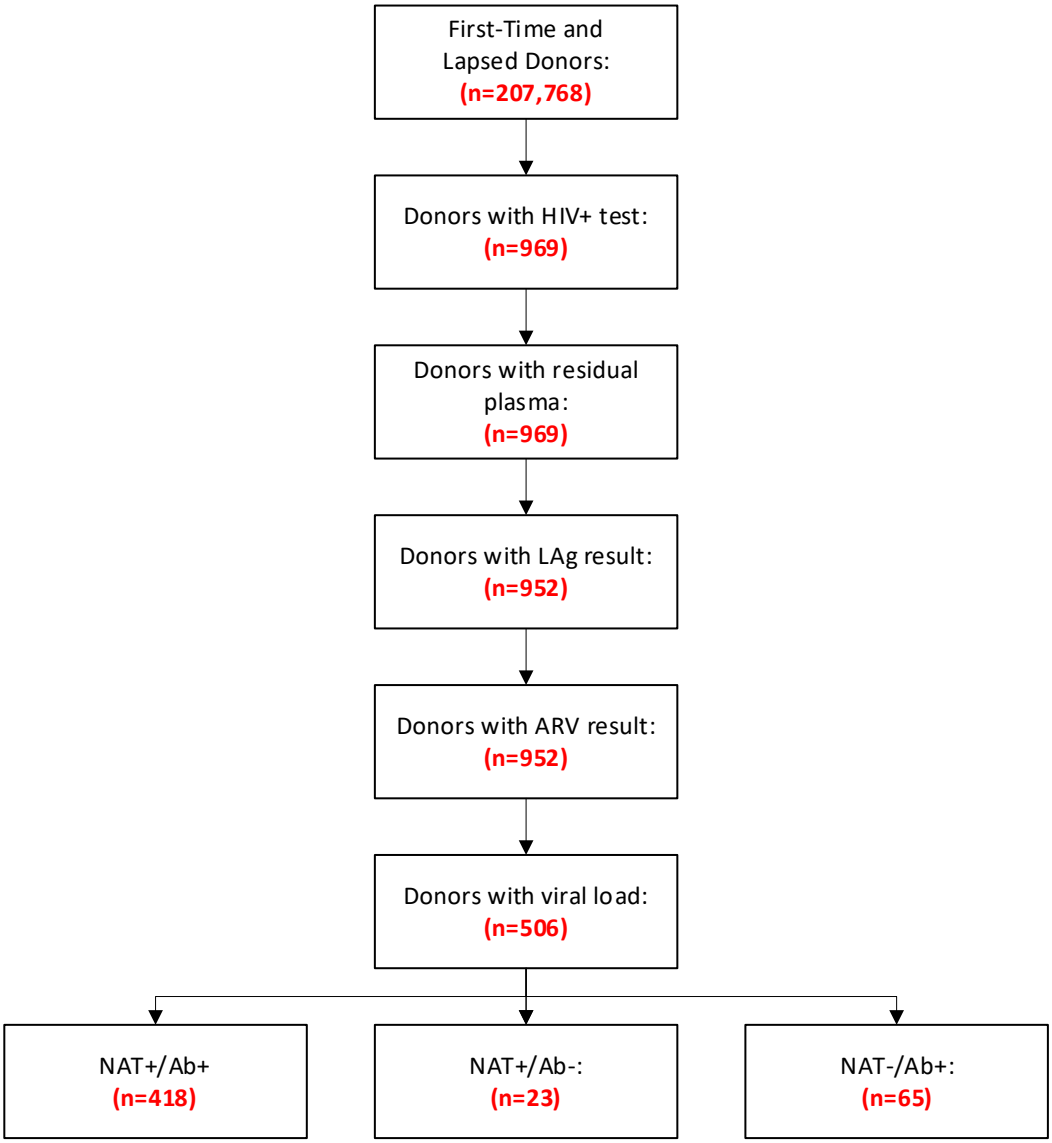
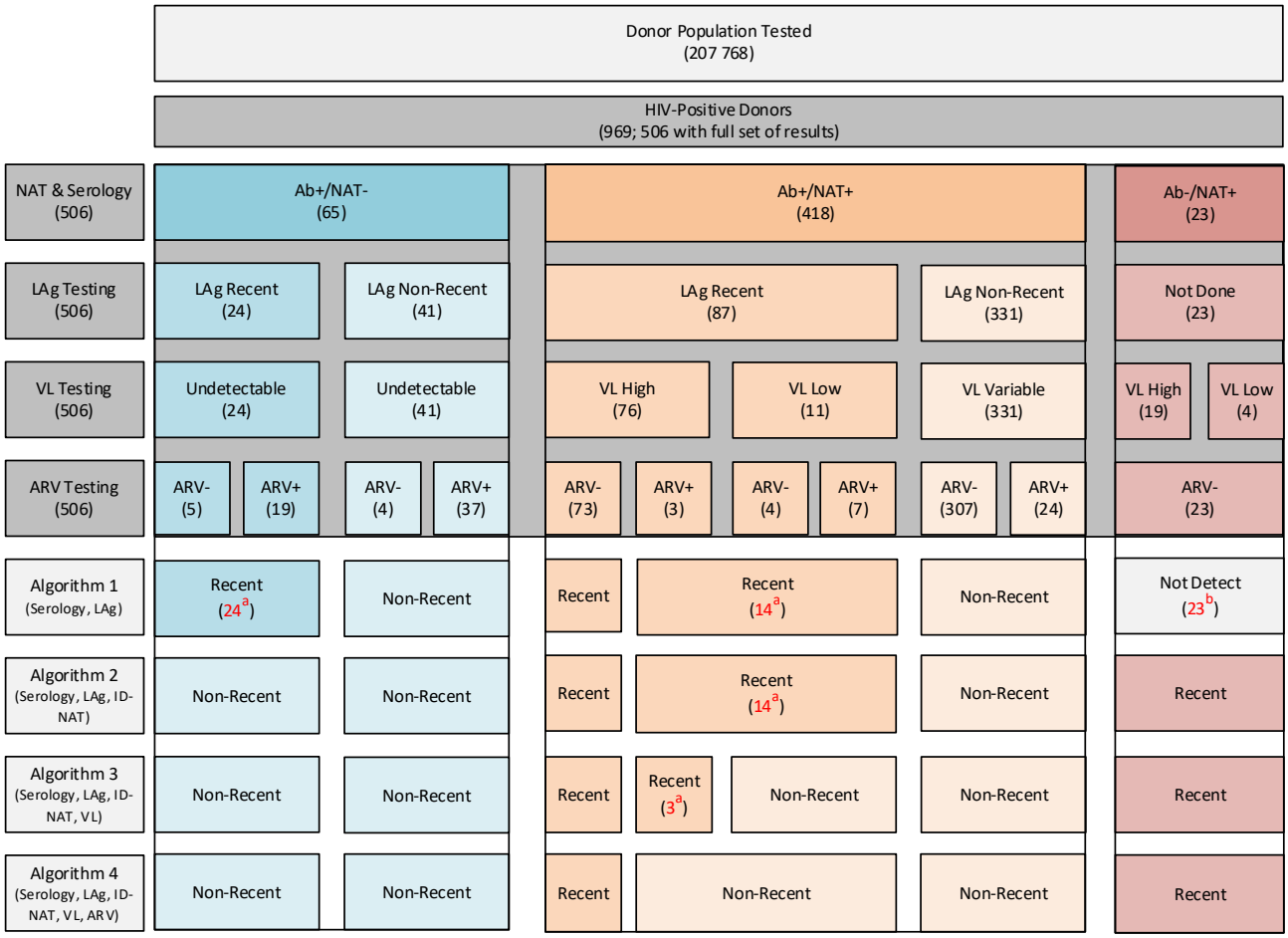


Figure 4: Flow diagram of available test results for the 207,768 first-time and lapsed donors who donated during January to December 2017.



[a] Donations incorrectly classified as "recent"; [b] Donations not detected as HIV-positive.

Figure 5: Testing algorithms and their outcomes applied to the 506 HIV-positive donations included in the analysis.

In comparison to Algorithm 4, Algorithm 1, which classified 111 (21.9%) of the 506 donors as recent, (**Figure 4, Table 7**) incorrectly classified 24 (36.9%) of the 65 Ab+/NAT- donors and 14 (3.4%) of the 418 Ab+/NAT+ donors as recent while failing to detect the 23 of the Ab-/NAT+. Adding ID-NAT (Algorithm 2) results in 110 (21.7%) donors being classified as recent; all 65 Ab+/NAT- donors were correctly classified as non-recent and all 23 Ab-/NAT+ donors were classified as recent. However, 14 (3.4%) of the 418 Ab+/NAT+ donors were still incorrectly classified as recent. Addition of the VL cut-off in Algorithm 3 reduces the incorrectly classified donors to three (0.7%). Algorithm 4 classified 73 (17.5%) of the 418 Ab+/NAT+ and all 23 Ab-/NAT+ donors, i.e. 96 (19.0%) of the 506 donors as recent.

Table 7: Donor counts for HIV incidence modelling derived from each testing algorithm.

Donor Parameters	Algorithm 1	Algorithm 2	Algorithm 3	Algorithm 4
HIV-negative	206822	206 799	206 779	206 799
HIV-positive	946	969	969	969
Tested for recency	483	506	506	506

Classified recent	111	110	99	96
Total Sample Size	207 768	207 768	207 768	207 768

Table 8 compares the outcomes of the calculated incidence estimates for each of the algorithms described above using first the MDRI and FRR published by Kassanjee *et al.*[4] and then separately the MDRI and FRR derived specifically for each algorithm. The estimated incidence for Algorithm 1, using the algorithm specific calculated MDRI and FRR compared to the published "Kassanjee" MDRI and FRR, is slightly lower (0.19% pa versus 0.22% pa) with a slightly wider 95% CI (0.13-0.26% versus 0.17 - 0.26%) but a substantially larger relative standard error (RSE) (15.91% versus 10.79%). The same pattern of slightly lower incidence estimates in the calculated algorithms held true for each of the algorithms when compared to its "Kassanjee" equivalent, although the difference in the RSE decreased with each more comprehensive algorithm, denoting improved precision.

Table 8: Outcomes of incidence calculations using a) "Kassanjee" and b) context-specific MDRI and FRR

	"Kassanjee" MDRI & FRR (95% CI)		**Context-specific MDRI & FRR (95% CI)**	
ALGORITHM 1 (Serology & LAg)				
Estimated incidence	0.22% pa	(0.17-0.26%)	0.19% pa	(0.13 - 0.26%)
RSE of incidence estimate	10.79%		15.97%	
ALGORITHM 2 (Serology, LAg, ID-NAT)				
Estimated incidence	0.21% pa	(0.16-0.25%)	0.18% pa	(0.13 - 0.22%)
RSE of incidence estimate	10.9%		13.39%	
ALGORITHM 3 (Serology, LAg, ID-NAT, VL)				
Estimated incidence	0.19% pa	(0.14-0.23%)	0.17% pa	(0.13-0.22%)
RSE of incidence estimate	11.47%		12.73%	
ALGORITHM 4 (Serology, LAg, ID-NAT, VL, ARV)				
Estimated incidence	0.18%	(0.14-0.22%)	0.17% pa	(0.13 - 0.21%)
RSE of incidence estimate	11.65%		12.51%	

MDRI: mean duration of recent infection; FRR: false recency rate; CI: confidence interval; LAg: limiting antigen avidity test; pa: per annum; RSE: relative standard error; ID-NAT: individual nucleic acid amplification test; VL: viral load; ARV: antiretroviral testing

We next compared the incidence estimates when using algorithm specific calculated MDRI and FRR with each other. When including ID-NAT testing (Algorithm 2) to serology and LAg testing (Algorithm 1), the estimated incidence decreased marginally from 0.19% pa to 0.18% but with a tightening of the 95% CI (0.13 - 0.26% versus 0.14 - 0.24%) and a decrease in the RSE from 15.97% to 13.39%. (**Table 8**) Subsequent addition of a VL cut-off (Algorithm 3) decreased the estimated incidence to 0.17% pa with a further narrowing of the 95% CI to 0.13-

0.22%, and a decrease in the RSE to 12.73%. The addition of ARV testing (Algorithm 4) yielded no change in the estimated incidence (0.17%) and almost no change in the 95% CI (0.13 - 0.21%) and a marginal decrease in RSE (12.51%).

4.5 DISCUSSION

Our study demonstrated a significant misclassification (61 of 506, 12.1%) of the recency status of HIV-positive donors when using a RITA that does not include ID-NAT, VL and ARV testing. Without ID-NAT, the 4.6% ID-NAT positive but seronegative donors were not identified as HIV positive. ID-NAT testing reduced the recency misclassification to 2.7% with marginal additional improvement to 0.6% provided by VL testing. The inclusion of ARV testing had a near negligible improvement in the identification of recent cases.

We confirmed a trend of decreasing incidence estimates with each increasingly comprehensive RITA. However, the use of context-specific MDRI and FRR resulted in only marginally lower incidence estimates (none of which were statistically significant) compared to the published "Kasssanjee" estimates. The "Kassanjee" estimates used in this analysis were also derived from South African blood donors but who donated prior to 2014 and for whom an HIV lysate-based Western blot assay was used to identify HIV-positive cases. In contrast, this analysis used a chemiluminescent immunoassay (Abbott Prism HIV1/2®) either alone or in combination with ID-NAT to identify HIV-positive cases. The use of ID-NAT resulted in an extended MDRI but simultaneously identified sero-negative recent cases. The net effect of which were remarkably, but co-incidentally, similar to that of the Kassanjee group.

Current recommendations[8] include the use of context-specific calibrated MDRI and FRR and multi-assay RITA aimed at limiting the impact of viral suppression, either due to elite controller status or ARV use, on FRR. The addition of a VL cut-off to differentiate between "true" and "false" recent cases has been the backbone of newer RITA.[18] However, the ongoing roll-out of ARV therapy and the not infrequent failure to disclose ARV use[19-21] have raised questions on the need to include testing for ARV in RITA to ensure the exclusion of persons on ARV with incomplete viral suppression.[22]

In our setting, a blood transfusion service in a country with a background HIV prevalence of ~13.9%[23] and estimated national incidence of 0.48% (in 2017)[24], ID-NAT testing was the biggest contributor in identifying misclassified cases as it identified both cases that had yet to seroconvert (and therefore assumed to recent) as well as seropositive donations with extremely low VL (below ~18 copies/mL)[3] (and therefore assumed to be longstanding infections), a third of whom tested LAg recent in Algorithm 1. However, in settings with significantly lower prevalence and especially lower incidence, the impact of ID-NAT versus simple VL testing

may be less obvious as, once identified as HIV-positive, VL testing would identify the overwhelming majority of HIV-positive donors with low VL, irrespective of the reason for the low VL.

Conversely, the use of ID-NAT only to identify recent cases is also problematic. In their publication on the HIV incidence among South African blood donors, Vermeulen *et al.* [25] essentially applied Algorithm 2 (ID-NAT and LAg) to identify recently infected HIV cases in their "LAg first-time donor" model and relied solely on ID-NAT to identify recent cases in the "NAT Yield Window Period" and "Classic Incidence/Window Period" model. Given the outcome of this study, it is likely that the "LAG first-time donor" model would have over-estimated incidence as it likely did not exclude LAg recent donors who had low, but detectable VL and those on ARV with poor viremic control. In contrast, the two models that rely solely on ID-NAT to identify recent infections, likely underestimated incidence as ID-NAT positive, seronegative cases accounted for only 23 of the 96 recent cases in our study.

In our study, the only benefit of adding ARV testing (in addition to ID-NAT and VL) was the identification of those donors on treatment that were not virally suppressed. Recognizing that NAT testing potentially introduces an overly low VL cut-off, the Transfusion Transmissible Infections Monitoring System program in the USA considered a RITA that included VL (but not ARV assessment) for defining incident HIV cases.[26] Their assessment, based on a cut-off of 1000 copies/mL, was that such cases would be unlikely in their donor population and as a result, did not include it in their final analysis. However, a contemporaneous publication by Custer *et al.*[12] confirmed that approximately 15% of HIV-positive donors from their regions in the USA were on ARV at the time of donation. Given the overlap between these two studies, there may have been an underappreciation of the impact of low VL and ARV use on identifying recent HIV cases. These findings would suggest that blood services should carefully consider RITA that sufficiently account for potential undisclosed ARV use.

In addition, we demonstrated a decreasing, more precise incidence trend when applying increasingly comprehensive RITA in combination with context-specific MDRI and FRR. A similar decreasing incidence trend was seen when using the RITA outcomes with the published "Kassanjee" MDRI and FRR, but in all instances these incidence estimates were higher than those calculated with the context-specific MDRI and FRR. While the difference in incidence estimates did not reach statistical significance, likely due to an underpowered sample size, the differences have epidemiologic importance. In a country such as South Africa with high HIV prevalence, the 0.05% per annum difference in estimated incidence between the "Kassanjee" Algorithm 1 and the "Calculated" Algorithm 4 may be of programmatic interest.

The outcome of these incidence calculations confirmed that accurate identification of recent infections will impact incidence calculations. However, it also highlighted that previous assertions that introducing a standardised time to "cut-off T" and the use of sophisticated, context-specific derived FRR and MDRI should offer "the opportunity to consistently account for imperfect accuracy and precision of the incidence estimator"[6,27] did not quite materialize. The fact that it did not, confirms that we do not fully understand the impact of each test on incidence calculation and that there may be other factors affecting recency not yet fully elucidated.

Our study had limitations. We had a full set of results on only 40% of the 969 lapsed and first-time donors who tested HIV positive during 2017. In particular, the sample included all the RNA-positive, seronegative and a disproportionate number (65 of 75 (86.7%) of the RNA-negative, seropositive donors. (Data not shown) This may have impacted the incidence calculations, especially of Algorithm 1 which incorrectly classified nearly 40% of these cases as recent. In addition, the inclusion of this group of donors resulted in a higher ARV use estimation for the overall sample as this group is known to have high levels of undisclosed ARV use.[10,11] However, this would not have had a material impact on the algorithms other than Algorithm 1 as the remainder of the algorithms correctly classified these cases as longstanding. Furthermore, we assumed that all persons with VL below 75 copies/mL and all those on ARV had longstanding infections. It is conceivable that some true elite controllers may have attained viremic control within the "cut-off time T" and therefore misclassified. Likewise, donors could have contracted HIV, been diagnosed, and started treatment within this same timeframe. Although this was a minor concern in 2017 when South Africa began to adopt the "test and treat" approach, it may pose a greater challenge in the future.

4.6 CONCLUSION

While in our setting the biggest gain in accurately identifying recent cases was derived from including ID-NAT testing, the biggest improvement in the incidence estimates was from the additional inclusion of VL with very limited gain by adding ARV testing. However, these results should be considered with caution as they are specific to our setting of high background HIV prevalence, incidence[23] and ARV uptake[28]. Further research is required to identify and clarify other factors that may influence HIV-positive individuals' and populations' progression from 'recent' to 'longstanding' infection.

Our work confirmed the need for both accurate identification of recently acquired HIV cases as well as the use of context-specific MDRI and FRR rates to derive meaningfully useful incidence estimates. While it appears theoretically feasible to derive MDRI and FRR estimates that can account for the inherent variability in incidence modelling, we have shown that it is not yet possible to achieve this with sufficient accuracy to fully account for such variability.

4.7 ACKNOWLEDGEMENTS

K.B. performed the research, data collection, initial data analysis and wrote the first draft of the manuscript; All authors contributed to the study design; E.G. performed extended data analysis; V.L., and E.M. supervised the research; all authors reviewed and approved the final manuscript.

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CHAPTER 5: DISCUSSION

In the "Action Framework to Advance Universal Access to Safe, Effective and Quality Assured Blood Products 2020-2023", the WHO reaffirmed the global need for a safe, sustainable blood supply for the effective delivery of health services and programs.[1] A key safety component is the reduction of the risk of TTI such as HIV, HBV and HCV.[2] To achieve this, blood transfusion services employ a multi-pronged approach. While laboratory screening is conducted on samples of all donated blood, donors must first pass through a combination of pre-donation education, donor assessment via a DHQ and potential deferral. Deferral is disqualification from donation, whether indefinitely (as for people living with certain infections, including HIV) or for a specified period (as for low iron).[3] The combination of donor assessment and laboratory testing has been remarkably successful[4], decreasing the number of transfusion-related HIV transmissions from thousands in the early 1980s[5] in the USA to an estimated risk of less than 1 in 1.6 million donations in 2021.[6] Even in South Africa, which has a generalized HIV epidemic, improved donor screening and testing strategies reduced the risk of HIV transmission from an estimated 22 per million transfusions in 1994[7] to less than 13 per million transfusions in 2015 despite an exponentially increasing epidemic.[8]

This success notwithstanding, the effectiveness of pre-donation donor assessment and deferral, as a blood safety strategy, depends on both the blood transfusion service asking the right questions (i.e., those that address behaviours and health conditions that truly pose risk) on the DHQ, and the willingness of would-be donors to disclose personal information in response to such questions.[9-11] This can pose challenges. For example, regarding TTI such as HIV and HCV, studies in Europe,[9,12,13] North America[14], Australia[15] and India[16] confirmed higher rates of nondisclosure of risk factors among donors who tested positive for TTI markers than among donors who tested negative. Most literature on non-disclosure among blood donors as it relates to HIV and other TTI revolves around *risk behaviours*. At the time of conceptualising this PhD, the non-disclosure of *known* HIV status or ARV use among blood donors had been much less frequently explored. In the intervening years, the phenomenon of undisclosed HIV+/ARV+ as well as undisclosed PrEP use have been described in multiple settings. [17-19] In addition, the potential deleterious effect of undisclosed ARV/PrEP use on routine blood donation testing, both molecular and serologic tests, have been demonstrated by researchers in Brazil.[20]

5.1 THE EPIDEMIOLOGY OF UNDISCLOSED HIV STATUS AND ARV USE

My interest in the impact of HIV on blood transfusion services started shortly after joining SANBS and coming to realise the extent to which the epidemic influences both blood collection and blood utilisation in South Africa. This interest let me to join the NHLBI funded REDS-III project and also informed the topic of my Master's dissertation "Transfusion Practices in the Eastern Cape Province of South Africa in the era of HIV and HAART". It was during the execution of the various REDS-III projects that we became aware that the increasing "prevalence" of what was at the time assumed to be HIV EC may be in fact be due to undisclosed ARV use.

During attempted enrolment in an EC follow-up study, some presumed EC, disclosed prior knowledge of HIV infection and ART use at the time of their index donation. This led us to send stored plasma samples of 152 potential EC, previously collected over a 5-year period, for ART testing. Disturbingly, 103 (68%) of the specimens in this limited sample tested positive for recent ART use at the time of donation.[21] This raised the broader research question regarding the prevalence among blood donors of persons previously diagnosed with HIV and taking ART.

To the best of my knowledge, this project was the first in Africa to systematically examine the prevalence of HIV-positive donors already on ARV donating blood and only the second globally.[17] I confirmed that a substantial proportion of HIV positive, prospective South African blood donors had undisclosed ARV use. The overall prevalence of undisclosed ARV use was 9.8%. Although the majority of HIV+/ARV+ donors were female and Black, these differences were not significant and were not associated with increased odds of undisclosed HIV status and ARV use. This likely is a simple reflection of the South African reality in which the HIV epidemic affects females and Black Africans disproportionally. Undisclosed ARV use was very high (85%) among donors who tested HIV RNA negative but antibody positive, a phenotype likely associated with their ARV intake and previously described as "false EC".[21] Factors independently associated with undisclosed ARV use were increasing age, first-time donor status, donation in provinces in South Africa with the highest HIV burden.[22]

While this project was one of the first to study undisclosed ARV use in blood donors, the phenomenon had been described in other settings, including HIV vaccine trials, household surveys, and high-risk populations. These studies have disparate results in relation to age, education and wealth levels, but consistently found no association with gender, which is similar to the findings of this work.[21,23-28] Household and community surveys generally reported younger age to be associated with greater odds of undisclosed ARV use, but had disparate results in terms of wealth and education.[23,26,27,29,30] Neither the HPTN 052[24] nor HPTN 075[31] HIV Prevention Trials Network trials demonstrated associations with age, but did show local as well as between-country regional differences. By contrast, undisclosed ARV use and HIV status were associated with older age among American MSM, which is more aligned with the results of this study.[25] These disparate results may suggest differing behavioural motivations for undisclosed ARV use among different populations.

In addition to the background population-level HIV prevalence, other factors that may potentially contribute to the observed demographic differences may relate to issues of access to care, ongoing high levels of HIV-related stigma and a low perceived risk personal risk of transmitting HIV through blood donation. For example, the Fifth South African National HIV Prevalence, Incidence, Behaviour and Communication Survey[22], conducted in 2017, noted that while they had the highest HIV prevalence in the country, the Free State, Eastern Cape and KwaZulu Natal provinces also had the highest proportions of VL suppression with lower rates of suppression

seen in the northern rural provinces. The higher odds of undisclosed ARV use seen among donors from these provinces may be associated with the higher uptake of ARV in these provinces. This also aligns with the findings of Sykes *et al.* who demonstrated the high proportion of undisclosed ARV use among RNA-/Ab+ donors.[21] The proportion of undisclosed ARV use among RNA-/Ab+ donors in our study period (2017) was 85% compared to the 76% reported by Sykes *et al.* for 2016 which correlates with increasing ARV coverage in South Africa.[32,33] Over this study period, Sykes *et al*. found a temporal increase in the proportion of HIV-positive blood donors who were RNA-/Ab+ and within this group, undisclosed ARV use increased from 39% in 2010 to 76% in 2016. Similarly, high rates of undisclosed ARV use have been reported among presumed ARV naïve HIV-positive persons who had suppressed viral loads, particularly in settings involving HIV treatment studies.[24]

In addition, a US study investigating the factors associated with patient nondisclosure of medically relevant information found that concerns about being judged, lectured or embarrassed to be independently associated with nondisclosure.[34] In relation to blood donors, feelings of being judged or embarrassed or having their HIV status disclosed to those around them, may similarly be reasons for not disclosing their HIV status and ARV use. The higher odds of undisclosed ARV use at mobile blood drives may be indicative of a relative lack of privacy at these settings which potentially creates a barrier to disclosed HIV status and ARV use.

Furthermore, a 2019 publication by Wentz *et al.*[35] concluded that young adult MSM in the USA donated frequently despite MSM being cause for permanent deferral from blood donation at the time. They suggested that these donors may be presenting to donate due to the belief that their blood is safe because of their low perceived personal risk of transmitting HIV through blood donation. Such "personal risk assessment" behaviour was also seen among donors who participated in a cognitive evaluation of the AABB uniform donor history questionnaire.[11] In this study, the authors demonstrated that rather than assess each question on the questionnaire strictly as asked, donors framed their interpretations such that each question was understood to ask, "Is my blood safe to donate?" which then informed their responses. This resulted in donors "filtering" their response based more on their own personal perceived risk rather than on the exact factual response for each question. Within the setting of HIV+/ARV+ blood donation, there may well be similar personal risk assessments, especially given the "U=U" campaigns and the significant media coverage, at the time, of HIV-positive to HIV-positive kidney transplants.

While this study confirmed similar prevalence of undisclosed ARV use among HIV-positive donors classified as LAg recent and those classified as LAg longstanding, it is likely that these data may contain "false" recent cases attributable to prolonged ARV use. Studies have shown that prolonged ARV use affects most tests, including the LAg avidity test, aimed at determining recency of HIV infection, resulting in overestimation of recent infections.[36-38] The FRR of these tests range from 50% to 76% among persons on ARV, which more likely

explains the high proportion of undisclosed ARV use among donors classified as having "recent infection" rather than a significant number of donors having commenced ARV therapy within a couple of months of HIV acquisition.[36] These concerns are more related to accurate estimations of recent infections and incidence modelling. However the significant ramp-up of ART delivery through the "test-and-treat" programmes PrEP roll-out (and the equal likelihood of disclosure failure by blood donors) raises additional safety concerns about the ability of current testing methods used by blood services to detect cases of seroreversion or delayed seroconversion. I further explored the impact of such undisclosed ARV use on the accurate identification of cases of recent HIV acquisition as well as on HIV incidence modelling, please see Section 5.3 "*The Effect of Undisclosed HIV Status and ARV Use on Recency Testing and Incidence Modelling*" below.

Both failed PrEP and early ART initiation are associated with delayed seroconversion and in some instances with seroreversion.[39,40] Rapid initiation of ART in persons with recently acquired HIV could therefore result in failure to identify donors with suppressed VL and delayed or reversed seroconversion with current blood banking testing algorithms.[41] Vermeulen *et al*. reported a 2% probability of transfusion transmitted HIV infection from a red blood cell product donated by donors who are RNA-/Ab+. This finding would suggest that blood products donated by HIV-positive donors who seroreverted or who failed to seroconvert due to very early ART initiation, would likely have a similar probability of transmitting HIV to recipients. While the absolute risk of transmission of HIV through a blood transfusion remains low compared to the overall incidence of HIV in the country, the effect on the public's trust may be significant. The impact of the international outcry of what was considered the failure of blood services to protect blood transfusion recipients from HIV and HCV infections during the 1980's[42], still affects existing and potential blood donor perception of the risks involved in donating blood, with >5% of both groups citing risk of "contamination" as a deterrent to blood donation.[43] Furthermore, the perceived incidence of TTI was 13.5% among patients surveyed in an academic hospital in Germany with 38% of those surveyed having a significant risk perception about possible TTI.[44]

The 9.8% prevalence of undisclosed ARV use among South African blood donors is alarming and has significant implications for blood transfusion safety in the country. The higher prevalence of non-disclosure among first-time donors was not unexpected since they had not been previously exposed to the blood service's behavioural screening. However, the non-disclosure among repeat and lapsed donors is more alarming because it indicates a failure of repeated exposures to the SANBS donor self-assessment questionnaire and one-on-one interview procedure to identify HIV-positive donors on ARV. A better understanding of the factors and motivations that contributed to this behaviour and decision making on the part of the HIV+/ARV+ donors were clearly required.

This study had limitations. First, no samples were available for ARV testing from 14.5% of the HIV-positive donations. While some plasma bags were destroyed due to leakage and other operational factors, the majority

of these was discarded following failure of a freezer at one of the two SANBS testing sites, which may affect some of the geographic sub-analysis as most of the discarded units were from two geographical areas. Second, due to the short half-life of some ARV drugs our testing algorithm may have missed donors on ARV who had interrupted therapy in the days prior to donation, which may result in underestimation of undisclosed ARV use. We only tested for four of the most commonly prescribed ARV in South Africa (nevirapine, efavirenz, lopinavir and zidovudine). However, at the time, efavirenz was standard in the three-drug fixed dose combination therapy prescribed as first-line therapy for the majority of adults diagnosed with HIV in South Africa and zidovudine was a key component of second-line therapy.[45] Third, we also did not test for tenofovir, which may have missed donors on pre-exposure prophylaxis (PrEP). At the time, though, PrEP was not yet widely available in South Africa and unlikely to have materially affected the overall results of this investigation.

We confirmed that almost 1 in 10 HIV positive blood donors in South Africa did not disclose their known HIV status and ARV use, likely associated with the major national ART rollout. The possibility of seroreversion and/or the lack of seroconversion due to early ART initiation, or delayed seroconversion resulting from PrEP use, signifies an increased risk to blood safety. This risk was illustrated by a case reported from Brazil where a donor in the acute phase of HIV acquisition at the time of donation, failed to disclose PrEP use.[20] The authors demonstrated the impact the PrEP use had on the kinetics of routine blood donor HIV testing, and the real risk of such donations escaping detection by routine testing. While the absolute number of ARV+/HIV+ cases (and presumably also the number of donors with unreported PrEP use) is currently still small, the impact of even one case of transfusion transmitted HIV by a blood donor who failed to disclose their HIV status and ARV use, may have a significant impact on the trust of both the donor and recipient communities. While we identified several factors associated with undisclosed ARV use among blood donors, the underlying cause and motivation remained unclear. The need to better understand the underlying motivation for this behaviour so as to devise appropriate mitigation strategies prompted the second part of this study, discussed further in section 5.2 below. Our research may help increase awareness of this phenomenon and help inform blood safety strategies globally, in particular in other high HIV prevalence countries with large-scale ARV availability.

In addition, it stands to reason that if PLWH who are on ARV fail to disclose their status and ARV use, those on PrEP will likely also not disclose, especially if they feel stigmatised in any way. Given that the large proportion of the South African population at risk of HIV acquisition, the push for large scale PrEP roll-out is a social imperative. However, local and international standards currently require the deferral from donation of any person who take any form of PrEP. In South African, donors are deferred for 3 months after cessation of oral ARV use which is in line with international practice, which also includes recommendations for a 2-year deferral for injectable PrEP (injectable PrEP is not currently available in South Africa). The large-scale roll out of PrEP therefore poses a two-fold risk to the blood supply, namely the potential impact on the availability of sufficient

blood but also on impaired kinetics of traditional blood donation testing algorithms, making this an important follow-up research focus area for blood services globally.

5.2 BEHAVIOURAL INSIGHTS INTO UNDISCLOSED HIV STATUS AND ARV USE

When I began formulating the research concepts for this PhD, my experience with qualitative research was very limited, and my grasp of its fundamentals was basic. However, under the guidance of Dr. Hughes, my comprehension of the intricate aspects of qualitative research deepened. Simultaneously, I acquired a new array of skills and knowledge essential for conducting mixed-methods research, thereby mastering this methodology. During the initial stages of ideation for this research, I engaged in extensive dialogues with a variety of colleagues to explore their perceptions regarding the motivations and behaviours that might lead certain donors to voluntarily disclose their HIV status and ARV medication use, in contrast to those who choose not to. As the qualitative dimension of this study progressed, I was confronted with the realization that many of our initial hypotheses were markedly inaccurate. The insights gained through the execution of the qualitative research have been enlightening, significantly influencing my comprehension of donor motivations and behaviours within this specific framework. Furthermore, these experiences have informed my appreciation for the necessity of fostering open-minded interactions with both current and prospective donors as a strategy to circumvent fallacious assumptions regarding their motivations and behaviours."

The mixed-methods study design utilised uniquely enabled the documentation of the motivations and contexts associated with blood donation by PLWH currently using ARV. The SEM provided the conceptual framework for this study. The SEM is frequently used in health research[46-48] and posits a complex interplay between multiple levels of influences, and human behaviour and health outcomes. A version of the SEM adapted specifically to deal with HIV risk guided development of data collection instruments capable of parsing the complex dynamics around blood donation by HIV+/ARV+ individuals (**Figure 2**).[49] Specifically, survey items and interview questions addressed influences at the individual, social, cultural, and policy levels. Examples include: the belief that SANBS screening can identify all HIV+ donations so donating will not cause harm; belief in the possibility of a cure for HIV, with the blood center being seen as a venue for confirmatory testing (individual); economic need making it difficult to resist when incentives are provided by the blood center; peer pressure to donate (social); pervasiveness and severity of HIV-related stigma inhibiting disclosure of HIV status (community).

By combining ACASI technology, and an individual, in-depth qualitative interview we confirmed that approximately two thirds of the participants had disclosed their HIV status to no more than three people. Eleven participants were identified as experiencing a high level of perceived stigma. Two (1101M and 2202M)

interviewees refused to acknowledge having knowingly donated while HIV+/ARV+. Reviewing interviewees' reported motivations for their SQDs, we grouped responses into three themes: 1) altruism, expressed both as a general wish to "save lives", and the specific intention of donating so that blood could be given to other PLWH; 2) a lack of privacy at the donation location, associated with a fear of status disclosure; and 3) other reasons. As the first two themes heavily predominated among interviewees' responses, the "other reasons" category of donation motivations is not be discussed in detail but is summarised in **Supplementary Table 2 (Appendix 8)**.

Notably, very few accounts suggested any kind of test-seeking and none provided evidence of an interviewee believing that s/he had been cured of HIV, both of these having been cited by multiple colleagues as what they believed would be potential motivations at the time of designing this project.

Also, regarding the data set as a whole, after stratifying by stigma scale score, we found no meaningful difference between the overall accounts of interviewees categorized as perceiving low and high stigma. For example, the following topics were mentioned by interviewees in both categories: not wanting to reveal their status to other people who were present, "knowing" that their blood would be discarded so donation wouldn't cause harm, donating as a blood management practice, and wanting to donate for the benefit of others—either generally or specifically for PLWH. Similarly, interviewees in both categories referenced highly restricted serostatus disclosure practices (not merely in the donation context), a lack of sufficient privacy at the donation location to complete the DHQ accurately or have a status-related discussion with SANBS staff, and the idea that asking specifically about HIV status on the DHQ is simply too sensitive. That being noted, interviewees who reported wishing to donate blood for use by other PLWH were more likely to be classified as perceiving low stigma, while those who reported privacy concerns being the predominant factor in their donation were more likely to be classified as perceiving high levels of stigma.

The following sections outline the study's main thematic findings pertaining to donation motivation (altruism and privacy) and context (understanding of donor eligibility and pre-donation screening experiences).

Motivation: Altruism

Overall, altruism was the most frequently reported motivation for donating blood among this sample, mentioned by over half of the interviewees. We identified two distinct sub-themes within these accounts. The first was a general wish to help others through donation. Some interviewees talked about friends or family members having previously needed or received a transfusion; others reported awareness of the general need for blood. For instance, as one man explained, "I wanted to donate blood because I knew my blood type was the most wanted one" and that traffic accidents had caused "a need for blood". A woman who was a repeat

donor shared her wish to donate "because my mother was sick and they donated blood for her. So, I thought if I could donate maybe I could help someone else just like they helped my mother". A first-time female donor told of coming across a mobile donation site at the mall. She reported thinking to herself about how it had always been easy to give blood samples at her medical appointments and concluded, "I have got a lot of blood, why don't I give to someone who needs blood?". Knowing that "during Easter there [are] lots of accidents, and you find that they are saying the blood bank is short of blood" seemed to strengthen her resolve.

The wish to engage in a more specific form of altruism was expressed by the majority of the altruism motivated interviewees. They were motivated to donate blood so that it could be given to a recipient also living with HIV. Some also asked the interviewer explicitly if a PLWH might donate for other PLWH, although that was not the primary motivation for their study qualifying donation.

Interviewees' talk about donating for other PLWH often drew on notions of "matching," seeming to equate matching serostatus with the matching of blood types required for transfusion. One woman, whose primary motivation for donating was a more general altruistic impulse, asked the interviewer hopefully: "If I donated blood for positive people…my blood group is B. So, if the other person…is positive and I'm positive too, and [the other person] needs blood group B, will I be able to help?". Some interviewees also spoke of themselves as being particularly good donors for other PLWH. This suitability was variously attributed to their overall health, serological indicators, and/or medication adherence. One interviewee, who started using ARVs in 2012, said of her blood, "I think it is better than the people who have just found out that they are HIV positive…. because I am drinking the medications regularly and then my health is fine. It will help the people who are HIV positive especially the ones with low CD4 count or whatever".

The level of certainty interviewees expressed about whether donation by a PLWH to another PLWH was feasible varied, though most were remarkably confident. Those who were least sure thought that "there *could* be something that can be done with HIV blood", and that "maybe [SANBS] divide[s] the blood" from "people who live with the disease". Others framed donating (for other PLWH) as a duty; the idea that PLWH would be ineligible to donate rarely surfaced in their stories: "If someone who is HIV-positive needs blood, I have to donate, because they need blood. I have a lot of blood, I have to [donate] so that I can help". For one interviewee, the idea that he could help other PLWH was extremely persistent; he continued to believe (and act on) it even after being explicitly told he was not eligible to donate. He explained his reasoning: "Blood is blood, whether infected or not. I still believe it can help other people in need".

Motivation: Privacy

After altruism, the most common factor raised by interviewees as playing a meaningful role in their decision to donate was a lack of privacy. This was described by over a third of interviewees, across all provinces, and both stigma categories. In most cases, interviewees reported that when the opportunity to donate blood arose, frequently at blood drives held at schools or workplaces, they were with other people (co-workers, classmates, friends, romantic partners) who were unaware of their HIV status. These participants believed that opting out of donation would unavoidably lead to speculation about their reasons for doing so. Some participants visited the donation site intending to inquire about the SANBS eligibility criteria for PLWH, but discovered the lack of a private and confidential space for this conversation. Similarly, interviewees were hesitant to disclose their HIV status on the Donor Health Questionnaire (DHQ) due to the proximity of co-workers and friends or because of doubts regarding the confidentiality of responses to screening questions.

The account offered by on participant illustrates many of these trends. She explained that SANBS ran a blood drive at her workplace and "everyone, most of the people in the office, they were going to donate…and yah, so I didn't have much of an excuse as to why I shouldn't go." She thought that "when I get there, I will [be] able to speak, maybe it will be in private ...but it was in the boardroom and you know they had the beds and stuff so we all just filled in the forms in one table and then you moved to the [phlebotomy]." Regarding the screening questions, she "didn't answer them truthfully because of the….situation, because everyone was looking at each other … it was like a joke because even those who had iron problems and [were] turned away, and then it was like people were like talking like, 'Oh why have [they] been turned away?'" This interviewee had thus observed that being deferred from donation, even for a reason like iron deficiency, made one the recipient of precisely the kind of attention (and, potentially, gossip) that she wanted to avoid. The fear of inadvertent disclosure of an interviewee's HIV status surfaced in multiple ways and was a thread in many of these narratives.

Only one person—a man—shared a notably different story of encountering a donation opportunity while amongst acquaintances. He had previously donated at blood drives held at his place of employment so when he stopped doing so, he says this prompted his colleagues to ask him, "'Look, you once donated, and you never go again' and I say, 'Argh, don't worry,' you know? I don't even explain because they know it's everyone's choice, you see'. Thus, he explicitly disavowed experiencing these circumstances as pressure (he framed his motivation for donating blood as helping other PLWH).

Slotting these research findings into an adapted version of the SEM that grounded our study, demonstrates the complex nature of the factors influencing the decision making of HIV+/ARV+ donors when donating blood. Despite the similar way altruism and privacy were described by interviewees (i.e., as "the reason" for their donation), they are different. This difference led us to locate these factors in different levels of the SEM.

Altruistic motivations were classified as an individual-level influence on donation as they were constructed as an expression of personal moral standards (recognizing, however, that all levels of the model are interconnected; for example, individual assessments take place within a cultural context that frames altruism as morally good). In contrast, privacy motivations were classified as a cultural factor. This was because our data show the desire for privacy stemmed from a general fear of disclosure of one's HIV status; therefore, it is more meaningfully understood as reflecting the still-pervasive HIV-related stigma in their communities rather than simply individually varying comfort levels.

Additional factors influencing HIV+/ARV+ donor decision-making and behaviour included donating in the presence of people to whom they had not and did not intend to disclose their serostatus (social level factor) as well as the physical environment and infrastructure at donation locations, especially mobile drives (physical environment factor). At an organisational policy level, blood services, including SANBS, have spent significant time and resources educating potential donors on the risk of "window period" donations and that the blood service should not be used as an HIV testing centre. To date, however, SANBS has not disseminated messaging that PLWH are permanently deferred from donation. In addition, while SANBS has recommendations on ensuring a private setting for the one-on-one screening of donors, the reliance on existing infrastructure at mobile blood drives, makes ensuring fully private conversations near impossible. That said, it would be of significant benefit, if alternative methods can be developed to allow donors to confidentially obtain information on donation eligibility or to withdraw their donation.

The nuances of the decision-making in interviewees' narratives cannot be adequately grasped, or responded to, without a multi-level model. In particular, the SEM helps highlight the fact that addressing factors related to HIV+/ARV+ donation at one level, such as individual understanding of the inability of the blood services to use blood donated by PLWH, may not eliminate the behaviour, as influences at other levels (HIV-related stigma and resulting desire for greater privacy) will still exist if measures have not been taken to mitigate them.

To the best of my knowledge, this is the first qualitative research to explore the motivations of HIV+/ARV+ donors and it uncovered complex, diverse motivations related to altruism and privacy leading to HIV+/ARV+ blood donations and noted multiple factors across different levels of the SEM influencing donor motivation and behaviour. As such, to be successful, interventions aimed at limiting donations by HIV+/ARV+ must therefore address all these factors and do so without increasing HIV-related stigma.

At a personal level, this research taught me two important lessons. The first was the need to think of PLWH as people within our community first and foremost. In this research, adapting the SEM allowed me to focus on

HIV risk; such tailoring is generally seen as an appropriate way to attend to context. Yet, this approach inadvertently led to the conceptualization of HIV+/ARV+ donors as distinct entities, essentially 'othering' them. A significant oversight of the adapted SEM employed in this investigation was its failure to illuminate the commonalities between HIV+/ARV+ donors and their counterparts. The second lesson was a reminder to temper my own and others' presumptuousness when evaluating the real-world experiences of individuals. At the project's inception, there was a widespread belief among my peers that the primary motivations for HIV+/ARV+ individuals donating blood were either a mistaken belief in their cure or a desire for testing. These assumptions were refuted, as not a single participant donated under the belief of being cured, and only a minority mentioned testing as a secondary motive.

5.3 THE EFFECT OF UNDISCLOSED HIV STATUS AND ARV USE ON RECENCY TESTING AND INCIDENCE MODELING

Throughout my 15-year tenure at the SANBS, addressing the risk of transfusion-transmitted (TT) HIV has been a central aspect of my work and a broader priority for the SANBS organization. Indeed, even four decades after the global recognition of HIV, preventing TT HIV infections continues to be a crucial objective for blood services worldwide. Since becoming Medical Director for SANBS, I see the risk of HIV transmission not only in relation to the broader safety of the blood supply, but also as it directly impacts my organisation's ability to meet the country's blood demand. Strategies to ensure a sufficient blood supply must simultaneously encompass strategies to mitigate the risk of transfusion-transmitted HIV. However, systems for tracking the outcomes of such mitigation strategies are complex and generally dependent on sophisticated (and costly) testing algorithms and statistical analysis.[50]

While early in the epidemic, the success of such mitigation strategies were generally evaluated by monitoring the prevalence of HIV-positive donations, the increasingly sensitive HIV testing assays and new methods for assessing HIV transmission risk required a different approach to monitoring the outcomes of such strategies.[51] Most rely upon estimation of HIV incidence, either through prospective studies or, more recently, modeling based upon cross-sectional data to impute incidence.[50] This was made possible through the development of recency assays and later recent infection testing algorithms. For a recency assay to be useful in modelling incidence estimates, it must have a sufficiently long MDRI and a low FRR.[52] The calculations of MDRI and FRR are per se complex, with the MDRI, for example, being significantly impacted by the testing used to identify HIV cases and the FRR by ARV used.

As a result, imputing incidence from cross-sectional data requires sophisticated statistical knowledge to calibrate context-specific MDRI and FRR requires and also detailed, recent HIV epidemiologic information, limiting its utility especially in resource constrained settings. However, the degree to which uncertainty for

each of these factors impacts incidence estimations, especially in the blood donation setting, is unknown. Gaining a clearer understanding of this uncertainty could potentially streamline the requirements necessary for this type of incidence estimation. This in turn may be of significant value in as resource constrained setting. Considering SANBS's ongoing international collaborations, its substantial data and sample repositories and its sophisticated donation testing infrastructure, this project presented an ideal opportunity to contribute to resolving some of these questions. It stands as the culminating step in comprehensively understanding how undisclosed HIV status and ARV use impact field of blood transfusion medicine.

While the most recent recommendations[53] on incidence modeling include the use of context-specific calibrated MDRI and FRR and multi-assay RITA aimed at limiting the impact of viral suppression, either due to elite controller status or ARV use, on FRR, the relative utility of each aspect remained unclear. The addition of a VL cut-off to differentiate between "true" and "false" recent cases has been the backbone of newer RITA.[54] The continued expansion of ARV therapy, coupled with the not uncommon instances of nondisclosure of ARV use, has prompted inquiries into the necessity of incorporating ARV testing within the RITA. [24,26,55] This is to guarantee the exclusion of individuals on ARV therapy who do not have complete viral suppression. Furthermore, it raises broader questions about what the ideal RITA would entail in specific contexts, such as a blood donation environment, which is the focus of this study.[56]

Using the data derived from quantifying recent cases through the four, increasingly comprehensive RITA, I demonstrated significant misclassification of the recency status of HIV-positive donors when using a RITA that does not include ID-NAT, VL and ARV testing. The omission of ID-NAT posed a significant challenge. This gap in the algorithm not only led to the failure in detecting HIV-infected donors who had not yet undergone seroconversion (thereby missing these individuals in any RITA that excludes NAT testing) but also erroneously categorized donors with a low viral load (<75 copies/mL), which was detectable by NAT, as recent infections when they were not. The addition of VL testing provided marginal improvement in recency misclassification as it was able to identify the small number of antibody positive donors with a low, but detectable VL. The inclusion of ARV testing had a near negligible improvement on the identification of recent cases.

In addition, I confirmed a trend of decreasing incidence estimates with each increasingly comprehensive RITA. However, the use of context-specific MDRI and FRR resulted in only marginally lower incidence estimates (none of which were statistically significant) compared to the published "Kasssanjee" MDRI and FRR estimates.[57] The context-specific MDRI differed from the "Kassanjee" estimates even though both were derived from South African blood donors among whom Clade C infections dominate. However, the Kassanjee estimate was derived from donors who donated prior to 2014 and for whom an HIV lysate-based Western blot assay was used to identify HIV-positive cases. In contrast, this analysis used a chemiluminescent immunoassay (Abbott Prism

HIV1/2®) either alone or in combination with ID-NAT to identify HIV-positive cases. In particular, the use of ID-NAT resulted in an extended MDRI while simultaneously identifying seronegative recent cases. The combined outcome of these methodologies resulted in findings that were remarkably, yet coincidentally, similar to those achieved with the Kassanjee MDRI and FRR.

Drawing from the discussion above, and considering our specific context—a blood transfusion service in a country with an approximate HIV prevalence of 13.9% and a national estimated HIV incidence of 0.48% in 2017[59], ID-NAT testing emerged as the most significant factor in correctly reclassifying misidentified cases. It successfully identified individuals who had not yet seroconverted (and were therefore presumed to be recent infections) as well as seropositive donations with an extremely low VL (below approximately 18 copies/mL)[60] (and thus presumed to be long-standing infections), a third of which tested recent in Algorithm 1. However, in settings with significantly lower prevalence and especially lower incidence, the impact of ID-NAT versus simple VL testing may be less obvious as, once identified as HIV-positive, VL testing would identify the overwhelming majority of HIV-positive donors with low VL, irrespective of the reason for the low VL.

Conversely, the use of ID-NAT only to identify recent cases is also problematic. In their publication on the HIV incidence among South African blood donors, Vermeulen *et al.* [61] essentially applied Algorithm 2 (ID-NAT and LAg) to identify recently infected HIV cases in their "LAg first-time donor" model and relied solely on ID-NAT to identify recent cases in the "NAT Yield Window Period" and "Classic Incidence/Window Period" model. Given the outcome of this study, it is likely that the "LAG first-time donor" model would have over-estimated incidence as it is likely that it did not exclude LAg recent donors who had low, but detectable VL and those on ARV with poor viremic control. In contrast, the two models that rely solely on ID-NAT to identify recent infections, likely underestimated incidence as ID-NAT positive, seronegative cases accounted for only 23 of the 96 recent cases in our study.

Notably, the only benefit of adding ARV testing (in addition to ID-NAT and VL) was the identification of those donors on treatment that were not virally suppressed (to a level below 75 copies/mL). Recognizing that NAT testing potentially introduces an overly low VL cut-off, the Transfusion Transmissible Infections Monitoring System program in the USA considered a RITA that included VL (but not ARV assessment) for defining incident HIV cases.[62] Their assessment, based on a cut-off of 1000 copies/mL, was that such cases would be unlikely in their donor population and as a result, did not include it in their final analysis. However, a contemporaneous publication by Custer *et al.*[17] confirmed that approximately 15% of HIV-positive donors from their regions in the USA were on ARV at the time of donation. Given the overlap between these two studies, there may have been an underappreciation of the impact of low VL and ARV use on identifying recent HIV cases. These findings

would suggest that blood services should carefully consider RITA that sufficiently account for potential undisclosed ARV use.

Accurately identifying recently acquired HIV cases is the first (and crucial) step in cross-sectional based incidence modeling, but using correct, context-specific MDRI and FRR are equally important. However, the extent to which these factors and the precision with which each are estimated, impacts final incidence estimations were unknown.[52] From this work, I was able to demonstrate a decreasing, more precise incidence trend when applying increasingly comprehensive RITA in combination with context-specific MDRI and FRR. A similar decreasing incidence trend was seen when using the RITA outcomes with the published "Kassanjee" MDRI and FRR, but in all instances these incidence estimates were higher than those calculated with the context-specific MDRI and FRR. Although the difference in incidence estimates did not achieve statistical significance, this likely resulted from an insufficiently powered sample size. Nonetheless, these differences could hold epidemiologic significance. In a country like South Africa with a high HIV prevalence, the 0.05% per annum difference in estimated incidence between the "Kassanjee" Algorithm 1 and the "Calculated" Algorithm 4 might carry programmatic significance.

The outcome of these incidence calculations confirmed that accurate identification of recent infections will impact incidence calculations. However, it also highlighted those previous assertions that introducing a standardised time to "cut-off T" and the use of sophisticated, context-specific derived FRR and MDRI should offer "the opportunity to consistently account for imperfect accuracy and precision of the incidence estimator"[50,63] did not quite materialize. This is demonstrated by the different incidence estimated delivered for each of the RITA despite best-attempts at estimating context-specific MDRI and FRR for each. The fact that it did not, confirms that we still do not fully understand the impact of each test on incidence calculation and that there may be other factors affecting recency not yet fully elucidated.

This study had limitations. We had a full set of results for only 40% of the 969 lapsed and first-time donors who tested HIV positive during 2017. In particular, the sample included all the RNA-positive, seronegative and a disproportionate number (65 of 75 (86.7%)) of the RNA-negative, seropositive donors. This may have impacted the incidence calculations, especially Algorithm 1 which incorrectly classified nearly 40% of RNA-negative, seropositive cases as recent. In addition, the inclusion of this group of donors resulted in a higher ARV use estimation for the overall sample as this group is known to have high levels of undisclosed ARV use.[21,64] However, this would not have had a material impact on the algorithms other than Algorithm 1 as the remainder of the algorithms correctly classified these cases as longstanding. Furthermore, we assumed that all persons with VL below 75 copies/mL and all those on ARV had longstanding infections. It is conceivable that some true elite controllers may have attained viremic control within the "cut-off time T" and were therefore misclassified.

Likewise, donors could have contracted HIV, been diagnosed, and started treatment within this same timeframe. Although this was a minor concern in 2017 when South Africa began to adopt the "test and treat" approach, it may pose a greater challenge in the future. Furthermore, there is a lack of comprehensive calibration data for calculating the FRR in treated but unsuppressed HIV cases. In this study, the estimated FRR for these cases were based on the FRR in untreated persons. This is likely a conservative estimation of the FRR but it had a minimal contribution to the overall weighted FRR. I consider these assumptions to sufficiently mitigate this limitation

The approach I used to in this methodological analysis allowed the evaluation of adding RNA screening (ID-NAT), VL and ARV testing to HIV incidence modelling and surveillance, with particular focus on the blood donor setting. In this context, the biggest gain in accurately identifying recent HIV cases was derived from incorporating ID-NAT testing. Meanwhile, the biggest improvement in the incidence estimates resulted from the further inclusion of VL. The addition of ARV testing provided marginal gains both in identifying true recent cases and in incidence estimation. Further research is required to identify and clarify other factors that may influence HIV-positive individuals' and populations' progression from 'recent' to 'longstanding' infection.

This work confirmed the need for both accurate identification of recently acquired HIV cases as well as the use of context-specific MDRI and FRR rates to derive meaningfully useful incidence estimates. While it appears theoretically feasible to derive MDRI and FRR estimates that can account for the inherent variability in incidence modelling, we have shown that it is not yet possible to achieve this with sufficient accuracy to fully account for such variability.

5.4 FUTURE DIRECTIONS

While this body of work has answered some of the key questions surrounding PLWH already on treatment presenting to donate, there remains a number of gaps in our knowledge. These gaps can be divided into those related to the delivery of blood donation services, i.e. programmatic, and those of a basic sciences nature including corroboration of the findings of this research as well as further assessment in to the transmissibility of HIV+/ARV+ (and even PrEP breakthrough) infections.

From a programmatic perspective, the key focus areas for future research should be to understand what mitigation strategies reduce the number of HIV+/ARV+ donors presenting to donate. Potential mitigation strategies could include targeted donor education on the ineligibility of HIV+ and/or ARV+ donors to donate blood, improving systems to ensure confidential one-on-one assessments and even processes for donors to efficiently withdraw their donations should the above processes fail. Such research questions are best answered through a mixed-methods approach, where semi-structured interviews, either at an individual or

small group level, are used to identify potential solutions which is then followed by quantitative methods such as surveys to establish which of the potential solutions would have the greatest impact.

From a basic scientific perspective, a key unanswered question is the actual transmissibility of HIV+/ARV+ donations not detected by current testing algorithms. The numbers of such donations are likely to remain small, but a similar (and growing concern), is the transmissibility of PrEP breakthrough infections which may go undetected for some time and for whom the same concerns re disclosure are likely to exist. A basic science approach would be most useful to address these questions. Some proposed investigations include animal-models but also the use of techniques such a probit analysis to estimate extremely low viral loads as was done in Vermeulen *et al*.[65]

It is incumbent upon me as the Medical Director of SANBS to shape future research in the area. In addition, the significant HIV burden in South Africa with one of the largest ARV programs in the world, combined with the donor assessment and testing infrastructure, make SANBS the blood service most suited to driving this research. We have already initiated discussions with various research groups in South Africa and internationally to drive this research agenda.

5.5 CONCLUSION

The genesis of this thesis started approximately 8 years ago and has been quite a dramatic journey both for me personally, but also for blood services in South Africa and elsewhere. Blood donation by persons with prior knowledge of their HIV status and already on treatment was an unknown entity until ~2015 when it was first identified by the REDS-III group of which I was the lead in-country investigator. The work done as part of this project greatly fostered my development as a researcher and also as the Medical Director for the largest blood service in Sub-Saharan Africa. It raised my profile at an international level and provided recognition for the work done by SANBS and the collaboration with the University of Cape Town. Importantly, the research played a significant role in acknowledging HIV+/ARV+ blood donation as a global issue. Initially perceived as a "South African" problem, it quickly became evident that this is a global concern. Now that we better appreciate the prevalence of and understand the motivation for HIV+/ARV+ donation, we need to develop mitigation strategies (that do not further stigmatise PLWH) and improve our tools to monitor the outcome of such strategies.

For me, this body of work left me with the overall impression of people inherently being the same; with the same challenges, the same wants and needs and the same desire to give back to the communities within which they live and operate. Altruism is not the exclusive territory of HIV-negative people. However, HIV stigma, even in a country with a background prevalence of ~15%, still dramatically shapes PLWH's daily behaviour.

Regardless of all the powerful science we have available, failure to address the stigma of HIV will doom any attempts to address the potential risk HIV+/ARV+ donors bring to the safety of the country's blood supply, to failure.

With the ever-increasing HIV-positive population in South Africa and the continued roll-out of ARV and PrEP, blood services in South Africa must carefully consider the breadth of the intersection of HIV and blood donation. Collecting safe and sufficient blood for the country's needs in the presence of a shrinking donor pool as the HIV prevalence increases remains challenging. The work I performed as part of this PhD project will assist in informing future strategies aimed at minimising blood donation by HIV+/ARV+ donors. The skill and experience I gained through these endeavours have equipped me to tackle future research and projects that will assist in ensuring a safe and sufficient blood supply for the patients of South Africa.

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ANNEXURES

ANNEXURE 1: SOUTH AFRICAN NATIONAL BLOOD SERVICE DONOR QUESTIONNAIRE

South African National Blood Service

SANBS
South African National Blood Service
Registration No. 2000/026390/08

DONOR QUESTIONNAIRE

Thank you for donating today

Your donation could save at least three lives. Remarkable, isn't it? – As a Service we are committed to providing safe blood and blood products to those who need them. We would like to thank you on behalf of the many people whose lives you have changed for the better. We hope that you will continue to support this cause and continue making a difference. We assure you that all the information that you supply to the SANBS will be treated as confidential.

We request that you complete all required information as accurately and honestly as possible.

SECTION 1: CONTACT DETAILS	SECTION 3: LIFESTYLE QUESTIONNAIRE	SECTION 5: CONSENT
SECTION 2: HEALTH QUESTIONNAIRE	SECTION 4: DECLARATION	

SECTION 1: CONTACT DETAILS

FIRST TIME DONORS: Complete in full **REGULAR DONORS: Only complete relevant fields if information has changed.**

SURNAME:		TITLE:	
FIRST NAME:	OTHER INITIALS:	FEMALE	MALE
DATE OF BIRTH:	ID NUMBER: Y Y M M D D		
HOME ADDRESS:			
		POSTAL CODE:	
POSTAL ADDRESS:			
		POSTAL CODE:	
TELEPHONE: (H)	TELEPHONE: (W)		
E-MAIL ADDRESS: (Complete in BLOCK LETTERS below)	CELL PHONE:		
PLEASE TICK THE APPROPRIATE BLOCK			
HOW WOULD YOU LIKE US TO REMIND YOU OF YOUR NEXT DONATION?	E-MAIL ☐ SMS ☐	PHONE HOME ☐	PHONE CELL ☐
PREFERRED BLOOD DRIVE OR DONOR CENTRE TO DONATE:			
LANGUAGE: ENGLISH AFRIKAANS	ETHNIC GROUP: ASIAN BLACK	COLOURED	WHITE

IMPORTANT: DO NOT DONATE BLOOD IF YOU MAY HAVE BEEN EXPOSED TO HIV/AIDS OR HEPATITIS

You may be endangering someone's life.

DANGER: the window period . . .

The window period refers to the time from when a person is first infected with the Human Immunodeficiency Virus (HIV) or Hepatitis Virus until the person tests positively. During the window period, laboratory tests are negative, but the person is still capable of infecting others. The window period may last for months. Even though a window period donation may be stored and re-tested, the virus ***will still not be detected***. Help keep the blood supply as safe as possible by looking HONESTLY at your lifestyle and answering the questions truthfully. If you have been in a situation where you could have been exposed or infected by HIV/AIDS or Hepatitis, do not donate blood. By donating, you will be putting the lives of patients who receive your blood at risk.

- The blood service is required to check the lifestyle of all those who wish to donate. Answer the questions you will be asked as honestly as possible, to help us keep the blood supply safe.
- **Every blood donation is tested for HIV/AIDS and Hepatitis and it must be understood that a positive result may have a profound impact on you both psychologically as well as on your future lifestyle.**

If you are unsure about any of the above, please discuss this in confidence with our staff.

FOR MORE INFORMATION ON AIDS COUNSELLING AND TESTING, CALL TOLL FREE: 0800 01 2322

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SECTION 2: HEALTH QUESTIONNAIRE

All donors must complete this section. Your answers will be treated confidentially.

	Please read all questions carefully and answer honestly	Please **TICK** your answers		Staff Comments
1.	1.1 Are you feeling well today?	YES	NO	
	1.2 Have you had something to eat or drink in the last 4 hours?	YES	NO	
2.	**Are you involved in any of the following:**			
	2.1 Driving a public or heavy-duty vehicle, flying an aeroplane, working on scaffolding or using power tools?	YES	NO	
	2.2 Sky diving, deep-sea diving or mountaineering?	YES	NO	
3.	**In the past 7 days:** 3.1 Have you been to the dentist?	YES	NO	
	3.2 Have you taken any pain killers, anti-inflammatories or aspirin?	YES	NO	
4.	**In the past 7 days:** Have you had a cold, flu, sore throat, fever, infection or allergy problem?	YES	NO	
5.	**In the past 30 days:** 5.1 Have you had diarrhoea or vomiting?	YES	NO	
	5.2 Have you taken any drug on the medication list?	YES	NO	
	5.3 Have you or your sexual partner travelled to any country outside of South Africa?	YES	NO	
6.	**In the past 3 months:**	YES	NO	
	Have you taken any medication (including traditional medication, herbal) injections or tablets?	YES	NO	
7.	**In the past 6 months:**	YES	NO	
	7.1 Have you or your sexual partner had a blood transfusion, received blood products or clotting factors?	YES	NO	
	7.2 Have you had acupuncture kavady prayers or dry needling?	YES	NO	
8.	**In the past 6 months:** 8.1 Have you taken part in a drug, vaccine trial or any other clinical research?	YES	NO	
	8.2 Have you had a vaccination or immunization (inoculation)?	YES	NO	
9.	**In the past 6 months:** Have you had a surgical procedure or been admitted to hospital?	YES	NO	
10.	Are you scheduled to have surgery in the next **6 weeks?**	YES	NO	
11.	**Have you ever had:**			
	11.1 High blood pressure?	YES	NO	
	11.2 Heart, lung or circulatory problems?	YES	NO	
	11.3 Epilepsy, convulsions or strokes?	YES	NO	
	11.4 Cancer, skin cancer or leukaemia?	YES	NO	
	11.5 Diabetes, asthma, TB or kidney disease?	YES	NO	
	11.6 Haemochromatosis ("high iron"), polycythaemia ("too much blood") or a bleeding disorder?	YES	NO	
	11.7 Have you ever had a severe allergic reaction, any serious illnesses (including diseases where you travelled to hot and humid areas e.g. Chagas disease, yellow fever, etc.), or used medication not mentioned in above questions?	YES	NO	
12.	**HEPATITIS:**			
	12.1 Have you ever had yellow jaundice, hepatitis, liver disease or a positive test for hepatitis?	YES	NO	
	12.2 Have you been in contact or lived with anyone with hepatitis (jaundice) in the past **6 months?**	YES	NO	
13.	**MALARIA:**			
	13.1 Did you grow up in a malaria prevalent area outside of the borders of South Africa?	YES	NO	
	13.2 If yes, have you been in any malaria area in the last **3 years?**	YES	NO	
	13.3 Have you been in a malaria area in the last **3 months?**	YES	NO	
	13.4 Have you had malaria in the last **3 years?**	YES	NO	
14.	**VARIANT CREUTZFELDT-JACOB DISEASE (also known as Mad Cow Disease):**			
	14.1 Have you ever had neuro-surgery, received dura mater (brain covering) graft or taken pituitary growth hormone?	YES	NO	
	14.2 Have you or your sexual partner ever received a tissue, cornea or organ transplant?	YES	NO	
	14.3 Have you visited the United Kingdom on one or more occasions adding up to a total stay of **12 months or more** between the years 1980 and 1996?	YES	NO	
16.	Are you participating in a regular training or athletic programme?	YES	NO	
17.	Have you ever injected yourself or been injected with unprescribed steroids (body building drugs)?	YES	NO	
18.	**FOR WOMEN ONLY:**			
	18.1 **Are you pregnant or undergoing fertility treatment?**	YES	NO	
	18.2 **In the last 3 months:** Have you had a baby, miscarriage or abortion?	YES	NO	
	18.3 Are you breastfeeding?	YES	NO	

SECTION 3: LIFESTYLE QUESTIONNAIRE

All donors must complete this section. Your answers will be treated confidentially.

Please read all questions carefully and answer honestly — Please **TICK** your answers — Staff Comments

1.	**In the past 6 months:**			
	1.1 Have you had a tattoo, body or ear piercing, or permanent make-up applied?	YES	NO	
	1.2 Have you had Rastib, ritual scarring, ritual piercing, ritual circumcision, blood sharing or been stabbed?	YES	NO	
2.	**In the past 6 months:** Have you or your sexual partner had a needle stick or skin penetrating injury; or had skin, eye or mouth contact with another person's blood?	YES	NO	
3.	**In the past 6 months:** Have you taken anti-retroviral medication including Truvada (WPBTS added)?	YES	NO	

Please Note: **The following questions are of a sexual nature. This includes oral, vaginal and anal sex. We ask these questions as sexual contact may result in infectious diseases such as HIV entering the bloodstream and so be transmitted to patients through your blood being transfused to them.**

Please read all questions carefully and answer honestly — Please **TICK** your answers — Staff Comments

4.	4.1 Do you have AIDS or are you HIV positive?	YES	NO	
	4.2 Have you ever had sexual contact with anyone who has AIDS or is HIV positive?	YES	NO	
	4.3 Is your motivation for giving blood to test for HIV?	YES	NO	
5.	**In the past 6 months (with or without a condom):**			
	5.1 Have you started having sexual contact with a new sexual partner?	YES	NO	
	5.2 Have you had sexual contact with more than one person?	YES	NO	
	5.3 Do you think your sexual partner might have or had other sexual partners?	YES	NO	
	5.4 Have you had sexual contact with someone whose sexual history you do not know?	YES	NO	
	5.5 Have you had sexual contact with anyone who takes money, drugs or other favours for sex?	YES	NO	
	5.6 Have you received money, drugs or other payment for sex?	YES	NO	
	5.7 Have you been sexually assaulted?	YES	NO	
6.	**In the past 6 months:** Have you or your sexual partner had any sexually transmitted disease (STD) including genital herpes, syphilis or gonorrhoea (drop)?	YES	NO	
7.	Have you or your sexual partner ever used recreational, street drugs by nose, mouth or injection needle?	YES	NO	
8.	Do you consider your blood safe to be transfused to a patient?	YES	NO	

SECTION 4: DECLARATION

1.	I have read and understood the pamphlet "Your blood saves lives".
2.	I understand the donation process and the possible risks involved as explained to me.
3.	To the best of my knowledge all the information I supplied is the truth and I understand that if I have not answered these questions truthfully it could endanger the patient and lead to legal proceedings against me.
4.	I confirm that I am 16 years of age or older.
5.	I undertake that should I for any reason deem my blood not safe for use, I shall immediately inform SANBS.

SECTION 5: CONSENT

1.	I consent to my blood being tested for blood grouping, syphilis, hepatitis B, hepatitis C and HIV as well as such extended testing which may be necessary to ensure the safety of the recipient.		
2.	I consent to being informed of any test results that are important to my health or affect my ability to donate blood and to my test results and information being kept confidential and stored indefinitely by SANBS in a secure facility.		
3.	I consent to the administration of fluids, additives and the re-infusion of my own blood components as may be required during apheresis and red cell collection procedures or in the management of an untoward reaction.		
4.	I consent to samples and components of my blood as well as my donation data be used to improve blood safety, but understand that my identity will always be protected.		
5.	I consent to my donation data and blood specimen to be stored at the SANBS Biorepository for use in research approved by the SANBS Human Ethics Research Committee. I understand that such research results will not be traceable back to me.	YES	NO

Please do not sign until you have answered all the questions and read the declaration.

Name and surname:		Contact telephone number:					
Date of birth:		Today's date:					
Donor's signature:		SAP #					
Interviewed by *(signature)*:							
Accepted by *(signature)*:							

FOR OFFICE USE

(TO BE COMPLETED BY STAFF MEMBER)

ACCEPTABLE FOR:	P	R	C	U	(TICK)			PLATELET USE?	YES	NO				
EDUCATION GIVEN BY: SIGN								DONOR VERIFICATION IDENTITY:						
EDUCATION GIVEN BY: SAP #								PHLEBOTOMIST ACCEPTED			YES		NO	
MNEMONIC:								PHLEBOTOMIST SIGN:						
DONOR NUMBER:								PHLEBOTOMIST SAP #						
								TIME NEEDLE IN:			TIME NEEDLE OUT:			
DATE:	TRANSFER		UNK		NEW		VISITOR	DISCONTINUED BY: SIGN						
TIME:								DISCONTINUED BY: SAP #						
CuSO4	1. PASS / FAIL	HEMOCUE		SAP #				NEEDLE CUT OFF BY: SIGN						
	2. PASS / FAIL							NEEDLE CUT OFF BY: SAP #						
BP:		PULSE:						WEIGHT OF BAG: g						
WEIGHT:								VOLUME: ml			DURATION:			
PRODUCT:		PACK:												
BLOOD GROUP:		DONATION COUNT:												
MALARIA STICKER UNTIL		MARKER									BAR CODE			
DATE:														
RECRUITED YOUTH PROGRAMME		YES		NO										

DONOR REACTION

CATEGORY	MILD	MODERATE	SEVERE	CATEGORY	MILD	MODERATE	SEVERE
Haematoma				Faint immediate, accident			
Arterial puncture				Faint delayed type			
Delayed bleeding				Faint delayed, accident			
Nerve irritation				Citrate reaction			
Nerve injury				Haemolysis			
Tendon injury				Generalised allergic reaction			
Painful arm				Air embolism			
Faint immediate type				Other			
Thrombophlebitis				More than 1 venepuncture			

REMARKS / DONOR STATUS:			
	SECOND PHLEBOTOMY	BARCODE	
	TIME NEEDLE IN:	PHLEB. SIGN:	
	TIME NEEDLE OUT:	PHLEB. SAP #	
	DEFERRED	DEFERRAL CODE	
	REASON	RETURN DATE	

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ANNEXURE 2: STANDARD OPERATING PROCEDURE FOR HIV+/ARV+ BLOOD DONOR RECRUITMENT, ACASI SURVEY AND QUALITATIVE INTERVIEWS

Project Overview

Research Aim: To gain an in-depth understanding of the context, decision-making and experience of blood donation from SANBS donors whose blood tested positive for HIV and ARV (hereafter, "HIV+/ARV+ donors").

- Identify any SANBS practices that contribute to this phenomenon
- As possible, develop recommendations re: ways to intervene/deter such donation

To accomplish the study aim, we will administer a survey and conduct individual, semi-structured interviews (N=30) with HIV+/ARV+ donors.

Project Team

- SANBS
  - PI: Karin van den Berg
  - Recruiters/Interviewers: Cynthia Nyoni (CyN), Cecilia Nomsobo (CeN), Wendy Ntaka (WN), Debbie Strydom (DS)
- University of California, San Francisco
  - Qualitative Lead: Shana Hughes

Standard Procedures

I. Generating Client Lists, Sampling

The sampling frame consists of donors who: donated between 1 January and 31 December 2017 (as well as all donors enrolled as HIV Cases in the REDS-III HIV/HBV Case Control study), and tested positive for HIV and ARV.

Once the list of these donors is available, KvdB and SH will review and establish guidelines for purposive sampling. Overall, the goal will be to obtain a pool of participants who are diverse in ways that may shape their experience (gender, race/ethnicity, age, place of residence, 1st time vs. repeat donor, date of donation, mobile vs fixed site, having previously been counselled by SANBS staff, etc.). This master list will be stored on secured servers at both SANBS and UCSF. These potential participants will be distributed onto lists that include information such as their name, donor ID and unit # for qualifying donation. These lists will be different for each recruiter/interviewer, who will be responsible for recruitment outreach to the individuals on her list.

II. Recruitment

A. Contacting Potential Participants

There are separate Recruitment Scripts for potential participants who were previously counselled vs. potential participants who will be counselled in conjunction with this study. Please make sure you use the correct script!

Interviewers should review their recruitment lists and identify any potential participants who have not been notified of/counseled about their HIV status. **For any such individuals, standard SANBS procedures for notification and counseling should be followed BEFORE undertaking study recruitment**. Study recruitment should only happen if the interviewer feels the participant is emotionally able to handle it (please note this in the Comments column of the Recruitment spreadsheet), and should follow the **Recruitment Script for HIV+ ARV+_UNCOUNSELLED**. Recruitment efforts can be postponed to a later time if interviewer determines this to be necessary (please note in the Comments column of the Recruitment spreadsheet). For potential participants who *have* already been counseled about their HIV status, please follow the **Recruitment Script for HIV+ ARV+.**

Each interviewer will attempt to contact her assigned potential participants by phone, using the following outreach scheme:

2 attempts over 5 days (e.g., Monday and Thursday) in week 1,
2 contacts in 2nd week (e.g., Tuesday and Friday), and
1 contact in 3rd week (e.g., Wednesday).

Each contact attempt should be made at a different time of day. If no response is obtained, discuss further action with Project Team at weekly meeting.

All contact attempts and recruitment outcomes should be documented in the Recruitment spreadsheet, which should be maintained on BOTH shared network folders. See recruitment scripts for example language to use in verifying client identity, assessing eligibility**, describing the study and participation incentive. Initially, all participants will be offered the chance to complete both the ACASI and the interview. All interviewees must ideally have completed the ACASI.

**Interviewers should assess the client's cognitive competence and comfort with completing an interview in English, and make a note in the 'Comments' column.

B. Documenting Outcomes

Recruitment outcomes should be documented in the 'Recruitment' spreadsheet Please note: there are separate columns for ACASI and interview. Record outcomes for both (as needed—use N/A if participant is not being recruited for the interview):

1. If the client consents to participate, mark '**CON'** under 'Outcome'.
2. If the client declines participation, mark '**DEC'** under 'Outcome' and indicate reason for not participating in 'Reason for Decline'. Delete client's name from recruitment spreadsheet.
3. If the client cannot be reached, mark '**UTL'** ('Unable To Locate') under 'Outcome'. Delete client's name from recruitment spreadsheet.
4. Please remember to delete ONLY the client name (not the entire entry) for clients who are UTL or DEC—these data are important for determining our recruitment outcomes.

C. Post-recruitment

1. During recruitment, interviewer will ascertain feasible interview location and reminder preference (call, SMS, email), and determine mutually-acceptable day and time. Interviewer will include this information on Recruitment spreadsheet ("Data collection appt" columns).
2. Right after the recruitment call, please send a message (SMS, WhatsApp, etc.) including the interviewer's contact info (so participant has easy access). Please book/reserve the private location for the study visit.
3. Once data collection is scheduled, the interviewer should contact the PI, KvdB, to transfer the necessary funds to the interviewer's account as payments will be done in cash. R600-00 will be paid into each interviewer's account upfront to ensure that there is always sufficient funds should an urgent or unscheduled interview come up. Each interviewer should therefore have R1200-00 available in the days leading up to the interview and a R600-00 base fund in the days following the interview. Any unused funds will be returned to KvdB at the end of the enrolment period of the study.
4. Interviewer should call/SMS/email to remind participant of scheduled interview 1-2 days prior to scheduled date. Repeat request that participant notify you if s/he needs to meet earlier/later (& provide contact info if necessary)

III. Study visit & immediately afterward

A. Preparations

i. The day before the interview, please confirm the location where data collection is scheduled to occur is still available/reserved.

ii. Assemble/check the data collection packet. It should contain:

1. 2 copies of Study Information Leaflet (one for interviewer, one for participant to keep if s/he wishes), 2 copies of Informed Consent signature page (an extra in case of mistakes)
2. Materials needed for ACASI:
    a. Well charged touch-screen computer with power supply
    b. Headphones
3. 2 copies of interview guide, copy of Must-Gets list
4. Incentive in envelope
5. Incentive distribution documentation
6. Digital recorder(s), extra batteries,
7. Pen, paper, tissues
8. Light refreshments—these should be obtained specifically for our participants. A cold drink and sandwich, or cold drink, chips and something else small should be sufficient.

iii. Interviewer should consult the recruitment spreadsheet to determine the appropriate participant identification number, or PTID. PTIDs are not actually assigned until data collection begins (to avoid assigning PTIDs to participants who no-show). For more on PTIDs, see section C, below.

iv. Interviewer should arrive at location for data collection 30 min. prior to scheduled appointment, to confirm availability and set up (This will be different if bringing ppt to location)

B. Data Collection

i. 10 minutes before scheduled time, interviewer waits for participant in lobby. When participant arrives, escort him/her to room.

ii. Perform SANBS counselling procedures for donors not yet counselled. Conduct Informed Consent process with participant (ACASI and interview procedures are same document—explain one or both processes with participant as needed, make time for questions, etc.).

iii. Set participant up with ACASI, allow ppt to complete. (For participants who will NOT be interviewed, skip to vi).
iv. For participants who will be interviewed, offer a bio-break and light refreshments.
v. Conduct Interview (for more details, refer to Interview guide and Must Get list). At outset of recording, please be sure to state interviewer's name, interview date and time.
vi. Disburse incentive (cash in person). Ask interviewees to complete documentation.
vii. Conduct Informed Consent process related to our ability to inform participant of research opportunities that may arise in the future.
viii. Once study procedures are completed, offer participant use of the restroom prior to leaving. In general, participants should be escorted to and from restroom as necessary.
ix. Escort participant to exit.

C. Post-Interview / Transcription

i. Assign PTID. PTIDs are four digits long and apply to both the ACASI and the interview (for ppts who complete both). PTIDs are made up of two components: the interviewer # and the numeric order of data collection (specific to the *interviewer*, not in the study as a whole).
    1. Interviewer #s are as follows: WN = 11, DS = 22, CeN = 33, CyN = 44
    2. "Numeric order of data collection" denotes the chronological order in which the interviewer in question collected the data. Thus, 3302 is CeN's second participant. 2202 is DS's second participant.
ii. Update recruitment spreadsheet and/or incentive documentation with ACASI and interview information, including PTID.
iii. Download ACASI data to secure SANBS server (follow updated ACASI Work Instruction sheet)
iv. For interviews: Transfer the recording (digital audio file) to appropriate folder on both secure file spaces (INTERVIEWS – AUDIO folder). Apply naming convention.
    1. Naming convention follows this structure: SANBS_PTIDPpt gender (M, F, O) (Ex: file name: SANBS_1107F)

IV. Verify integrity of audio file on both secure file spaces. Once verified, delete the audio file from the recorder.

V. Interviewer securely transfers audio file to transcriptionist. Provide a brief description and any relevant details about audio file.

VI. Use template and notes taken during interview to complete Fieldnote, upload to both secure file spaces.

VII. When transcripts are completed, [[transcriptionist gets document(s) to interviewer]]. The interviewer uploads file to "Original Interview Transcripts" folder on both secure file spaces, and updates spreadsheet with date received.

VIII. Interviewer reviews and de-identifies her own transcripts, then saves in designated folder ("Transcripts_ReadyForAnalysis") in both secure file spaces. Interviewer updates spreadsheet (input date in "Transcript cleaned by Interviewer" column).

IX. Email Shana to let her know a new transcript is ready for review.

X. Shana reads through, confirms de-identified transcripts are ready for analysis (RfA)/import to qualitative data analysis software, records date in spreadsheet.

XI. Data Analysis

A. In weekly team meetings during recruitment and data collection, interviewers share issues, experiences, and progress ("Unable To Locate" participants, scheduling issues, languages other than English, any ACASI tech problems, debrief about interviews, discuss any problematic questions, share what's coming out, etc.). All team members should read each other's interview fieldnotes prior to each meeting. Team keeps notes on emerging themes/patterns, questions, new directions to pursue, etc. These notes are saved in "Weekly Meetings" folder on secure file space.

B. As transcripts are RfA, Shana reads each one, writing a brief analytic memo to complement the Fieldnote drafted by the interviewer. She also keeps a list of potential codes and definitions, adding and revising as data collection continues. These can be discussed in weekly meetings as well. These materials are saved in the "Analysis" folder on the secure file space.

C. First pass coding

D. Second pass coding (by secondary analyst)

E. Reconciliation of coding, standardization of code book

F. Karin and both analysts confer and select themes to summarize

G. Summarization/pattern identification and negative case-finding

H. Results write-up and draft rec's for SANBS (as possible); presentation?

I. Member reflection (?) and results refinement

J. Manuscript planning, writing

ANNEXURE 3: TELEPHONE RECRUITMENT SCRIPT FOR HIV+/ARV+ DONORS (SANBS)

NOTES TO INTERVIEWERS:

1) Prior to calling a potential participant, please make sure the individual has already been notified/ received counseling for his/her HIV status. **If not, please follow standard SANBS guidelines for those activities, then continue with the alternate recruitment script for this study**.
2) During these phone calls, interviewers should assess the donor's cognitive competence and comfort with completing an interview in English, and make notes as needed in the 'Comments' column on the recruitment spreadsheet.

SCRIPT:

Hello. May I speak to _____?

➔ If no answer/donor not available, please leave this message:

Hello, this is _____ (*caller first name*) with the South African National Blood Service calling to speak with _____ (*potential participant name*). I can be reached at [provide interviewer's number for call-back]. Thank you.

[If you are speaking with correct person:]

Hello, (*potential participant name*). This is _____ (*caller first name*) and I'm with the South African National Blood Service. **Do you have about ten minutes to talk**?

If NO: "I understand. It is important that I speak with you, so when could I call back?"

_____ [*document if donor refuses a call-back*].

If YES: Great. First, let me set your mind at ease: this call is no cause for worry. I'm reaching out to you to let you know about an important research study SANBS is inviting you to be part of. Now, I know we were in touch with you in _____ (month/year donor was counseled) to share some test results and provide counseling about a previous donation you made. Does that sound familiar? *[Likely the person we are calling will say yes, and then recruiter can go on from there. If potential participant says No, recruiter can respond, "That's OK, I just mentioned it because those same test results qualify you to be in this study. I'd like to tell you what we'd be asking of you if you decide to participate…." and continue with script]*

Those same test results qualify you to be in this study, so I'd like to tell you what we'd be asking of you as a participant. Feel free to jump in and ask questions as we go along, OK? (wait for affirmation from donor)

Ok, the goal of this study is to help SANBS understand how to serve certain kinds of donors better and keep the blood supply safe. If you decide to take part, you would come to a study visit and ideally do two things: First, use a computer to take a survey, which usually lasts about 30 minutes. Participants who take the survey receive R250. Second, do an in-person interview with a research nurse. That usually takes about

another hour. People who do the interview get an additional R300, making R550 for both the survey and the interview. *[Only provide this information for ppts being invited to the interview.]* It's important that you know that everything you share with us will be kept private. Do you have any questions so far? [answer any questions]

***If* donor declines at this point:** I understand. Could I ask why you have decided not to participate? [if donor has misunderstood something or is declining for a reason we can fix, please try to clarify/adapt. If decline is firm, document reason in spreadsheet]

OK. If you change your mind or have any further questions, I can be reached at [provide interviewer's call back number]. Thank you for your time. Enjoy the rest of your day.

If OK to proceed: Ok, I'd like to go over a few more details with you. You might want to have privacy to hear about this. Are you in an OK place for that? [[wait until OK]] We're only doing this study with people who had a previous donation test positive for HIV medication—this is often called ARVs, or ART or just "HIV treatment." The test results from your last donation make you eligible, so that's why we want to hear from you. At your study visit, I will probably be the one helping you and we'll talk about how you feel about blood donation, how you found out you had HIV, your HIV care, and things like that. [*If recruiter knows ppt has denied HIV or ARV use, she can add here: "If you feel those questions don't apply to you, we'll be able to have a discussion about that."*] You can refuse to answer or skip questions that make you feel too uncomfortable. We know sometimes people have questions about who will have access to what they share during the study, so let me assure you that it will only be members of the study team. In addition, to protect your privacy, we will use a study number instead of your name in reports and written records. Also, nothing you share will be used against you in any way. I know this is a lot to take in; do you have any questions? [Answer ppt questions] **Do you think you might be interested in helping us**?

***If* participant protests s/he is not taking ARV/Not HIV+:** Ok, I understand that you don't feel you qualify. The thing is, SANBS does lots of tests on samples of donated blood, and yours did test positive, so it's important for us to learn how this might have happened. We'd really like to give you a chance to tell your story….[continue at **&&**]

***If* NO:** I understand. Could I ask why you have decided not to participate? [if donor has misunderstood something or is declining for a reason we can fix, please try to clarify/adapt. If decline is firm, document reason in spreadsheet]____

OK. If you change your mind or have any further questions, I can be reached at [provide interviewer's call back number]. Thank you for your time. Enjoy the rest of your day.

If YES: That's wonderful, we really appreciate that. [continue at **&&**]

&& In terms of planning for the study visit, we estimate that doing both the survey and the interview will take about 2 hours. Would it be possible to find 2 hours for you to participate?

[[Listen to and address ppt concerns/questions about time needed, if any. NOTE: In this project, there is a strong preference for participants to do *both* the survey and interview, but if needed/desired, offer survey only. e.g., "For participants who complete only the survey the study visit should take 1 hour or less—would that be easier for you?"]]

Another important detail is that all data for this study are being collected in English. We know not everyone may feel comfortable speaking about these issues in English—how do you think that would be for you?

→ ***If ppt expresses discomfort:*** Discuss this with him/her and reassure ppt that his/her English does not have to be perfect, that words here or there in another language are OK. Note relevant details in the recruitment spreadsheet "Comments" column. If ppt self-determines (or if interviewer decides) that s/he is unable to participate due to language issues, please include what language the ppt *would* feel comfortable in. You can tell ppt that we will call back if our research procedures change in a way that would allow their participation. Be sure to thank him/her for considering the study.

➔ ***If ppt feels comfortable in English:*** That's wonderful. Ok, something else we want to make sure you know about now is that we would like to make an audio recording of the interview. The reason we do this is so the study team can do a better job with data analysis. We just want to make sure we get your story right. **Do you have any questions about that**? [address ppt concerns]

Now, as I mentioned, you will receive an incentive at the end of the study visit, and this will be in cash. We will have to complete a little bit of paperwork about that but it goes pretty quickly. **Would you like to talk about scheduling a study visit?**

***If* NO:** I understand. Could I ask why you have decided not to participate?

_____

"In case you have any further questions or if you change your mind about participating, we can be reached at #####. Thank you for your time. Enjoy the rest of your day."

If YES: Great! Now, as I mentioned before, the study visit will most likely be with me, though if necessary one of my colleagues may help instead. In your area, we are able to host you at [[list available options]]. At these locations we know we can ensure privacy for you to complete the study activities. Is it possible for you to meet us at one of these places?

***If* NO:** TBD WITH TEAM!! [*As interviewers feel comfortable/safe, they can offer to pick up participant, or can arrange for some other transport for ppt.]*

***If* YES:** Thank you [agree on location with participant]. We try to work with people to find a study visit time that's convenient for them. We can usually be available from [[TIME]] to [[TIME]] on [[LIST DAYS]]. Is there a day and time in that range that works well for you? Remember, we're asking for a block of time when you would available to be at the study location for about 1 hour //2 hours.

***If* NO: [[If the** times/days participant would be available are slots the recruiter can accommodate, please schedule and then check in about this with the team (esp. Karin). If the interviewer cannot make the times/days the ppt is available, please document the preferred visit times and reach out to the team to see if another interviewer can handle the appointment. Tell the participant you will be back in touch with him/her as soon as possible.]]

***If* YES:** Thank you—that sounds great. I will meet you at [[agreed-upon LOCATION]] so you can complete the survey//the survey and interview on [[DAY, DATE, at TIME]].

To help the study visit go smoothly, we ask that you do your best to be on time and come by yourself. Friends/partners will be asked to return after you have finished. If anything should prevent me from meeting you as we've planned, I will get in touch—what's the best way for me to do that? [[document in spreadsheet]] Also, could I ask you to let me know if you need to see me earlier or later? Again, my name is _____ and my number is _____.

I'd like to just confirm the telephone number we have for you [confirm number used to speak with participant]. Is this the number you want us to use in the future? [[**Obtain alternate number if ppt says**

NO]]. Is this a home or mobile number [note H/M for spreadsheet]? Is it ok for the interviewers to leave a message for you at that number [note Y/N for spreadsheet]? [For mobile numbers:] Is it ok to send an SMS to this number [note Y/N for spreadsheet]? **Is there another number you'd like the interviewers to have**?

If YES: [Note additional number for spreadsheet] Is this a home or mobile number [note H/M for spreadsheet]? Is it ok for the interviewers to leave a message for you at that number [note Y/N for spreadsheet]? [For mobile numbers:] Is it ok to send an SMS to this number [note Y/N for spreadsheet]?

If NO: (skip to next section)

Ok, you also have the option to give us an email address. Would you like to do that? [If YES: note in spreadsheet].

Ok, thank you.
We usually give study participants a reminder a day or two before our meeting. Because we know there are situations where other people might hear or read your messages, to protect your privacy as much as possible, we will not mention HIV in any messages. We can remind you by phone call, SMS or email—which works best for you? [[Document ppt preference]].

Wonderful. So I'm looking forward to seeing you for your study visit at [[TIME on DAY, DATE at LOCATION]]. We will remind you a day or two before the visit by [[PHONE/SMS/EMAIL]]. Do you have any questions right now? [[answer ppt questions, if any]]. If you think of other questions later, you can reach me at _____ [provide recruiter's phone number]. Please do call if something comes up and you can't make the appointment. Thank you for your help; enjoy the rest of your day.

ANNEXURE 4: INFORMED CONSENT DOCUMENTS FOR HIV+/ARV+ DONORS PARTICIPATING IN THE QUALITATIVE STUDY

Informed Consent – Blood donors with prior knowledge of HIV and ART use

South African National Blood Service

Consent for Research

Blood donation by persons with previously diagnosed HIV infection and concurrent antiretroviral therapy in South Africa: prevalence and motivations.

Dear Blood Donor,

Thank you for taking the time to review the information below before considering whether you are willing to participate in this research project. You are being asked to take part in a research study with the above title and short title we will use in this document, "HIV+/ARV+ Study".

The person in charge of this study is Dr. Karin van den Berg from the **South African National Blood Service (SANBS).** Before you decide if you want to join this study, we want you to learn about the study. The study staff will talk with you about the study and answer your questions. Before you agree to join this study, please read this consent form carefully. Take your time in deciding if you wish to join this study. This consent form might contain some words that are not familiar to you. Please ask questions about anything you do not understand.

Who is conducting this research study?

The **South Africa National Blood Service (SANBS)** is leading this study in collaboration with researchers from the University of California San Francisco and Blood Systems Research Institute in the United States as well as the University of Cape Town. The data collected for this study will be analysed in South Africa and the United States and results reported in medical journals. The results of the study may be used to improve blood safety in South Africa and other countries in Africa. The study is supported financially by the South African National Blood Service as well as the National Heart, Lung and Blood Institute in the USA.

What is the purpose of this research study?

The study will perform research on HIV-positive donors who have donated blood while on antiretroviral therapy (ART). The aim of the study is to understand why such donors present to donate blood.

What will happen if you participate in this study?

This study consists of two parts, namely:

- Completing a computerized survey
- Participating in a semi-structured interview with a research nurse

Procedures:

If you agree to participate in the study, you can elect to do only the survey or both of the study procedures.

For the computerized survey:

You will complete a confidential questionnaire using a computer to answer questions about your HIV diagnosis, your knowledge about HIV/AIDS and about the motivations that took you to donate blood. You may skip any questions that you are not comfortable answering.

For the semi-structured interview with a research nurse:

The study nurse will ask you questions about your history of HIV testing, diagnosis, and treatment initiation; the impact of HIV on your life; your ideas about the risk of passing HIV to someone else; perceptions of blood donation and the healthcare system; and the timing, context, and reason for your donation. These interviews often feel a lot like normal conversation, though some of the questions may make you feel uncomfortable. You may refuse to answer any of the questions and all your answers will be kept confidential. We ask permission to audio record the interview to ensure high quality analysis of the information you share.

How many people will be in this part of the study?

We anticipate that about 100 people will complete the computer interview and about 15 to 30 people the semi-structured interview.

Are there risks to you for participating in the study?

Risks:

1. Survey and Interview: Some of the questions could make you upset or embarrassed you can refuse to answer any question or stop the survey or interview at any time.

2. Confidentiality: As in any research, there is a very small chance that your personal information could become known to others. To lessen the chance of this, the survey and blood samples will be identified by code numbers and not your name. Audio recordings of all interviews will be stored securely and deleted at the end of the study. Identifying details will be removed from interview transcripts.

Will I be paid and are there any costs to the research?

You will be to compensated you for your time and transportation to the study center. For participating in the computerized survey interview you will be compensated R250-00. If you also participate in the semi-structured interview, you will receive R550-00 and (R250-00 for the survey and R300-00 if you participate in the semi-structured for the interview). If you participate in both studies, you will receive R550-00 in total.

What if I don't want to participate after I have completed the study?

You do not have to participate in this study and you may retract your consent for participating at any time by contacting the investigator listed on this consent form. If you decide to remove yourself from the study, your questionnaire and interview responses will be deleted from the study databases. However, if the data have already been analysed and reported in medical journals we will not be able to remove you from the study. Your decision to remove yourself from the study will not affect your relationship with SANBS in any way.

Questions you may have:

You can have any questions you may have answered by the responsible investigator, before and during the research. If you have questions right now please ask them before signing this consent.

Informed Consent Signature Page – BLOOD DONORS WITH PRIOR KNOWLEDGE OF HIV AND ART USE

Blood donation by persons with previously diagnosed HIV infection and concurrent antiretroviral therapy in South Africa: prevalence and motivations.

If you have any questions about this research study or if you are injured as a result of the research you may contact the following at any time:

South African National Blood Service Contact Person:

Name:	Dr Karin van den Berg
Telephone Number:	041-391-8269 or 082 578 7045
E-mail:	Karin.vandenberg@sanbs.org.za

You may also contact the Secretariat of the Ethics Committee of SANBS, at telephone number 011-761-9135 or email Valencia.Simmadari@sanbs.org.za if you have questions about your rights as a research participant.

Your participation in this research is voluntary, and you will not be penalized or lose benefits in anyway if you refuse to participate or decide to stop participating.

If you agree to participate, you will be given a signed copy of this entire informed consent document, which provides you with a written summary of the research.

I DECLARE THAT I HAVE READ AND UNDERSTOOD ALL THE INFORMATION CONTAINED IN THE CONSENT DOCUMENT AND I AGREE TO PARTICIPATE IN THIS RESEARCH STUDY. I AM FREE TO RETRACT MY CONSENT IN ANY PART OF THE RESEARCH IF I DECIDE THAT I DO NOT WANT TO CONTINUE PARTICIPATING.

I agree to participate in the computerized survey: Yes / No

I agree to participate in the semi-structured interview: Yes / No

Name: _____

Signature: _____

Date: _____/_____/_____

Signature of study staff taking consent:

I declare that the above participant has been fully informed about the nature, conduct and risks of the above study.

Name: _____

Signature: _____

Date: _____/_____/_____

ANNEXURE 5: SEMI-STRUCTURED INTERVIEW GUIDE FOR ONE-ON-ONE QUALITATIVE INTERVIEWS WITH HIV-POSITIVE DONORS ON ARV AT TIME OF DONATION

Introduction

Thank you once again for responding to the questions on the ACASI. Because everyone in the study will answer those same questions, it will help us learn the same information about each participant. Now we'll do something a little different. We know that everyone who has donated blood while living with HIV and taking ARVs has their own story that might not be like anyone else's, so in this part of the study, we will ask questions and let you respond. Some of these questions might seem similar to what you just answered, but I don't have access to the ACASI answers, so don't worry if you feel like you're repeating yourself, or if you talk about things in a different way. There is a list of topics for us to cover, but we want you to feel at ease, a lot like a normal conversation. Some of the questions we'll ask might seem embarrassing or a little uncomfortable, but this isn't an evaluation or a test—there are no "wrong" answers. What we want most of all is just to hear your story, and the whole study team is grateful you've chosen to share it. It would be really great if you can include concrete examples from your life, though, remember, you can decide not to answer any question that you feel is too upsetting or private. Finally, rest assured that information you share with us will not be used against you, and we'll protect your confidentiality by using an ID number for you instead of your name. When we share the results of the study, you may see your words, but no one will know who said them.

[[*Check interviewee comprehension of topic and process*]]

Ok, great. We have many things to talk about, and I want to make sure we get to hear your thoughts on all of them, so I apologize in advance if I cut in to make sure I understand what you're saying or to change the topic. Please let me know my questions are unclear or you have questions as we go along. We also talked before about making an audio recording of our conversation, so once I start the recorder I will state the date, time and interview number, then ask your permission to be recorded. Let's both try to remember to speak clearly and loudly enough to be heard on the recording, ok?

Do you have any questions? …Are you ready to start the interview?

[[*Once interviewee is ready, start recording, state who is conducting the interview, date, time and interview number. Ask "As we mentioned earlier, we are recording this conversation to aid in data analysis for the study. Do you accept being recorded?"]]*

Section 1: Intro questions (life context)

[This section allows interviewees to become more comfortable with talking to the interviewer, the recording, etc. It gives interviewers a chance to demonstrate they are friendly and safe. Use probes/follow-up questions as needed, but don't let this go too long. Ideally <10 minutes. The reason there are two separate questions is to give the interviewee a chance to experience having a back-

and-forth with the interviewer, and to give the interviewer her first chance to see how talkative the interviewee might be in this context (sometimes people act differently during the interview vs. recruitment phone calls).]

Great, thank you for that. Now, when I'm doing an interview like this, it really helps me to get to know the person I'm talking to a little. Would you mind telling me a bit about yourself?

Possible probes: *Anything you feel comfortable sharing: where you grew up, your family, or your favorite things to do with friends?* [[*Interviewer can also ask about age, place of current residence, work situation, etc.*]]

I'm interested in understanding a little about what your life is like these days. What can you tell me about family, work, hobbies, church/mosque—things like that?

Section 2: HIV (diagnosis, ARVs, impact, perception of transmissibility)

[Where a donor insists that they did not know that they are HIV or taking ARV, the interview can proceed asking the participant to respond in the hypothetical, but be sure to allow the participant to refuse to continue]

Thank you for sharing that. It will help me understand your answers as we move through the interview. You know you were invited to do this interview partly because of your HIV status, so could you tell me when and how you found out you had HIV?

[In the HIV research I've done, sometimes this is a long story…be mindful of interviewee's need to tell the story AND need to move through this story to get to the rest of the questions! It will be helpful to understand the context of testing (why test? was the intv in ill health? Did s/he suspect status? Where was testing done and why there? Was the intv alone or with someone?]

How did you feel when you received the news? …How did you handle it?

Possible probes: *Do you remember getting any counseling or information about HIV when you were diagnosed? …Do you/Did you use any other resources to learn about HIV and/or treatment?*

And what about treatment? When and how did you start taking ARVs?

Possible probes: *How was that for you? …Are you taking treatment now? …Have you ever had to stop or switch your medication for any reason? …Have you taken any "treatment holidays"?*

How is your health now? …What kinds of things do you do to take care of yourself?

[Some interviewees tend to answer this question with their viral load or CD4 count, and that's fine if that happens—it tells us about how people understand "health". But let's also think about health in a way that includes more than HIV!]

I'm curious what you think about passing HIV to someone else—how easy or difficult do you think that would be?

[[*Probe for participant understanding of UVL and/or any differences associated with particular modes of transmission Here is one of the places where we need to listen for unexpected understandings of what "undetectable" or "functional cure" mean to interviewees and how that might lead them to think they can donate even if they don't stop taking their HIV treatment.*]]

I'd like to understand your thinking around sharing or not sharing your HIV status with others. Have you told anyone? …Who have you shared this with?//Tell me about your decision not to tell people

Possible probes: *Could you tell me what that conversation was like? Is there anyone who knows your status that you can count on for help/support? Is there anyone you're thinking about telling but you haven't decided yet (how does ppt decide who to tell?)?*

Thinking about your life as a whole, how would you describe HIV's impact on you?

Possible probe: How does your life now compare to before you were diagnosed?

Section 3: Perceptions of care

Where do you receive care for HIV? …What's that like?

Possible probes: *How often do you go? …What usually happens when you go there? …How did you end up getting care there and not somewhere else? …How do you feel about the care you receive?* [[*Probe for feeling about medical providers/staff, understanding of and satisfaction with labwork, VL, resistance testing, etc.]]*

Is there anything you wish you could get that isn't part of your treatment at (fill in location)?

Section 4: Timing, context, and motivation of blood donation

Ok, let's switch gears and talk a little now about blood donation. How many times would you say you've donated blood?

[[***If the donation that qualified them for the study was their first, skip to next question***. ***If they've donated more than the most recent time that qualified them for the study, ask****: Could you tell me about the first time—how old were you, where did you donate, why did you want to—all that stuff? …Has blood donation changed for you in any way since that first time? How so? …Has there been a time when you were unable to donate? → If intv has not mentioned it yet, ask "And what was your HIV status that first time you donated?"]]*

Ok, how about this (most) recent donation—could you tell me the story of how that happened? I'd love to really understand the details: When was it, who were you with, what were you doing when you decided to go, things like that.

What made you want to donate at that time?

I'd like to learn a little more about the place where you donated. Could you tell me what was it like? [[*esp. check for mobile vs. fixed*]]

[If participant does not mention it on his/her own, this section is where we should probe about privacy issues in terms of completing the questionnaire and the location for the pre-donation interview.]

Ok, and I understand you fill out a questionnaire before each time you donate blood—would you tell me about that? [[*Where did intv complete questionnaire? In what format (paper, electronic, etc.)? With others? Alone? etc.*]]

And how about the screening interview before your donation--How did that go? [[*Assess comfort with talking to staff, privacy concerns*]]

Possible probes: How did you feel discussing any possible health risks with the staff?

Was there ever a moment when you felt like not going through with the donation?

What was your understanding then of reasons why people might not be eligible to donate? …What about HIV? …What's your understanding of this now? …How do you feel about that?

There's one more thing that I understand happens lots of times after people donate blood that I'd like to ask you about. I think staff are supposed to give you a card that has some information on it, like the date you donated, a number that's assigned to your donation, a number you can call—do you remember getting anything like that? …[[*only if intv seems confused/unsure*:]] they might have called this an "honesty card"—does that sound familiar? …[[*if intv received HC, can ask, "Was there ever a time you thought of calling the number on the card? …Can you tell me about that?*]]

I'd like to know about the tests the blood center does on donated blood—what's your understanding of that? [[*probe for specific types of tests? Window periods?*]]

Do you ever talk to anyone about blood donation or have you ever encouraged anyone else to donate blood?

[This is a question I use in interviews with donors as it gets at their larger social network and how they have communicated their knowledge about blood donation. Reveals what kinds of questions/perceptions people have around them and how they contribute to this conversation in their social milieu. This may be important to understanding those conversations in relation to ARVs/HIV.]

Section 5: Wrap Up

Alright, I have just a few questions left. Is there anything the blood services could do to……communicate more effectively with the public about donation eligibility//help PLWH who are taking ARVs avoid donating blood?

[What interviewers should say here really depends on the rationale the interviewee shared for his/her donation.]

Is there anything else you want to share with me about this that we haven't discussed?

Is there anything I didn't ask you that you thought would be part of this interview?

This section is to be completed by the research assistant or other research staff.

A1. Participant ID

__ __ __ __ __ __ __

A2. Participant Donor Number

__ __ __ __ __ __ __ __

A3. Month of interview (Choose one)

01	January
02	February
03	March
04	April
05	May
06	June
07	July
08	August
09	September
10	October
11	November
12	December

A4. Year of interview (please enter four numbers)

__ __ __ __ yyyy

A5. Research Assistant Initials: __ __ __ __ __

If study participant is not already sitting at the computer, at this time please make sure the study participant is sitting at the computer and has put the headphones on.

READ: This study has been approved by Ethical Committees in South Africa and the USA.

READ: Thank you for agreeing to participate in this scientific study. Please answer these questions to the best of your knowledge and as truthfully as you can. You may skip any questions that you are not comfortable answering. Please keep in mind that while the researcher will make every effort to keep your responses confidential, there is a small chance they will not be able to. However, your name is not collected on the questionnaire and your responses cannot be easily traced back to you.

READ: In this section of the questionnaire the research assistant will show you how to use the computer to answer the interview questions. After completing this section, with the help of the research assistant, you will be left to complete the interview in private. If you have any questions at any time or are unsure of what to do, please ask for help.

B1. What is your gender? (Choose one)

1	Male
2	Female
3	Other
7	Don't Know
8	Refuse to Answer

If B1 is not equal to 3, then skip to B3.

B2. How would you describe your gender?

__ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __

B3. What is your birth year?

__ __ __ __

9997 Don't Know

9998 Refuse to Answer

B4. What is your birth month? (Choose one)

__ __ January
__ __ February
__ __ March
__ __ April
__ __ May
__ __ June
__ __ July
__ __ August
__ __ September
__ __ October
__ __ November
__ __ December
__ __ Don't Know
__ __ Refuse to Answer

B5. What is your birthday?

__ __

97 Don't Know

98 Refuse to Answer

B6. What is your country of birth? (Choose one)

01 South Africa
02 Zimbabwe
03 Malawi
04 Mozambique
05 Swaziland
06 Botswana
07 Lesotho
08 Namibia
09 Nigeria
10 Other
97 Don't Know
98 Refuse to Answer

If B6 is not equal to 10, then skip to instruction before B7.

B6B. Please specify your country of birth

__ __
__ __ __ __ __ __ __ __ __ __ __ __ __ __ __

READ: From now on, you will be left alone. It means that you will have total privacy to answer all these questions. Please, if you have any questions call the research assistant for help.

B7. What is your race / ethnic origin? (Choose one)

1 Black
2 White
3 Coloured
4 Asian
5 Other
7 Don't Know
8 Refuse to Answer

B8. What is the primary language that you speak at home? (Choose one)

01 isiZulu
02 isiXhosa
03 Afrikaans
04 Sepedi
05 Setswana
06 English
07 Sesotho
08 Xitsonga
09 siSwati
10 Tshivenda
11 isiNdebele
12 Other
98 Refuse to Answer

B9. What is your current marital status? (Choose one)

1 Single, never married
2 Living with partner, but not married
3 Married to one partner (including traditional marriage)
4 Married to more than one partner (including traditional marriages)
5 Separated/divorced
6 Widowed
97 Don't Know
98 Refuse to Answer

B10. What is the highest level of education you have completed? (Choose one)

0 Never been to school
1 Up to Grade 7 / Standard 5
2 Up to Grade 10 / Standard 8
3 Up to Grade 12 / Standard 10
4 Incomplete further degree or qualification (some college or technical school)
5 College or technical qualifications
6 University or professional degree
97 Don't Know
98 Refuse to Answer

B11. What is your religion or affiliation? (Choose one)

01 Christianity
02 Islam
03 Hinduism
04 Judaism
05 African traditional beliefs
06 Other faiths
07 No religion
97 Don't Know
98 Refuse to Answer

B12. Do you have medical aid?

1 Yes
0 No
8 Refuse to Answer

B13. Do you self-fund access (pay out of pocket) to private doctor or private hospital?

1 Yes
0 No
8 Refuse to Answer

B14. Are you currently working? (Choose one)

1 Yes, self-employed
2 Yes, employed full time
3 Yes, part time
4 No, unemployed
97 Don't Know
98 Refuse to Answer

If B14 is equal to 4, then skip to instruction before C1.

B15. What type of work are you doing? (Choose one)

01 Mining
02 Transport / cargo delivery
03 Military / police
04 Medical / healthcare
05 Business / sales / retail
06 Farming
07 Teacher / education / student
08 General labour (domestic worker, gardener, janitorial)
09 Civil service (examples including working in government office, post office)
10 Other
Don't know
Refuse to answer

If B15 is not equal to 10, then skip to B17.

B16. What is your occupation

____

B17. What is the combined monthly income after tax for your household? (Choose one)

1 Less than R5,000-00
2 R5,001-00 to R15,000-00
3 R15,001 to R30,000-00
4 More than R30,000-00
97 Don't Know
98 Refuse to Answer

READ: From now on, you will be asked more sensitive questions. Please be assured that we are just trying to learn more about this situation. Please, if you have any questions call the research assistant for help.

C1. What year did you first find out that you were HIV positive? (If you don't know exactly, please enter your best guess)

__ __ __ __ yyyy
0097 Don't Know (Year)
0098 Refuse to Answer (Year)

C2. What month did you find out that you were HIV positive? (Choose one)

01 January
02 February
03 March
04 April
05 May
06 June
07 July
08 August
09 September
10 October
11 November
12 December
97 Don't Know
98 Refuse to Answer

C3. Where were you tested for HIV when you first found out your status? (Choose one)

1 General Practitioner
2 Local clinic/health facility
3 Hospital
4 HIV Testing Centre (known as a Voluntary counseling and Testing of VCT Centre)
5 Other test site
7 Don't Know
8 Refuse to Answer

If C3 is not equal to 5, then skip to C5.

C4. Please tell us the other test site.

__ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __ __

C5. Can you remember if your CD4 count was tested at that time?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

If C5 is equal to 0, then skip to C8.

C6. Can you remember more or less what it was?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

If C6 is equal to 0, then skip to C8.

C7. Please tell us what it was.

— — — —

9997 Don't Know
9998 Refuse to Answer

C8. Can you remember if your viral load was tested at that time?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

If C8 is equal to 0, then skip to C11.

C9. Can you remember more or less what it was?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

If C6 is equal to 0, then skip to C11.

C10. Please tell us what it was.

— — — —

9997 Don't Know
9998 Refuse to Answer

C11. What year did you start anti-retroviral treatment for your HIV? (If you don't remember exactly, please enter your best guess)

— — — — yyyy
0097 Don't Know (Year)
0098 Refuse to Answer (Year)

C12. What month did you start anti-retroviral treatment for your HIV? (Choose one)

01 January
02 February
03 March
04 April
05 May
06 June
07 July
08 August
09 September
10 October
11 November
12 December
97 Don't Know
98 Refuse to Answer

C13. Has your anti-retroviral treatment ever been changed?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

If C13 is not equal to 1, then skip to C17.

C14. How many times has your anti-retroviral treatment been changed?

__ __

97 Don't Know
98 Refuse to Answer

C15. Please tell us what the reason for changing your medication was? (Choose one)

0 The medication wasn't working
1 The medication cause side effects or problems
2 My usual medication was not available
3 Other
7 Don't Know
8 Refuse to Answer

If C15 is not equal to 3, then skip to C17.

C16. Please tell us what the reason for changing your medication was.

__ __

97 Don't Know

98 Refuse to Answer

C17. Are you still taking your anti-retroviral treatment?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

If C17 is equal to 0, then skip to instruction before D1.

C18. How regularly do you take your treatment? (Choose one)

0 Every day

1 At least 5 days a week

2 At least 3 days a week

3 Less than 3 days a week

7 Don't Know

8 Refuse to Answer

Section D - HIV Knowledge Domain

READ: The following questions will ask you about things that you may know about HIV/AIDS.

D1. Is HIV/AIDS spread by kissing?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

D2. Can a person get HIV/AIDS by sharing kitchens or bathrooms with someone who has HIV/AIDS?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

D3. Can men give HIV/AIDS to women?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

D4. Can women give HIV/AIDS to men?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

D5. Must a person have many different partners to get HIV/AIDS?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

D6. Does washing after sex help protect against getting HIV/AIDS?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

D7. Can a pregnant woman give HIV/AIDS to her baby?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer
9 Not Applicable

D8. Can a person get rid of HIV/AIDS by having sex with a virgin?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

D9. Is HIV the virus that causes AIDS?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

D10. Is there a cure for HIV/AIDS?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

Section E - HIV Stigma

READ: The following questions will ask you about things that you may have experienced since finding out about being HIV-positive. Use the following scale to indicate to what degree the following is true for you or have happened in your life: 1 - Not at all, 2 - Very little, 3 - Somewhat, 4 - Very much.

E1. Some people avoid touching me once they know I have HIV. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E2. People I care about stopped calling after learning I have HIV (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E3. I have lost friends by telling them I have HIV (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E4. Telling someone I have HIV is risky. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E5. I work hard to keep my HIV a secret. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E6. I am very careful who I tell that I have HIV. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E7. People with HIV are treated like outcasts. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E8. Most people believe a person who has HIV is dirty. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E9. Most people are uncomfortable around someone with HIV (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E10. I feel guilty because I have HIV. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E11. People's attitudes about HIV make me feel worse about myself. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

E12. I feel I'm not as good a person as others because I have HIV. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

Section F - Blood Donation Knowledge and Perceptions

READ: The following questions will ask you about what you know and understand about blood donation. Please answer these questions to the best of your knowledge and as truthfully as you can.

F1. Did anyone ever speak to you about having HIV and donating blood?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

If F1 is equal to 0, then skip to F4.

If F1 is equal to 97, then skip to F4.

If F1 is equal to 98, then skip to F4.

F2. Who was it that spoke to you about having HIV and donating blood? (Check all that apply)

__ Doctor
__ Nurse
__ Family
__ Friends
__ Blood Centre staff
__ Don't Know
__ Refuse to Answer

F3. Did they tell you that you should no longer donate blood?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

F4. Did you ever speak to anybody about donating blood after you were diagnosed with HIV?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

F5. At the time of your last donation did you think that SANBS would test your blood for HIV?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

F6. At the time of your last donation did any of the SANBS staff speak to you about the tests that would be done on your blood?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

F7. Did you think SANBS would find HIV in your blood?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

F8. At the time of your last donation did any of the SANBS staff speak to you about the risk of HIV to the people who receive your blood?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

F9. At the time of your last donation did you ask any of the SANBS staff anything about HIV?

1 Yes
0 No
7 Don't Know
8 Refuse to Answer

F10. How do you think the testing done by SANBS compares to what is done in the public clinics or hospitals? (Choose one)

1 Worse
2 Better
3 The same
7 Don't Know
8 Refuse to Answer

Section G - Blood Donation Motivation

READ: The following questions will ask you about factors that may have influenced your decision to donate blood. Use the following scale to indicate how much the factors influenced your decision to donate blood. 1 - Not at all, 2 - Very little, 3 - Somewhat, 4 - Very much

G1. I felt I could help someone by donating blood. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G2. Everybody who can donate blood should do so. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G3. People who donate blood will get their blood for free if they need it in future. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G4. I heard there was a blood shortage. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G5. I felt I could not say no. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G6. I wanted to receive the gift that was being handed out. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G7. I wanted to receive my 4th donation gift. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G8. The blood drive organizer or recruiter strongly encouraged me to donate. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G9. My family or friends strongly encouraged me to donate. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G10. My co-workers or someone else strongly encouraged me to donate. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G11. A doctor or nurse told me to donate for health reasons. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G12. A doctor or nurse told me to donate for an HIV test. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G13. Who most influenced your decision to donate blood? (Choose one)

1 Family member
2 Friends
3 Co-workers
4 Blood drive organizer
5 SANBS recruiter
6 Doctor or Nurse
97 Don't Know
98 Refuse to Answer

G14. An advertisement of the radio or TV or in a newspaper or magazine encouraged me to donate. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G15. A post on Facebook, Twitter, Instagram or other social media encouraged me to donate. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G16. A letter, telephone call or SMS from SANBS encouraged me to donate. (Choose one)

1 Not at all
2 Very little
3 Somewhat
4 Very much
97 Don't Know
98 Refuse to Answer

G17. What most influenced you to donate blood? (Choose one)

01 Radio
02 TV
03 Newspaper or magazine
04 Facebook
05 Twitter
06 Instagram
07 Letter/SMS/Call from SANBS
08 None of these
97 Don't Know
98 Refuse to Answer

G18. When you last donated, did you know what tests SANBS would do on your blood?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

G19. Did you think that the blood you donated may infect a patient if it was transfused to a patient?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

G20. On the day you last donated, you were asked whether you were HIV positive. Did you understand the question?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

G21. On the day you last donated, you were asked whether you were taking anti-retroviral drugs. Did you understand the question?

1 Yes

0 No

7 Don't Know

8 Refuse to Answer

READ: Thank you for taking the time to complete this questionnaire. Please return this questionnaire to the research staff.

READ: If you have any questions or concerns, please talk to the research assistant or nurse. You can also contact the medical director at our blood bank.

ANNEXURE 7: SUPPLEMENTARY TABLE 1: HIV STIGMA SCALE SECTION OF THE SURVEY CONDUCTED AMONG HIV+/ARV+ DONORS

Adapted from Reinius *et al*.

READ: The following questions will ask you about things that you may have experienced since finding out about being HIV-positive. Use the following scale to indicate to what degree the following is true for you or have happened in your life: 1 - Not at all, 2 - Very little, 3 - Somewhat, 4 - Very much.		
Subscale 1: Personalised stigma		
E1.	Some people avoid touching me once they know I have HIV. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E2.	People I care about stopped calling after learning I have HIV. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E3.	I have lost friends by telling them I have HIV. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
Subscale 2: Disclosure concerns		
E4.	Telling someone I have HIV is risky. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E5.	I work hard to keep my HIV a secret. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E6.	I am very careful who I tell that I have HIV. (Choose one)	

	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
Subscale 3: Concerns about public attitudes		
E7.	People with HIV are treated like outcasts. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E8.	Most people believe a person who has HIV is dirty. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E9.	Most people are uncomfortable around someone with HIV. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
Subscale 4: Negative self-image		
E10.	I feel guilty because I have HIV. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E11.	People's attitudes about HIV make me feel worse about myself. (Choose one)	
	1	Not at all
	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer
E12.	I feel I'm not as good a person as others because I have HIV. (Choose one)	
	1	Not at all

	2	Very little
	3	Somewhat
	4	Very much
	97	Don't Know
	98	Refuse to Answer

1. Reinius M, Wettergren L, Wiklander M, Svedhem V, Ekström AM, Eriksson LE. Development of a 12-item short version of the HIV stigma scale. Health and quality of life outcomes. 2017;15(1):115.

ANNEXURE 8: SUPPLEMENTARY TABLE 2: OVERVIEW OF "OTHER REASONS" FOR DONATION BY HIV+/ARV+ DONORS

Donation motivation	**Summary**
Donation as a blood management technique	This was nearly always a secondary motivation and often revolved around using donation to address interviewees' perceptions that they had "too much" blood (2202M, 4408F, 3304F, 4405F).
Donation as HIV status confirmation	In two cases (3307F, 4407F) this was secondary to interviewees' primary motivation of avoiding HIV-status disclosure or related suspicion in the donation context. Mentions of status confirmation were brief and seemed almost incidental: "I just wanted to take a blood – to see if it's safe to give to another person" (3307F), "I think [the year of the SQD] was the year I found out I'm HIV. Now I was looking for different result. But unfortunately, I got the same result" (4407F). In the other case, an interviewee, cited "to know if [SANBS] will see that this blood is HIV positive or what" (4408F) as her primary motivation. Despite having been diagnosed with HIV in 2009, and asserting that HIV had no impact on her, this interviewee still seemed somewhat confused about how she could have gotten the virus.
Belief that SANBS and the HIV service within the public health system were connected/could share clinical information	One interviewee (1103F) framed this as her primary motivation for donation; she also believed PLWH could donate for other PLWH.

ANNEXURE 9: CODE BOOK FOR THE THEMATIC ANALYSIS OF THE QUALITATIVE INTERVIEWS

Id	Parent Id	Title	Description
1		ACQU	Acquaintances: any time the interviewee talks about people with whom s/he has some kind of ongoing exposure to, but would not consider friends, lovers, family, etc. Examples: co-workers, classmates, people who live in their neighbourhood, etc. Will often be applied with another code, as participants will talk about other people in conjunction with certain activities, knowledge, desires, etc.
2		BLOOD BEL	Blood-related beliefs: Any time interviewees discuss their ideas about blood itself. This could be different kinds of blood (blood types, "strong" vs. "weak" blood), blood-related health beliefs (e.g., donation as a remedy for headaches caused by having too much blood), etc.
3		DISCUSS	Any time you would like to talk about a segment with the team. For example, if you think we should apply a new code, you've applied a code and you're not sure about it, or you just have a question.
4		DON ELIGIB	Donor Eligibility: any time the interviewee talks about his/her thoughts about or understanding of eligibility criteria for donation, even if incorrect or the "talk" is simply to say s/he doesn't know.
5		DON EXP	Donation Experience: any time the interviewee talks about his/her experience(s) donating blood. These segments may be long. Do not apply to general/hypothetical examples.
6		DON GUILT	Guilt around Donation: Any time the interviewee talks about feeling guilty for having donated blood, whether or not s/he was aware of eligibility criteria at the time of donation or not.
7		DON HC	Honesty Card: any time the interviewee talks about the "Honesty Card" donors should receive after donation, even if the "talk" is just to say s/he didn't get it. Will often be co-coded with (or inside a segment coded) DON EXP.
8		DON HESITAT	Hesitation around Donation: to ALL interviewee responses to the question "Was there ever a time you thought about not going through with the donation?" regardless of the answer. Also apply to any mention of hesitation or ambivalence about donation.
9		DON HIV 4 HIV	Donation of HIV+ blood for other PLWHIV: Apply this code any time the interviewee talks about the idea/belief that HIV+ recipients might be able to use blood given by an HIV+ donor
10		DON INTERV	Donation Screening interview: any time the interviewee talks about specifically reviewing answers to the questionnaire or having a one-on-one conversation with SANBS personnel prior to phlebotomy, even if the "talk" is just to say it didn't happen. Will often be co-coded with (or inside a segment coded) DON EXP.
11		DON LOC	Donation Location: any time the interviewee talks about the place where s/he donated: mobile vs. fixed, description of physical set-up, people present, etc. Will often be co-coded with (or inside a segment coded) DON EXP.
12		DON MOTIV	Donation Motivation: any time the interviewee talks about reasons why s/he donated. Will often be co-coded with (or inside a segment coded) DON EXP. Do not apply to general/hypothetical examples.
13		DON QUEST	Donation Questionnaire: any time the interviewee talks about the donor history questionnaire that should be completed prior to donation, even if the "talk" is just to say s/he didn't fill it out. Will often be co-coded with (or inside a segment coded) DON EXP.

14		DON TALK	Talk about donation to others: Any time the interviewee shares having discussed donation with other people, whether this is in response to the question on the interview guide or otherwise (e.g., with a physician, with a friend, with a SANBS worker ahead of the time ppt went to donate)
15		EMP/ECON SIT	Employment or Economic Situation: any time the interviewee talks about paid work (or lack of paid work), finances/money, material goods, being supported by or supporting someone else.
16		FAM/FRIENDS	Family or Friends: any time the interviewee talks about people to whom s/he is related (by blood, marriage, or other type of kinship), or who are friends
17		HIV ATT	HIV Attitudes: Any time the interviewee talks explicitly about her/his conception/the nature of HIV. Examples include: HIV as a death sentence, HIV as "just another illness," HIV as something the interviewee ignores/is rarely aware of, HIV as life-changing, etc.
18		HIV BIOMED	HIV Biomedical measures: any time the interviewee talks about ways HIV is typically assessed in medical contexts, like CD4 count, Viral Load (including "undetectable" viral load), etc., even if the ppt is misusing those terms/concepts
19		HIV CARE	HIV care: any time the interviewee talks about the care s/he receives for HIV in a clinical context. Do not apply for "caring" done by family/friends, for example. May often be applied with HIV Tx/ARVs
20	19	DIS/SATISFACTION	Any time interviewees discuss being satisfied or dissatisfied with the HIV care they receive.
21		HIV DISC	HIV Disclosure: any time the interviewee talks revealing or not revealing his/her HIV status, including to whom, for what reasons, why not, etc. May also be applied if interviewee talks about keeping HIV status "secret" or "private" (in that case it would be co-coded with PRIV).
22		HIV Dx	HIV Diagnosis: any time the interviewee talks about the circumstances or context of receiving her/his diagnosis with HIV.
23		HIV EMO	HIV Emotional Reaction: any time the interviewee talks about how s/he feels about having HIV, or about others' reactions to his/her status. This could be immediately in response to the diagnosis, or later on (including currently). Also, can use for things participant did to adjust to, accept, deny, or ignore HIV.
24		HIV LANG	Language around HIV: apply to interviewee talk that is about or references HIV in ways that suggest stigma or an attempt to distance oneself from the virus. This can apply to text that does not necessarily rise to the level of applying the STIGMA/SHAME code.
25		HIV PREV	HIV Prevention: Any time the interviewee talks about specific ways s/he has (or has not) attempted to prevent HIV transmission. This will most often be condom use or avoiding (penetrative) sex, but could also include serosorting (e.g., only having sex with partners of the same HIV status—that is, positive with positive or negative with presumed negative), not breastfeeding, taking PrEP, or only engaging in sexual interaction when his/her viral load is undetectable. Do not apply to general discussions of HIV prevention.
26		HIV SELF-CARE	Any time the interviewee talks about things s/he does outside of engagement in clinical care to maintain health after being diagnosed with HIV. Examples include eating or not eating certain foods, avoiding alcohol, getting exercise or sufficient sleep, engaging in meditation, support groups, etc.

27		HIV SIL	HIV Silences: Any time the interviewee talks about times or situations when HIV is not discussed or cannot be talked about, especially if the people who would be involved are aware of ppt's status. The idea here is that HIV in these interactions is taboo, ignored, the absence a kind of presence itself, a weight that the ppt carries alone.
28		HIV TEST	HIV testing: Any time the interviewee talks about specific instances of getting tested for HIV, whether their own testing or that of a partner. Should also be applied to talk about interviewee or partner refusing, avoiding, or failing to seek an HIV test when appropriate or necessary.
29		HIV TRANS	HIV Transmissibility: any time the interviewee talks about his/her understanding of how easy or difficult it would be to transmit HIV from one person to another (in general and in their specific case).
30		HIV Tx/ARVs	HIV Treatment or ARVs: any time the interviewee talks about when and how s/he began taking ARVs, the experience of taking (or stopping) ARVs, general feelings or effects of ARVs, etc.
31		HOBBIES	any time the interviewee talks about activities or pursuits on which s/he spends time regularly.
32		INTERESTING	Use this code to tag any excerpt that is interesting and you don't want to lose it, but there isn't an existing code that applies. Also apply the DISCUSS code so we can look at these together as a team.
33		NON-W MED	Non-Western medicine: any time the interviewee talks about any treatment s/he uses for HIV that is outside of that provided by the clinic. Also apply this code to all responses that indicate interviewee does NOT use non-Western medicine.
34		OTHER DON	Other Donation: any time the interviewee talks about a donation that is NOT the study-qualifying donation. Will often be applied with other codes.
35		OTHER PRE-DON PROC	Other pre-donation screening procedures: Any time the interviewee talks about pre-donation screening procedures OTHER THAN the questionnaire and specifically reviewing answers to the questionnaire or having a one-on-one conversation with SANBS personnel prior to phlebotomy (those themes have their own codes). This includes BP, weight, and iron check. OTH PRE-DON PROC Will often be co-coded with (or inside a segment coded) DON EXP.
36		PARTNERS	Romantic/sexual partners: any time the interviewee talks about a partner with whom s/he has an emotionally intimate (on a more than platonic level) or sexual relationship
37		PEER PRESS	Peer pressure/influence of others: any time the interviewee talks about other people (not necessarily just peers) playing an influential role in his/her decision-making, whether this influence comes from explicit statements/actions of the other person ("C'mon, everyone in our group is donating; don't be the only one who doesn't help!") or is more subtle. Can be applied in context of HIV testing, blood donation, ARV (non-)adherence, and other domains. Be aware, however, how this might be different from perceived stigma, in which the other person doesn't have to do or say anything at all but the interviewee still takes (or avoids) action based on his/her thought/assumption of being stigmatized.
38		POST-DON ACT	Post-Donation Action: Any time the interviewee talks about actions s/he took regarding the donation after leaving the donation location (e.g., calling SANBS, inquiring about donation

			eligibility for PLWHIV, etc.), as well as any actions taken by SANBS (e.g., reaching out to the interviewee, counselling, etc.)
39		PPT DESC	Participant description: to all interviewee self-description in terms of what s/he likes, dislikes, what kind of person s/he is, etc.
40		PRIV	Any time interviewee talks about privacy (including lack of privacy, wish for privacy, etc.), especially around donation. May also be applied if interviewee talks about keeping HIV status "private" (in that case it would be co-coded with HIV DISC).
41		QQ/GEMS	Quotable quote/Gems: quotes that are so good/so clearly illustrate an important point or trend in the data that they are obvious candidates for use in publications
42		RECS	Recommendations: any time the interviewee talks about what SANBS should do to increase clarity of communication around donation eligibility, help PLWHIV avoid donating blood, etc.
43		RELAT DYN	RELATIONSHIP DYNAMICS: Apply to talk about interviewee's relationship with significant people s/he is close with, and the functioning and impact of that relationship. This will most often be intimate/romantic partners, but could also be family members or even roommates in some cases. The point is to capture not just that a relationships exists (e.g., "I have a boyfriend") but the way it works in practice (e.g., "I have a boyfriend and we fight all the time, but he accepts my HIV"), and potentially how that shapes the rest of an interviewee's life.
44		RELIG	Religion/spirituality: any time interviewee talks about religion, spirituality, church, etc. Do not apply to mentions of God that seem purely cultural/casual (e.g., "Thank God that she didn't see me in the clinic") but DO apply when God is mentioned in a way that seems spiritually meaningful (e.g., "I thank God every day for the medicine that keeps me alive.").
45		SANBS TEST	SANBS Testing: any time the interviewee talks about his/her understanding of testing the blood centres do on donations, even if incorrect or the "talk" is simply to say s/he doesn't know.
46		SOC SUPPORT	Social Support: apply this code to talk about giving / receiving (or not giving/ receiving) non-monetary and non-material help or assistance, especially emotional or psychological.
47		SQD	Study Qualifying Donation: any time the interviewee talks about his/her "study qualifying donation." Will almost always be applied together with other codes.
48		STIGMA/ SHAME	any time the interviewee talks about feeling shame around HIV status, having experienced discrimination due to HIV status, feeling that others (may) treat people who are living with HIV differently than those they assume to be HIV-negative. Note: Can also be applied if the coder feels the interviewee is saying something motivated by stigma or shame, even if the interviewee him/herself does not recognize it. In that case, use the code if there is at least one explicit indication you can point to that stigma (and not some other reason) is at the root of the utterance, and also apply the code "Discuss." Important: If other sources of stigma/shame (i.e., other than HIV) surface in the data set, please code with "Discuss" and be sure to call team's attention to them.