Project 4 – Does pre-donation behavioral deferral increase the safety of the blood supply?

Rationale: While it is well-accepted that deferrals as part of the "layers of safety" concept increase the safety of the blood supply, studies with sufficiently large sample size to quantify HIV infection and other infectious marker rates in deferred donors are lacking. Evidence in support of increased safety is frequently inferred from studies conducted in other health care settings. For example, a small hospital-based case control study conducted in Brazil examined the association between infectious markers and body tattoos. Even though tattoos are not used as a criteria to determine blood donor eligibility in Brazil, having a tattoo was associated with HCV (Odds Ratio (OR) 6.4, 95% CI 1.3-31.8), and also with having at least one positive infectious marker (OR 2.1, 95% CI 1.1-3.8).(1) Significant associations were not independently observed for HIV, HBV, syphilis or Chagas. The authors reported an overall sensitivity of 11% and specificity of 97% for the presence of a tattoo as indicator of having HIV, HCV, HBV, or syphilis infection. The researchers then estimated the impact on blood donor selection and disease marker testing using the results from their hospital-based case control study. However, the assumptions such as disease marker prevalence of as much as 15% in donors who are deferred for tattoos and a prevalence of 4% of the potential donor base having a tattoo (2) do not represent current temporary deferrals in Brazil and do not address the most common behavior-related deferrals. A more detailed and targeted assessment of the value of relevant deferrals could be used to help inform blood donation policies in Brazil.

Background

In USA, current HIV prevalence is so low that studies of deferral are impractical due to the large sample size required. The ten-fold higher HIV prevalence in Brazil [1,2] allows us to design a hypothesis-based study whose results will be directly applicable to Brazil, and more broadly may be relevant for other countries considering the utility of their donor deferral policies. Although technically recommended by the Brazilian Government, lack of funds and the poor cost-benefit ratio of HIV nucleic acid testing (NAT) have delayed its implementation in this upper-middle income country. Plans to implement NAT are currently being developed. Even if NAT were to be implemented, the residual risk of HIV infection would still be substantially higher than in USA. Additional studies that assess recruitment and deferral are therefore essential in further reducing the risk of transfusion-transmitted HIV infection. The Brazil Ministry of Health recommends a number of HIV risk factor questions in the standard blood donor questionnaire, but the effectiveness of these questions are unknown. Only preliminary studies of the HIV risk factor profiles have been conducted among Brazilian blood donors.

Donor deferral is initiated either by the blood center based on information disclosed by prospective donors or by the donor through self-deferral. Either type of deferral occurs because of the belief that a donor's behavior, exposures, or history represents an increased risk to the safety of the blood supply. Some self-deferrals occur before a person presents to be a donor. This study does not address those self-deferrals. In addition the process for donor deferral could impact the number of deferrals. Two of the REDS-II Brazil centers (and HemoRio) use detailed codes to record specific deferrable behaviors. While the other center (HemoMinas) uses a very limited set of codes that are applied to all donors regardless of the specific deferrable behavior reported by the donor.

Data on the frequency of all deferrals and specifically for behaviors that could represent higher risk of infection were obtained from two years of the REDS-II Brazil donation database (Table 1). High risk behavioral deferrals are not captured in the same way at each of the REDS-II Brazil blood centers. The high risk behavior category is a combination of several behavior categories at the different blood centers. A high risk exposures category combining some or all of the specific codes now located in different REDS II deferral code categories of 403, 404, and 501, 503, and 600 was created. Internally this combined category is referred to as the 500c. Table 2 provides the specific codes captured in 500c that are

applied to donors who present to donate from the different centers. (A similar mapping for the Rio blood center has not been completed, but will be necessary before Rio can participate in the study.)

Table 1.Donor presentations and reasons for deferral in participating REDS-II Brazil blood centers 2007 – 2008.

	Sao Paulo (%)	Belo Horizonte (%)	Recife (%)	Rio de Janeiro (%)
Candidate Presentations	349,002	197,047	269,415	203,008
Total Donations	271,880 (77)	139,429 (71)	204,122 (76)	179,604 (78)
Total Deferrals	77,122 (22)	57,618 (29)	65,293 (24)	50,404 (22)
Deferrals				
Low hematocrit/hemoglobin	12,266 (16)	5,236 (9)	14,748 (23)	9,788 (19)
Blood Pressure/Pulse	6,618 (9)	5,025 (9)	7,924 (12)	2,296 (5)
Medical diagnoses	15,926 (20)	10,127 (18)	6,579 (10)	6,077 (12)
Higher risk behaviors	***7,129 (9)***	***17,295 (30)***	***6,243 (10)***	***8,227 (16)***
Unwell, colds, high temperature	3,101 (4)	2,680 (5)	4,884 (8)	2,270 (5)
Medication	4,398 (6)	2,499 (4)	3,310 (5)	2,667 (5)
Other infectious exposures	6,984 (9)	0	1,239 (2)	1,432 (3)
Couldn't wait, Changed Mind	2,282 (3)	5,126 (9)	3,628 (6)	2,013 (4)
Vaccination	4,673 (6)	994 (2)	1,298 (2)	4,064 (8)
Weight	1,299 (2)	898 (2)	817 (1)	836 (2)
Other Deferral	6,356 (8)	5,835 (10)	12,867 (20)	13,734 (27)

Table 2. REDS-II deferral grouping codes and individual blood center deferral codes for the deferred donor protocol.

REDS-II Deferral Category (Grouping codes)	Sao Paulo (Specific codes)	Belo Horizonte, Minas Gerais (Specific codes)	Recife, Pernambuco (Specific codes)	Rio de Janeiro (Specific Codes)
403 – HIV exposure	23G-Exchange drugs or money for sex		61-Sexual partner of HIV suspicious 62-Unsafe sex with heterosexual partner <12 months 73-Rape	069-Sexual partner of a HIV positive person*
404 – STD exposure	11-Syphilis 11A -Other STD	017-Self reported STD	39-STD exposure	036-STD exposure*
501 – High risk (includes high risk sexual partner)	08A- Sexual partner of hepatitis patient 17A- Sexual partner of blood recipient 23B1- Sexual partner actually in prison or in the past 23C3- Sexual partner of injection drug user 23C4- Sexual partner of not injection drug user 23D1- Bisexual partner 23E- High risk sexual relations= 6 or more sexual partners 23E1-sexual intercourse without a condom/casual one time partner. 23F- Sexual partner of prostitute 23F1 - Promiscuous sexual partner 23H- Contact with Infectious Disease Carrier 23K- High risk sexual partner 24- Professional activity of high risk (prostitute, men and women dancers, rent boy, male hustler, etc) 51- Sexual partner of hemodialysis patient 60- Sexual partner of organ/tissue receiver	012-Behavioral risk – TD* 013-Behavioral risk-PD** 067-Sexual promiscuity	100-Sexual partner to a HIV risk person 103-Prostitution 107-Sexual partner to a HIV positive 108-Sexual partner of prostitute <12 months 112-High Risk suspicious 74-Bisexual 75- Promiscuous 124- Sexual partner to a HTLV positive 132- Sexual partner to a HCV positive 88- Sexual partner of ex-inmate or convict	033-High risk sexual relations= 5 or more sexual partners** 063-Sexual partner of a blood recipient*
503 – Male who has sex with other males (MSM)	23D-Same sex sexual relation 50-bisexual		118-Homosexual contact, just once 77-MSM	034-MSM **
600 – Other deferral	23A- Came to blood bank to get blood tests/also HIV test 23C2-Drug user (not ID user) 78-Past Drug user (not ID user)	073-Illegal drug user	87- Came to blood bank to get blood tests/also HIV test 92-Drug user (not IDU)	067-HIV test seeker ** 054-Inhaled Drug* 104-Other drugs* (LSD, Ecstasy, etc) 035-Drug User (IDU)*

* Temporary Deferral ** Permanent Deferral

Preliminary Studies

While the previous studies by Nishioka and imply that deferred donors may have higher disease marker rates, to our knowledge there have been no Brazilian or studies in other countries focused on measuring actual disease marker rates in deferred donors that could then be used to estimate the positive predictive value of specific deferral categories. There are two studies that have addressed deferrals and disease markers in blood donors in Brazil.

In a previous study by Goncalez and colleagues[3] at FPS/HSP, prevalence of infectious disease markers were measured in a small sample of 238 deferred donors. 72 (31%) of those deferred were deferred because of self-acknowledged HIV-related risk behavior and 165 (69%) were deferred for other reasons (anemia being the most common cause, n=47). Deferred donors were more likely to have anti-HCV reactivity ($p=0.03$) and syphilis ($p=0.04$) but no statistical difference was found for HBV, HTLV-I, or Chagas disease. There was no anti-HIV reactivity within accepted or deferred blood donors in this study, likely due to the small sample size.

Almeida Neto and colleagues[4], also from FPS/HSP, conducted an analysis of HIV risk factors in deferred blood donors in 2004. In that study EIA reactive blood donors were recalled to the counseling center where another blood sample was taken for Western blot confirmatory testing. Donors were then notified and counseled regarding the results of this confirmatory testing, and referred to medical care if indicated. A risk factor questionnaire was administered to all returning donors as part of the counseling procedure. Controls blood donors were defined as being HIV EIA reactive but Western blot negative or indeterminate. The results showed gender-specific associations with HIV seropositivity confirmed by Western blot, men age 21 through 40 ($p=0.011$), male to male sex ($p<0.001$), having had more than two heterosexual partners in the past year ($p<0.001$), sex with a prostitute and sex with a promiscuous or HIV positive partner were also significantly associated with HIV. Among women, high school education ($p=0.03$) and multiple sexual partners ($p=0.02$) were significantly associated with HIV status. Although reported by few women, sex with an IDU, having a promiscuous or HIV positive partner were also significant risk factors. This preliminary study had sample size limitations, did not ask donors about motivations for donating, and was not able to control for volunteer (community) versus replacement (directed) donor status. The study also focused on behaviors in disease marker positive donors as opposed to disease marker prevalence in deferred donors.

Study Overview

The three blood centers that are participating in REDS-II Brazil are Fundaçao Pro Sangue/Hemocentro de Sao Paulo (FPS/HSP), HemoMinas in Belo Horizonte, Minas Gerais, and Hemope in Recife, Pernambuco. In addition HemoRio in Rio de Janeiro is participating in an HIV case control study and will participate in the deferral study because of the similarity in the study procedures between the HIV case control and deferral studies. The present study will expand upon the previous research by specifically assessing disease marker prevalence in deferred donors while also determining motivations for attempting to donate and whether additional risk factors are present using a short version of the audio-computer assisted interview (ACASI) developed for the HIV case control study. This study will focus on the safety impact of several sexual exposure category deferrals; multiple sexual partners, male to male sex, exchanging money or drugs for sex, sex with a partner who has HIV, etc.

Aim 1

Assess infectious disease marker prevalence in donors who are deferred for higher risk sexual and non-injection drug use behavior.

Hypothesis

1. The prevalence of infectious markers in donors who are deferred for sexual risk behaviors will be higher than those of accepted blood donors.

Aim 2

Determine if the deferral classification procedures and coding used by different blood centers in Brazil leads to a measurable difference in disease marker prevalence in deferred donors between those centers that use specific behavior codes and those centers that use non-specific deferral codes.

Hypothesis

2. The deferral assessment and coding processes used at different centers lead to different infectious maker prevalence in deferred presenting donors. Non-specific deferrals codes will have different predictive value than behavior-specific deferrals codes.

Study Design

Methods:

Deferred donors who agree to participate in this study will be asked to complete an audio computer assisted self interview (ACASI) questionnaire that measures two content areas 1) motivations for attempting to donate, 2) additional information on the deferral and other potentially undisclosed deferrable behaviors. A phlebotomy of approximately < 24 ml will be collected from the deferred donors and tested for the panel of infections currently screened for in Brazil (HIV, HCV, HBV, HTLV, syphilis, and *Trypanosoma cruzi*) using the same high-throughput laboratory reagents and procedures that are used to screen donations. Self-disclosed motivations and reasons for deferral will be reported. Comparison of deferred donor marker rates will be made to infectious marker testing of accepted donors with the same demographic characteristics captured in the available from the REDS-II Brazil donation database during the same time period of enrollment of the deferred donors in this study. Marker rates in deferred donors will also be compared between the blood centers that use detailed versus non-specific deferral codes.

Inclusion criteria: Subjects will be recruited for the study from those prospective donors who are deferred during the health history assessment in categories of infectious disease exposures related to sexual and/or non-injection drug use (Table 2).

Exclusion criteria:

- **a.** Prospective blood donors who refuse to participate
- **b.** Prospective blood donors who are not Portuguese literate
- **c.** Prospective donors who are deferred for reasons other than sexual or non-injection drug use behavior.

Deferred donors will be taken to a private location in the blood center. At that time the purpose of the study will be explained in more detail and written informed consent will be sought to allow for blood sample collection, disease marker testing, collection of interview data, and linking interview and testing data to operational blood center records.

Data Sources:

Original data collection as part of study protocol:

a) Laboratory results for blood samples collected from deferred donors and accepted donors (HIV, HCV, and other infectious disease markers).
b) Self-administered structured ACASI questionnaire.

Linkage to existing operational data:

c) Health history questionnaire responses. The content for this questionnaire is issued by the Brazilian Minister of Health and has been designed primarily to measure risk behavior for STDs. The health history form is tailored to the procedures at each center, but covers the same general content.

Study Procedures:

This study will take place only at the primary donation for each blood center. At 4 Brazilian blood centers (Sao Paulo, Belo Horizonte, Recife, and Rio), the clinical screening process and eligibility assessment for blood donation, is usually performed by a physician or nurse. All members of the interview staff at the main center for each organization will complete the standard eligibility assessment. A work-aid will be developed that is specific for each blood center indicating which deferrals for that center trigger recruitment for participation in the study. Donors who report one of these deferrable behaviors will be approached about possible participation in the deferred donor study. These donors will be directed to meet with REDS-II research staff for an explanation of and recruitment into the study in a private setting. If the deferred donor (subject) agrees to participate and provides informed consent, the subject will have a phlebotomy of 24 mL of whole blood drawn. The study subject will then complete the risk factor and motivations interview using the ACASI application. Blood samples will be submitted for infectious marker screening.

The blood samples will receive a research study label, but will be routed through the blood bank laboratory for routine testing by standard procedures at each blood center. This includes supplemental or confirmatory testing to be conducted at each local blood center using standard procedures specific to each center. The collected samples from all deferred subjects from the 4 blood centers will be kept for potential additional infectious diseases markers tests in a repository at Fundacao Pro-Sangue/Hemocentro de Sao Paulo. Each center is required to keep a donor sample for at least 6 months following donation, we will use the same process for this study and at the end of study accrual the residual volume of blood remaining following testing will be transferred to the Sao Paulo repository.

Deferred donors who confirmed positive by initial or supplemental testing for any infection will be informed of the results, re-tested and counseled in accord with Brazilian and local regulations, and blood center procedures.

Results: One of three different letters will be sent to the participants first thanking them for participating in the study and then notifying them of their testing results: 1) Negative letter: to the subjects with negative serology for all the markers tested, 2) Positive letter: to the subjects that tested positive or inconclusive for any of the markers, and 3) Laboratory issues: to the subjects that for technical reasons we were unable to perform all tests.

The reason for deferral as recorded in operational records will be obtained from the donor history form, and transferred together with the questionnaire to the coordinating center. All health history and operations data come from computerized records and are part of IT system at each center.

IT specialists will merge these three data sources into one file using a new coded number that will link the health history data, the ACASI interview, and the results of the infectious marker testing. Once the new file is created and data integrity checks have been performed, donor identifiers will be removed before

providing the data set for analysis. The file will retain a coded number, but the USA researchers will not have access to the code. Only if necessary, information systems specialists can update information. Researchers will never have access to this code and patient name. Data will be transferred to the coordination center using procedures that have previously been defined for the HIV case control study.

Measures:

Outcome variables. The primary variables are the categories of infectious disease exposure deferrals related to sexual and non-injection drug use. Donors will be classified as having been deferred for the reason recorded on operational (donor health history) records. The operational records classification will be compared to deferrable behaviors that each donor reporting during the ACASI.

Infectious maker prevalence will be estimated overall and by infection for the deferrable behaviors.

Predictor variables and covariates. The blood center and donor status (volunteer v. replacement) are the primary independent variables. Among other independent variables is donation type (first time *vs*. lapsed vs. repeat). At each blood bank, repeat blood donors will be defined as those who donated previously at least once in the last 12 months, and lapsed blood donors will be defined as donors who donated previously, but not in the past 12 months. Further variables include: demographic characteristics (age, gender, salary, education, socio-economic status), site of donation, use of CUE, etc.

We expect that approximately 50% of the people who are approached will agree to participate in the study. If the recruitment period lasts 6 months, the total number of deferred donor participants should be over 4800 (Table 3).

Table 3. Expected deferred donor participation by center.

Estimated Participation	Sao Paulo	Belo Horizonte	Recife	Rio de Janeiro	Total
50%	891	2161	780	1028	4860
33%	588	1426	515	679	3208

Sample size:

The expected sample size is 4860 deferred donors. Roughly 75% of the sample will be FT donors who are deferred, and the other 25% will be RPT donors who are deferred. The disease marker rates among these two subgroups of deferred donors will be compared to the known disease marker rates of corresponding ‘accepted’ donors. An accepted donor is a donor who successfully completes a donation. Screening tests on these donations determines disease marker rates among donors. These rates have been compiled in the 2008 REDS-II Brazil donation database and are shown in Table 4 for the three markers of primary interest.

Since FT deferred donors constitute the majority of the sample and since the marker rates are presumed higher in FT donors, a difference in marker prevalence between FT deferred donors and FT ‘accepted’ donors will be easier to detect (as exhibited by the smaller Odds Ratios in Table 4). With the estimated sample size and 80% power we will be able to detect an odds ratio of 1.3 for the combined infectious marker prevalence in first time deferred donors compared to first time accepted donors when all deferred donors are combined together during analysis.

Aim 2 will compare the marker prevalence in the center that use non-specific deferral codes (i.e. Belo Horizonte) to marker prevalence in centers that use specific deferral codes (i.e. all other centers). Table 5

shows the Odds Ratios (i.e. odds of positive marker in Belo Horizonte compared to odds of positive marker in other centers) that can be detected in the minimum expected sample size. Since the prevalence depend greatly on FT/RPT status of the deferred donor, the Odds Ratios will be tested stratified by FT/RPT status (hence the latter two columns of Table 5 are most relevant). With the estimated sample size and 80% power we will be able to detect an odds ratio of 1.6 for the combined infectious marker prevalence in first time deferred donors compared to first time accepted donors when all deferred donors are combined together during analysis for the Belo Horizonte blood center.

Table 4. Odds Ratios pertinent to Aim 1 that can be detected with 80% Power.

	Marker prevalence			Marker prevalence		
Infection	FT 'accepted' donors *	FT deferred donors	Odds Ratio	RPT 'accepted' donors *	RPT deferred donors	Odds Ratio
HIV	0.41%	0.74	1.8	0.19%	0.65%	3.4
HBV	0.26%	0.53%	2.0	0.02%	0.25%	12.5
syphilis	1.5%	2.1%	1.4	0.39%	1.03%	2.6
combined**	2.6%	3.4%	1.3	0.63%	1.31%	2.1

* Confirmatory marker prevalence of 'accepted' donors are derived from 2007-2008 REDS-II Brazil donation database.
** Combined prevalence of HBV, HCV, HIV, HTLV, and Syphilis

Table 5 Odds Ratios pertinent to Aim 2 that can be detected with 80% Power.

Infection		Overall		FT		RPT
HIV		2.3		2.5		4.4
HBV		2.7		2.9		7.5
syphilis		1.8		1.8		3.5
combined		1.6		1.6		3.1

Data Analysis:

Aim 1

Disease marker rates (as a summary measure) and by each pathogen will be reported for the deferred donors. The prevalence of disease markers will be compared to those of demographically (age, gender, donor type and status) similar eligible donors captured in the REDS-II Brazil database during the time the same time period as this study.

Data Analysis: We will estimate the positive predictive value the behavioral deferrals using infectious marker test results as the relevant "gold standard". The positive predictive value of a deferral is dependent on the prevalence of the deferral in the presenting donor population and the specificity of the tests used to detect each disease marker. Within deferral categories we will determine the disease marker positive test yield for all disease markers combined.

Multivariate modeling of the predictors of disease marker positive donors who are deferred among all donors who are deferred for higher risk behaviors will be conducted.

Expected Results: We expect to find a higher seroprevalence of HIV, HBV, and syphilis among donors with these deferrals when compared to demographically similar accepted blood donors. We do not expect

that marker rates for HCV and HTLV will be higher because infection by sexual contact is less common for these viruses.

Aim 2

Compare the prevalence of HIV, HBV, HCV, HTLV, and syphilis for donors who are deferred in a the center that uses non-specific deferral codes to the other blood centers that use specific deferral codes using t-test or other appropriate statistical tests.

Expected Results: The rate of infectious markers will be different (and likely lower) for the blood center that uses non-specific deferral codes than for the centers that use specific codes.

Alternative Approaches:

Deferred donors will not be compensated for participating in the study. However, we expect that deferred donors will be interested in the study question, will choose to participate, and that participation rates will be high. If deferred donors do not participate in sufficient numbers to achieve study sample size requirements in a six-month study period, we will consider only including multiple sex partner deferrals in the study. This deferral is expected to be the most common and as a result the easiest to evaluate and also the potentially most important deferral that could be modified if there is no evidence of increased risk of infectious markers in donors who are deferred due to having multiple sexual partners. An evaluation of participation will be necessary within 2 months of initiation the study to determine if enrollment targets will be met. The two month evaluation will allow us to alter the deferral inclusion criteria if necessary, or to develop strategies to increase subjects' enrollment.

Limitations:

Deferral definition: Am important potential limitation is the difference in deferral categories at the blood centers. It will be necessary to develop center-specific work aids to identify deferred donors that should be approached for study participation. However, this study also seeks to understand how the different deferral procedures impact the rates of disease markers at blood centers and will use the differences to further address relevant policy questions in Brazil.

Human Subjects Considerations:

Positive serological markers: Blood samples will be submitted for infectious marker screening. If the screening results for any marker are positive we will use the same operational procedures that each the blood center uses for the regular blood donors who have reactive results, including additional testing, notification, counseling and permanent donor deferral.

Risks of this protocol include phlebotomy, testing for HIV and other ID markers, and a questionnaire inquiring in detail about HIV risk behaviors such as sexual practice or injection drug use. As with any study, there is a risk of lost confidentiality. There are no benefits to the subjects beyond possible early diagnosis and treatable HIV, hepatitis and other infections. There is a probable benefit to Brazilian society in knowledge that may be useful in increasing blood safety. We shall minimize the risks to the subjects by using trained phlebotomist, by providing test results and counseling using trained blood bank staff.

Brazil doesn't have a disqualified donor registry. A person at risk might donate blood in many different blood banks, therefore, subjects found to be marker positive will be told they cannot donate blood in the future and will be placed on a list of permanent deferral donors at blood bank where he/she has donate at the last time. However, this information cannot be communicated to other blood centers.

The collected samples from all deferred subjects from the 4 blood centers will be kept, for potential additional infectious diseases markers tests, in a repository at Fundacao Pro-Sangue/Hemocentro de Sao Paulo. Study subjects will provide consent for placing the residual sample volumes into the repository for potential future testing by indicating their willingness to have sample kept in the repository.

The study protocol will be approved by the IRB's at each Brazilian center, the Brazilian government IRB, and the UCSF Committee on Human Research; written informed consent will be obtained from all subjects prior to enrollment. The study will also require Office of Management and Budget (OMB) approval.

References:

1. de Nishioka SA, Gyorkos TW, Joseph L, Collet JP, MacLean JD. Tattooing and transfusion-transmitted diseases in Brazil: a hospital-based cross-sectional matched study. European journal of epidemiology. 2003;18(5):441-9.

2. Nishioka SA, Gyorkos TW, MacLean JD. Tattoos and transfusion-transmitted disease risk: implications for the screening of blood donors in Brazil. BrazJInfectDis. 2002 08;6(4):172-80.

3. Goncalez TT, Sabino EC, Murphy El, Chen S, Chamone DA, McFarland W. (2006) Human immunodeficiency virus test-seeking motivation in blood donors, Sao Paulo, Brazil.*Vox Sang*, Apr;90 (3):170-6

4. de Almeida Neto C, McFarland W, Murphy EL, Chen S, Nogueira FA, Mendrone A Jr, Salles NA, Chamone DA, Sabino EC,(2007) Risk factors for human immunodeficiency virus infection among blood donors in Sao Paulo, Brazil, and their relevance to current donor deferral criteria, Transfusion, 2007 Apr;47(4):608-14.