Supporting Statement B for

Evaluation of Risk Factors Associated with HIV, HBV and HCV in Chinese Donors (NHLBI)

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B. Collection of Information Employing Statistical Methods

B.1. Respondent Universe and Sampling Methods

Sampling Method for HIV Risk Factor Protocol:

All five blood centers will participate in this study. Cases will be donors with confirmed anti-HIV antibody reactivity. The control group will include randomly-selected HIV non reactive donors matched by blood center and fixed versus mobile donation sites.

All donors with anti-HIV results confirmed by local C-CDC laboratories will be eligible as Cases for this study. During C-CDC's routine notification/counseling meeting with a donor with confirmed anti-HIV results, a trained staff member from C-CDC (or the blood center) will explain the purpose and procedures of this study and administer an informed consent form. Consented donors will be interviewed by trained staff using the RFQ. The location for this activity can be either at the local C-CDC office or blood center. The REDS-II China Data Center will select a random group of donors with negative infectious disease test results as Controls for this study. Controls will be matched to the Cases by blood center and donation month. Blood Centers will contact potential Controls by phone and/or mail, inviting them to come back to participate in this study. Controls will be consented and interviewed using the same RFQ by C-CDC or blood center staff, either at the local C-CDC or blood center.

Sampling Method for HBV/HCV Risk Factor Protocol:

Three groups of donors will be included in the protocol:

- "HBV Group": HBV (HBsAg) positive donors either from prescreening (rapid testing) or routine screening testing. Confirmatory testing for HBV will be done for these donors.
- "HVC Group": HCV (anti-HCV) positive donors from routine screening testing (blood centers do not do prescreening rapid testing for anti-HCV). Confirmatory testing for HCV will be done for these donors.

- "Control Group": Donors with negative results for all prescreening and routine screening tests. No additional testing is done for these donors.

Using confirmatory testing results for HBV and HCV respectively, the REDS-II China Coordinating Center will generate a list of cases for each blood. A list of controls (randomly selected and matched by fixed versus mobile collection site) will also be provided to each blood center.

Sample Size Calculations for HIV Risk factor Protocol:

Table B.1.1 below shows the sample size that would provide 80% power to detect ORs of 1.5, 2.0 or 3.0 with an alpha of 0.05 for various risk factor prevalence values (from 1% to 10%). The OR compare the odds of having a risk factor between seropositive and seronegative donors.

Table B.1.1: Sample size required to detect with 80% power for a given OR with alpha of 0.05

	Risk Factor Prevalence in Controls				
OR	0.01	0.02	0.03	0.05	0.1
1.5	8,075	4,100	2,800	1,725	930
2	2,400	1,225	**830**	**525**	290
3	810	420	285	180	103

For this study, using the estimate of identifying about 200 HIV confirmed seropositive donors per year at the five blood centers, enrolling cases and controls over a 30 month period (10/08 to 3/11) will allow us to identify about 500 HIV confirmed seropositive donors. Assuming a 70% response rate from donors, we anticipate that about 350 donors will be enrolled in this group which would permit detection

of an OR of about 2.0 with a risk factor prevalence of about 6.5%. We are assuming a comparable response rate for controls, therefore we plan to approach approximately 1000 controls to enroll 700.

Sample Size Calculations for HBV/HCV Risk factor Protocol:

Table B.1.2 shows the sample size that would provide 80% power to detect ORs of 1.5, 2.0 or 3.0 with an alpha of 0.05 for varying prevalence of risk factors (from 1% to 10%) in the control group. The ORs compare the odds of having a risk factor between seropositive and seronegative donors. Sample sizes have been adjusted for multiple comparisons. We will use the same control group for both HBV and HCV cases. The 'optimal' ratio is √2 (1.41) controls to one HBV seropositive and one HCV seropositive. Based on our prediction that about 30% of HBsAg or anti-HCV screening test reactive samples will have confirmed reactive results, and about 70% donors will respond to our survey, we plan to do supplemental testing on 4,000 screening HBsAg reactive donors and 4,000 screening anti-HCV reactive donors with the goal of enrolling 830 donors into each study group. Considering the same 70% response rate, we plan to approach 1,700 donors with the goal of enrolling 1,170 as controls.

Table B.1.2: Sample size required to detect with 80% power for a given OR with alpha of 0.05

			Risk Factor Prevalence in Controls				
OR		Group	0.01	0.02	0.03	0.05	0.1
	1.5	Seropositive HBV	8,075	4,100	2,800	1,725	930
		Seropositive HCV	8,075	4,100	2,800	1,725	930
		Control	11,386	5,781	3,948	2,432	1,311
		Total	27,536	13,981	9,548	5,882	3,171

2	Seropositive HBV	2,400	1,225	**830**	525	290
	Seropositive HCV	2,400	1,225	**830**	525	290
	Control	3,384	1,727	**1,170**	740	409
	Total	8,184	4,177	**2,830**	1,790	989
3	Seropositive HBV	810	420	285	180	103
	Seropositive HCV	810	420	285	180	103
	Control	1,142	592	402	254	145
	Total	2,762	1,432	972	614	351

B.2. Procedures for the Collection of Information

B.2.1. Questionnaire

Questionnaires will be administered only once to all subjects in a paper format for both the HIV and HBV/HCV studies. Donors participating in the HBV/HCV study will also have an option to complete a web based survey. The questionnaire will collect general demographic and risk factor information pertinent to all three viruses (HIV, HCV, and HBV).

HIV Risk factor Protocol:

Cases: During C-CDC's routine notification/counseling meeting with a donor with confirmed anti-HIV results, a trained staff member from C-CDC (or the blood center) will explain the purpose and procedures of this study and administer an informed consent form. Consented donors will be interviewed by trained staff using the RFQ. The location for this activity can be either at the local C-CDC office or blood center.

Controls: Blood Centers will contact potential Controls by phone and/or mail, inviting them to come back to participate in this study. Controls will be consented and interviewed using the same RFQ by C-CDC or blood center staff, either at the local C-CDC or blood center.

HBV/HCV Risk factor Protocol:

The paper questionnaire will be sent to all participants along with instructions for completing the questionnaire online as an alternative way of participating.

B.2.2 Data Analysis

HIV confirmatory results and results from the risk factor interviews will be entered into the REDS-II China database on a routine basis (weekly) using double key entry. Data will be checked for logical errors and consistency.

We propose separate case-control analyses to examine, in turn, the risk factors associated with each virus. Case/control status will be the main outcome variable (e.g., HIV-positive vs. control), whereas the various risk factors will be the independent variables of main interest. Other variables such as age, gender, ethnicity, first-time/repeat status could be potential confounders or effect modifiers and will be evaluated. Frequency tables and associated Chi-square tests (or Exact tests for small sample size) will be used to review potential associations. We will calculate odds ratios (OR) with 95% CI using logistic regression analysis to compare the odds of seropositive donors having a risk factor compared to seronegative donors. Models will be constructed both unadjusted and adjusted for factors that may affect the association between the risk factor and the infection of interest. We will also determine what final set of independent risk factors appear to be associated with the infection of interest by building a multivariable model that includes as independent variables all risk factors that are independently associated with each infection. We will probably use a stepwise selection procedure to build this multivariable model and will examine interactions between independent variables, as appropriate.

B.3. Methods to Maximize Response Rates and Deal with Non-response

A trained staff member from C-CDC (or the blood center) will explain the purpose and procedures of this study and administer an informed consent form. All consented donors for HIV

study will be interviewed by trained staff using the RFQ. A web based survey option is also given to the HBV/HCV study donors to improve the response rate.

B.4. Test of Procedures

We have conducted focus groups discussions and cognitive testing to improve the potential reliability of responses.

B.5. Individuals Consulted on Statistical Aspects and Individuals Collecting and/or Analyzing Data

Individuals consulted include biostatisticians on statistical aspects of the study design; the blood centers researchers responsible for enrollment, administering questionnaires, and collection of samples; and the CC staff for protocol development, study monitoring, and data management. Data analysis will be performed by the analytic staff at the CC that includes epidemiologists and biostatisticians, with assistance and oversight provided by the REDS Steering Committee (see Attachment 3.3 for a complete list of Steering Committee members.)