Supporting Statement (0925-NEW)

RETROVIRUS EPIDEMIOLOGY DONOR STUDY–II (REDS–II)

THE REDS-II DONOR IRON STUDY:PREDICTING HEMOGLOBIN DEFERRAL AND DEVELOPMENT OF IRON DEPLETION IN BLOOD DONORS

RETROVIRUS EPIDEMIOLOGY DONOR STUDY – II (REDS – II)

THE REDS-II DONOR IRON STUDY: PREDICTING HEMOGLOBIN DEFERRAL AND DEVELOPMENT OF IRON DEPLETION IN BLOOD DONORS

Sponsored by:

The National Heart, Lung, and Blood Institute

Transfusion Medicine Branch

National Institute of Health

OMB Submission

Project Officer/ICD Contact: George J. Nemo, Ph.D.

Transfusion Medicine Branch

Division of Blood Diseases and Resources

National Heart, Lung, and Blood Institute

Two Rockledge Center

Suite 361

6700 Rockledge Drive

Bethesda, MD 20892

Phone: (301) 435-0075

Fax: (301) 480-0868

Email: nemog@nih.gov

TABLE OF CONTENTS

SUPPORTING STATEMENT 1
Introduction and Summary 1
A. Justification 3
A.1. Circumstances Making the Collection of Information Necessary 3
Introduction 3
A.2. Purpose and use of the information 4
A.3. Use of Information Technology and Burden Reduction 5
A.4. Efforts to Identify Duplication and Use of Similar Information 6
A.5. Impact on Small Businesses or Other Small Entities 6
A.6. Consequences of Collecting the Information at a Chosen Frequency 6
A.7. Special Circumstances Relating to the Guidelines of 5 CFR 1320.5 6
A.8. Comments in Response to the Federal Register Notice and Efforts to Consult Outside Agency 7
A.9. Explanation of Any Payment or Gifts to Respondents 7
A.10. Assurance of Confidentiality Provided to Respondents 7
A.11. Justification for Sensitive Questions 7
A.12. Estimates of Hour Burden Including Annualized Hourly Costs 9
A.13. Estimates of Other Total Annual Cost Burden to Respondents or Record Keepers10
A.14. Annualized Cost to the Federal Government 10
A.15. Explanation for Program Changes or Adjustments 10
A.16. Plans for Tabulation and Publication and Project Time Schedule 10
A.17. Reason(s) Display of OMB Expiration Date is Inappropriate 12
A.18. Exceptions to Certification for Paperwork Reduction Act Submissions 12
B. Collection of Information Employing Statistical Methods 12
B.1. Respondent Universe and Sampling Methods 12
B.1.1. Donor Universe 12
B.1.2. Donor Sample 13

B.1.3. Site selection by Centers 14
B.2. Procedure for the Collection of Information 15
B.2.1. Convenience Sample 15
B.2.2 Estimation Procedures 16
B.2.2.1 Hemoglobin Measures 16
B.2.2.2 Iron Status and Donation Intensity Association 17
B.2.2.3 Models Predicting Iron Status 18
B.2.2.4 Comparison to NHANES-III Hemoglobin distribution 19
B.2.3. Quality Control 19
B.3. Methods to Maximize Response Rates and Deal with Non-response 20
B.4. Test Procedures 20
B.5. Individuals Consulted on Statistical Aspects and Individuals Collecting and/or Analyzing Data 20
REFERENCES 21

ATTACHMENTS

Attachment 1:Legal authority. Congressional mandate, Sec 421 [285b-3] and 422 [285b-4]
Attachment 2: Iron Study Questionnaires
I. Baseline Questionnaire:
a. Donor Iron Status Survey (Cohort version)
b. Iron Status Survey (Deferred donor version)
II. Final Questionnaire
Attachment 3: Invitation to participate brochure
Attachment 4: Consent & Letters
4.1 Consent form
4.2 Targeted mailing for interim visits
4.3 Targeted mailing for final visits
Attachment 5: 60-Day Federal Register Notice
Attachment 6: Supporting Documents
6.1 Committee members
6.2 National Heart, Lung, and Blood Institute Strategic Plan FY 2002-2006
6.3 Iron Questionnaire: Description of each item, source, and goal.
6.4 OSMB Memo

Section A

Introduction

Introduction and Summary

Iron loss is a known consequence of blood donation. Although the overall health significance of iron depletion in blood donors is uncertain, iron depletion leading to iron deficient erythropoiesis and lowered hemoglobin levels results in donor deferral and, occasionally, in mild iron deficiency anemia. Hemoglobin deferrals represent more than half of all donor deferral, deferring 16% of donation attempts by women. This Iron Cohort study has been developed under the National Heart, Lung, and Blood Institute's (NHLBI) Retrovirus Epidemiology Donor Study-II (REDS-II) contract. The contract for REDS-II was awarded in August 2004 to six blood centers and a coordinating center (CC). The blood centers include the American Red Cross New England Region, Dedham, Massachusetts; the American Red Cross Southern Region, Atlanta, Georgia; Blood Center of Wisconsin (BCW), Milwaukee, Wisconsin; Hoxworth Blood Center, Cincinnati, Ohio; the Institute of Transfusion Medicine, Pittsburgh, Pennsylvania; and Blood Centers of the Pacific, San Francisco, California. Westat, located in Rockville, Maryland, serves as the CC. The CC is responsible for protocol implementation, monitoring, data management and analysis. A REDS-II central laboratory contract was also awarded to Blood Systems Research Institute in San Francisco, California.

Among the six REDS-II blood centers, a longitudinal study of iron status in two cohorts of blood donors will be conducted. The first cohort will be comprised of 1920 first time and returning (reactivated) donors for whom baseline iron and hemoglobin status can be assessed without the influence of previous donations, and a second cohort of 1440 frequent donor, where the cumulative effect of additional frequent blood donations can be assessed. For each cohort, donors will routinely donate blood during the study period. As part of the research study, a sample of their blood will be collected for laboratory testing and on specific visits, for repository storage. Additionally we will enroll a group of 500 first time and reactivated donors who are

deferred by the blood center for low hemoglobin to assess these donors' baseline iron and hemoglobin status.

At the baseline and final study visits, hemoglobin levels and a panel of iron protein and red cell and reticulocyte indices will be measured. The final study visit is defined as the latest visit occurring 15-24 months after the initial enrollment visit. At these visits donors will also complete a self-administered survey assessing past blood donation; smoking history; use of vitamin, mineral and iron supplements; use of aspirin; intake frequency of heme rich foods; and for females, menstrual status and pregnancy history. A DNA sample will be obtained once at the baseline visit to assess three key iron protein polymorphisms. Further, throughout the study when a subject returns, fingerstick hemoglobin or hematocrit values will be obtained and plasma will be collected for selected measurements of ferritin, and sTfR levels to the extent possible. Test results and survey data will be combined with demographic, anthropomorphic, racial/ethnic, and zip code (to estimate altitude) data routinely compiled at all REDS-II centers. Finally, a plasma and DNA linked repository will be established to allow for future assessment of new genetic, chemical or cellular markers of iron status, as related to blood donation behavior.

The primary goal of the study is to evaluate the effects of blood donation intensity on iron and hemoglobin status and assess how these are modified by demographic, reproductive and behavioral factors. This study aims to identify laboratory measures for predicting the development of iron depletion, and hemoglobin deferral in whole blood and double red cell donors as that could be potentially useful for donor management. The data collected will help evaluate hemoglobin distributions in the blood donor population (eligible and deferred donors) and compare them with NHANES data. It will also help formulate a predictive model for the development of iron depletion, hemoglobin deferral and/or iron deficient hemoglobin deferral in whole blood donors, which could be beneficial to developing optimal whole blood donation frequency guidelines.

Secondary objectives include elucidating key genetic influences on hemoglobin levels and iron status in a donor population as a function of donation history; and establishing a plasma and DNA archive to evaluate the potential utility of future iron tests and genetic polymorphisms.

A.1. Circumstances Requiring the Collection of Data

A. Justification

A.1. Circumstances Making the Collection of Information Necessary

Introduction

Deferral for low hemoglobin levels is the most common cause of presenting donor loss, particularly in females. Within the New England Region they represent more than half of all deferrals,1 nearly all being women. Approximately, 15% of women presenting at blood drives are deferred for their hemoglobin level, in contrast to approximately 5% of male donors. The deferral of large numbers of presenting female donors, many of whom do not return or are again deferred on repeat presentation, leads to significant problems maintaining, let alone increasing the blood supply –a principal goal of REDS-II.

Several cross sectional studies of blood donors, using older measures of iron status have indicated that being female, frequent donation and not taking iron supplements are predictors of iron depletion. However, none of these studies have included racial/ethnic, anthropomorphic, or behavioral factors and none have evaluated the impact of newly discovered iron protein polymorphisms.

Collecting the proposed information is a vital part of the overall responsibility of the Federal Government and U.S. blood collection centers to ensure the safety and availability of the national blood supply. NHLBI has a Congressional mandate, Sec 421 [285b-3] and 422 [285b-4] to ensure the overall safety of the blood supply (See Attachment 1). An important aspect to this assurance is ongoing epidemiology and laboratory research regarding blood donation practices and procedures to ensure the safety of donors while ensuring a blood supply adequate to fulfill

the nation's needs. The NHLBI strategic plan for FY 2002-2006 includes work to continue improving the safety and supply of blood for transfusion (See Attachment 6.2) and this study supports this NHLBI mission.

This study will develop better predictive models for the development of iron depletion and hemoglobin deferral (with or without iron deficiency) in blood donors. It will allow for improved donor screening strategies and open the possibility for customized donation frequency guidelines for individuals or classes of donors thus helping protect the well being of blood donors. It will provide important baseline information for the design of targeted iron supplementation strategies to replace iron loss in blood donors, and improved counseling messages to blood donors regarding diet or supplements. Finally, the elucidation of the effect of genetic iron protein polymorphisms on the development of iron depletion will enhance the understanding of the role of these proteins in states of iron stress, using frequent blood donation as a model.

A.2. Purposes and Uses of the Data

A.2. Purpose and use of the information

Data collected in this study will be of practical use to the blood banking community and to the Federal Government (See Section A.1.). In addition to the traditional route of peer reviewed scientific publication, previous REDS-I study data were the subject of numerous requested presentations by Federal and non-Federal agencies, including the FDA Blood Products Advisory Committee, the HHS Advisory committee on Blood Safety and Availability, the AABB Transfusion-Transmitted Diseases Committee, and the Americas Blood Centers Association. We anticipate similar requests for data generated from this study. The questionnaires for data collection have been designed to meet the analysis objectives detailed in Section A.16. There are three versions of the questionnaire for the purpose of collecting data at the baseline and final visits and a separate version for deferred donors. Attachment 2 lists the questions by study

objectives along with their specific goals and source. The broad categories of information to be collected are:

I. Baseline Questionnaire:

1. Donation history
2. Smoking history
3. Diet
4. Use of vitamins, supplements and aspirin containing pain killers
5. Reproductive history (for female donors only)

II. Final Questionnaire:

1. Smoking history
2. Use of vitamins, supplements and aspirin containing pain killers
3. Reproductive history (for female donors only)

III. Deferred Donor Version:

1. Donation history
2. Smoking history
3. Diet
4. Use of vitamins, supplements and aspirin containing pain killers
5. Reproductive history (for female donors only)

A.3. Use of Information Technology To Reduce Burden

A.3. Use of Information Technology and Burden Reduction

Due to the simplicity of the questions, we believe a self-administered, paper questionnaire that donors can quickly complete in privacy at the time of donation is the least burdensome form of data collection. The responses to the questionnaire will be entered into a secure, web based, data capture system by specially trained REDS-II research coordinators.

Donors will be assured of the confidentiality of their responses. A label will be placed on the paper questionnaire with a unique study ID number assigned to each enrolled donor. Use of the study ID on the questionnaire will allow for tracking of survey responses without entering identifying information into the study database. The link between the study ID number and the identity of the donor is only maintained by the blood centers. This link is maintained should there be a need to re-contact the donor in the future or should the donor wish to withdraw from the study. The CC will not have access to any donor identifying information.

Efforts to minimize respondent burden are described below:

1. The questions are presented along with easy-to-read instructions and skip patterns to avoid having respondents answer unnecessary questions.
2. The questionnaire contains tried and tested questions from previous REDS survey, the California Smoking Survey,[2] the NIH Diet History Questionnaire;3 the National Health and Nutrition Examination Survey (NHANES)[4] and the Mansfield-Voda-Jorgensen Menstrual Bleeding Scale [5].

A.4. Efforts To Identify Duplication

A.4. Efforts to Identify Duplication and Use of Similar Information

None of the iron status studies conducted in the past have included racial/ethnic, anthropomorphic, or behavioral factors and none have evaluated the impact of newly discovered iron protein polymorphisms. This information is not routinely collected by U.S. blood collection centers in the course of their regular donor screening operations.

A.5. Small Business

A.5. Impact on Small Businesses or Other Small Entities

Small businesses or entities are not involved. All respondents are individual blood donors.

A.6. Consequences of Not Collecting the Information

A.6. Consequences of Collecting the Information at a Chosen Frequency

Questionnaires will be administered at the baseline and final follow-up visit for the two donor cohorts (first time/reactivated and repeat donors). This will help to study the impact of diet, behavior, use of supplements, reproductive history for females, and frequency of donation on the iron status of the donor. Blood samples will be collected at these two visits and also during the interim visits. For the hemoglobin deferred donor group, it is proposed to collect this information only at the baseline visit.

A.7. Special Circumstances Justifying Inconsistencies with Guidelines in 5 CFR 1320.6

A.7. Special Circumstances Relating to the Guidelines of 5 CFR 1320.5

The proposed data collection is consistent with 5 CFR 1320.5.

A.8. Consultation Outside the Agency

A.8. Comments in Response to the Federal Register Notice and Efforts to Consult Outside Agency

The 60-day Federal Register Notice requesting comments was published on August 28, 2006, (Attachment 5). No comments were received in response to this notice. There has been consultation outside of NHLBI to conceptualize and design the proposed study. The final study design was developed, reviewed, and approved by the REDS-II subcommittee, the REDS-II Steering Committee, and the Observational Study Monitoring Board (OSMB) (See Attachment 6.1 for a complete list of members). The OSMB reviewed the final protocol and provided input and comments (See Attachment 6.4). Revisions were made to the sample size and consent document incorporating the suggestions of the OSMB.

A.9. Payments or Gifts to Respondents

A.9. Explanation of Any Payment or Gifts to Respondents

Study participants will not receive any payments or gifts.

A.10. Assurance of Confidentiality

A.10. Assurance of Confidentiality Provided to Respondents

The Privacy Act does not apply to the proposed data collection since identifiable information will not be collected on this questionnaire.

A.11. Questions of a Sensitive Nature

A.11. Justification for Sensitive Questions

The deferral of large numbers of presenting female donors, many of whom do not return or are again deferred on repeat presentation, leads to significant problems maintaining, let alone increasing the blood supply – a principal goal of REDS II. The relationship of blood donation to iron stores was first elucidated by Simon, et al. in a cross-sectional observational study of blood donors.[6] The overall frequency of iron depletion was 8% in male blood donors and 23% in female blood donors. The loss of approximately 230 mg iron with each whole blood donation along with a limited absorption capacity leads to a high incidence of iron deficiency in frequent donors, especially women. More than two thirds of body iron is contained in red cells, and red cell loss is the major route for iron loss from the body.

To satisfy study objectives, it is essential to collect information on factors that influence iron balance in women. This includes menstrual history, as well as the number, outcome (live birth, still birth, or whether they were miscarriages/terminated pregnancy) and date of pregnancy.

The average menstrual period results in a loss of 15-25 mg of iron contained in shed red cells. This causes pre-menopausal women to have a dietary requirement that is 50% higher than men. There is considerable variability in the frequency and amount of menstrual bleeding. The age of onset of menopause is quite variable as well. Because there is this significant variation

among women in their iron loss from non-blood donation sources, it is necessary for the protocol objectives to accurately assess these variables. In fact, it is expected that menstrual variables may be important in determining the ability of a woman donor to donate at her desired frequency.

Pregnancy places an even more extreme, although short-tem iron stress on women. The average pregnancy causes a woman to lose 700-1000 mg of iron, equivalent to the donation of 3-4 units of blood. Miscarriage or other earlier termination of pregnancy will have a lesser, but meaningful impact on body iron stores. Thus a woman's pregnancy history is highly important as a variable to explain her ability to donate blood subsequently.

For additional information see Attachment 6.3. There is no attempt to recruit female donors based on specific reproductive histories. Special attention has been devoted to carefully design possible sensitive questions related to pregnancy history in a straightforward and non-judgmental way. Demographic variables (e.g., age, gender, race/ethnicity), genetic factors (e.g., transferrin G277S polymorphism, HFE markers), behavioral factors (e.g., mineral supplements, smoking) are all known to impact iron or hemoglobin status. The study proposed here will investigate how these factors interact with one another to influence the development of iron depletion and deficiency in blood donors. These questions have been developed, tested, and are used by the National Health and Nutrition Examination Survey (NHANES) conducted by Health and Human Services (HHS). In addition, being aware of the possibly sensitive nature of the pregnancy related questions, the following steps will be taken to assure the confidentiality of respondents:

3. Identities of donors who enroll in the study are known only to the blood center. All information is transmitted to the CC via a secure web portal and will contain only Study ID numbers. These numbers can be linked to the donor's identity only by the blood center. Once entered and data verified, the blood center will not retain the hard copy questionnaires.
4. To help ensure privacy, the questionnaire will be self-administered.

5. All data will be stored in a secure location, accessible only to authorized study personnel.
6. Donors are advised of the voluntary nature of their participation in the study and of the steps taken to ensure the confidentiality of the information collected. See Informed Consent Document, Attachment 4.1.

A.12 Estimates of Response Burden

A.12. Estimates of Hour Burden Including Annualized Hourly Costs

The annualized cost to respondents is estimated at $9,270 for baseline visit and $3,672 for final visits based on $18 per hour. It is estimated that each respondent will spend about 7 minutes (0.12 burden hours) completing the baseline questionnaire and about 6 minutes (0.1 burden hours) completing the final follow up questionnaire. The respondent population of U.S. blood donors represents a wide range of wage rates. Therefore, the $18 per hour wage rate was selected based on reported overall labor force mean hourly earnings in 2004[7].

Table A.12. ESTIMATES OF HOUR BURDEN AND ANNUALIZED COST TO RESPONDENTS

Type of Respondents	Estimated Number of Respondents	Estimated Number of Responses per Respondent	Average Burden Hours per Response	Hourly Wage Rate ($)	Estimated Total Annual Burden Hours Requested
Blood donors at Baseline Visit	4,290	1	0.12	18	515
Blood donors at Final Visit	2,040	1	0.1	18	204
					719(Total)

A.12.1. Number of Respondents, Frequency of Response, and Annual Hour Burden

A.12.2. Hour Burden Estimates by Each Form and Aggregate Hour Burdens

A.12.3. Estimates of Annualized Cost to Respondents for the Hour Burdens

A.13. Estimate of Total Capital and Startup Costs/Operation and Maintenance Costs to Respondents or Record Keepers

A.13. Estimates of Other Total Annual Cost Burden to Respondents or Record Keepers

There are no capital or start-up costs, and no maintenance or service cost components to report.

A.14. Estimates of Costs to the Federal Government

A.14. Annualized Cost to the Federal Government

The total cost to the Federal Government for the proposed study is estimated to be approximately $3.2 million for a 24 month period, or an annualized cost of approximately $ 1.6 million.

A.15. Changes in Burden

A.15. Explanation for Program Changes or Adjustments

This questionnaire constitutes a new collection of information.

A.16. Plans for Publication, Analysis, and Schedule

A.16. Plans for Tabulation and Publication and Project Time Schedule

The schedule for study activities is shown in Table A.16.

Table A.16. PROJECT TIME SCHEDULE

Activity	Time Schedule After OMB Approval
Donor enrollment and baseline assessment visit	June-October 2007
Baseline Laboratory Testing	November-December 2007
Baseline data compilation and cleaning	January 2008
Baseline data analyses/interpretation	February 2008-May 2008
Interim Period	August 2007 – December 2008
Follow-up period	August 2008 – May 2009

Laboratory Testing	June-July 2009
Data compilation and cleaning	August 2009
Data analyses	September–December 2009

Subject to NHLBI approval, data will be disseminated to the scientific and blood banking community and others through peer-review journal publications, and presentations at government (FDA Blood Products Advisory Committee) and professional meetings (American Association of Blood Banks). We will use descriptive statistics to evaluate the distributions of all variables. We will use log-likelihood χ^2 statistics (or exact tests if cell sizes are too small) to evaluate if the distribution of a categorical characteristic (e.g., high vs. low baseline hemoglobin in first-time/reactivated donors; high vs. low HYPOm) is significantly different among groups (e.g. gender, race/ethnicity, iron supplementation vs. not). For comparison of continuous characteristics among groups, we will compare means among several groups by conducting t-test (two groups) or analysis of variance (> 2 groups); or if a non-parametric method is more appropriate by conducting a Wilcoxon rank-sum test (two groups) or a Kruskal-Wallis test (> 2 groups). Correlations and coefficients of determination may also be used to evaluate the association between two continuous variables. To evaluate whether two continuous variables are equivalent (such as evaluating if the hemoglobin level obtained by fingerstick HemoCue® is similar to that obtained from a pre-donation venous draw), a paired t-test could be used. Examples of longitudinal analyses to evaluate changes in fingerstick hematocrit/hemoglobin over time as a function of donation intensity are:

1) Analysis where fingerstick hemoglobin (HemoCue®) is the outcome variable of interest; this analysis will be restricted to the centers that will use HemoCue® as their test-of-record.

2) Analysis where fingerstick hematocrit is the outcome variable of interest; this analysis will be restricted to the centers that use a fingerstick hematocrit as their test-of-record.

3) Analysis where pre-donation venous hemoglobin is the outcome variable of interest. In this analysis, we will not use actual observed pre-donation venous hemoglobin values even if available but rather transform the fingerstick hemoglobin or hematocrit value obtained at each visit into a pre-donation venous hemoglobin equivalent using the appropriate conversion factors.

The baseline venous hemoglobin measures from FT/reactivated, deferred and accepted, donors will be weighted (to reflect differential sampling probabilities of deferred and non-deferred donors) to determine estimates of the mean hemoglobin levels for the blood donor population by age, gender and race. These means will be compared to the NHANES-III data results. Study hypothesis will be tested using cross-sectional analyses of end-of-study variables (primarily end-of-study iron status variables and donation intensity). These analyses will be based on the log-likelihood χ^2 statistics associated with the corresponding 2x2 tables. Males and females will initially be analyzed separately.

A.17. Approval to Not Display Expiration Date

A.17. Reason(s) Display of OMB Expiration Date is Inappropriate

The OMB expiration date will be displayed in the upper-right hand corner of the questionnaire.

A.18 Exceptions to Item 19 of OMB Form 83-I

A.18. Exceptions to Certification for Paperwork Reduction Act Submissions

There are no exceptions to the certification statement of OMB Form 83-I.