

Subprotocol
*Study of Donors with Equivocal Assays for Antibody to Human Immunodeficiency Virus:
Evaluation and Follow-up of Recipients*

INTRODUCTION

In 1984, under emergency powers, the National Heart, Lung, and Blood Institute (NHLBI) issued a contract (NO1-HB-4-7002) for the collection of serum samples from routine blood donors prior to the availability of screening tests for antibodies to the human immunodeficiency virus (HIV). The immediate use for the TSS/NHLBI Donor Repository was to identify potentially infectious donors and evaluate transmission to recipients by the components of the donation.

Samples were taken from donors in four geographic areas with the highest prevalence of AIDS: the Greater New York area, served by the New York Blood Center; the South Florida area, served by the American Red Cross Regional Blood Services; San Francisco and Bay area counties to the north of that city, served by the Irwin Memorial Blood Bank; and Los Angeles County, served by the American Red Cross Regional Blood Services.

From early September, 1984, until early February, 1985, donors at the participating blood services were asked to permit the collection of an additional 10 ml of clotted blood for HIV antibody testing. They were also asked for permission to contact them later if the results of such tests indicated that further observations would yield useful scientific information.

At the end of Repository collections in early 1985, a total of 200,000 donor serum specimens had been stored. Their distribution was as follows: New York Blood Center, 77,760; American Red Cross Blood Services, South Florida Region, 31,104; Irwin Memorial Blood Bank, 27,500; and American Red Cross Blood Services, Los Angeles-Orange Counties Region, 67,418.

After licensure of an enzyme-linked immunoassay (EIA), work began at each blood service to screen samples in the Repository for antibodies to HIV. Initially positive samples were retested in duplicate and if found to be repeatably positive an aliquot was sent to the Central Processing Laboratory (CPL) for corroborative testing.

Whenever a repeatably positive sample was identified, a negative control was selected matched with the EIA-positive donor on age group, sex, geographic classification, and date of donation within one week. Samples from potential control donors were retested in duplicate and if found to be repeatably negative an aliquot was sent to the CPL for corroborative testing.

In addition to the above samples, aliquots of all specimens giving non-repeatable screening results, whether initially positive or negative, were sent to the CPL for further testing. The procedures performed for verification of screening results included repetition by a different EIA from another manufacturer, and the identification of antibodies to specific viral antigens by protein immunoblot (PIB) and radioimmunoprecipitation (RIP).

Confirmed anti-HIV positive donors and confirmed negative donors (defined as Group A and B donors, respectively) and the recipients of blood components derived from their donations (defined as Group A and Group B recipients, respectively) are currently being studied as described in the primary protocol. The study of donors whose samples yielded discrepant or equivocal results was deferred, and is the subject of the present subprotocol. The purpose of the proposed study of donors for whom test results were equivocal, and of the recipients of their donated blood is to determine the relative sensitivities of EIA screening at a blood service and corroborative procedures.

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SCIENTIFIC QUESTION TO BE ASKED

What is the meaning of discrepancies between EIA testing and the "confirmatory" procedures (PIB and RIP)? These can best be clarified by determining the later status of the donor and the status of the recipient.

OBJECTIVE

To use observations of donors, follow-up of recipients, and new laboratory procedures to determine the meaning of discrepancies among tests used in screening of the TSS/NHLBI Donor Repository.

STUDY POPULATIONS

1. Persons in the geographic areas listed below who gave permission at the time of blood donation to have serum tested for anti-HIV and to be contacted concerning further participation if results of that testing indicate that additional observations would add to knowledge concerning the meaning of the test.
 - a) Donors to the New York Blood Center's facilities in all four of its divisions (New York City, Northern New Jersey, the Hudson Valley, and Long Island).
 - b) Donors to the South Florida Region of the American Red Cross Blood Services (formerly, South Florida Blood Service).
 - c) Donors to the Irwin Memorial Blood Bank at its facilities in San Francisco and adjacent counties to the north.
 - d) Donors to the American Red Cross Regional Blood Services' facilities located in Los Angeles County.
2. Recipients of blood or blood components at hospitals in the areas served by the participating blood collection agencies.

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The following characteristics of anti-HIV test results will be used to define further the TSS donors and their recipients to be studied under this subprotocol.

Probably negative donors are those who had results summarized as one of the following:

- (1) repeatable anti-HIV positivity on EIA screening, with either H9 specificity or non-specificity, and with either PIB or RIP negative and the other of these two confirmatory assays equivocal, with both PIB and RIP equivocal, or with both PIB and RIP negative;
- (2) non-repeatable positivity on EIA screening, with either H9 non-specificity or no H9 result available, and with either PIB and RIP negative and the other of these two confirmatory assays equivocal or with both PIB and RIP negative; or,
- (3) repeatable anti-HIV negativity on EIA screening, no H9 result, and with either PIB and RIP negative and the other of these two confirmatory assays equivocal, or with RIP negative and PIB positive.

The following are the patterns of interest and their frequencies in the Repository:

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Initial   Repeat EIA
EIA      mean      H9      PIB      RIP      Frequency
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P         P         P         Q         N         2
P         P         NS        Q         N         20
P         P         P         N         Q         1
P         P         NS        N         Q         6
P         P         NS        Q         Q         2
P         P         P         N         N         11
P         P         NS        N         N         174
P         N         -         N         Q         1
N         P         NS        Q         N         1
P         N         -         N         N         1
N         N         -         Q         N         14
N         N         -         N         Q         7
N         N         -         P         N         1
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These donors will be defined as *Group C donors*, and their recipients will be classified as *Group C recipients*.

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Possibly positive donors are those who had repeatable anti-HIV positivity on EIA screening, with either H9 specificity or non-specificity, and PIB *or* RIP positivity. The following are the patterns of interest and their frequencies in the Repository:

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Initial   Repeat EIA
  EIA     mean      H9      PIB      RIP      Frequency
-----
  P        P        P        Q        P         2
  P        P        P        P        Q         2
  P        P        NS       P        N         1
=====
  
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These donors will be defined as *Group D donors*. Their recipients will be classified as *Group D recipients*.

PROCEDURES

Donor Follow-up

The Director of the CPL will provide the Study identification number of each Group C or D donor to the person responsible for donor recruitment at the clinical center where the donation was made.

Throughout the identification and follow-up of donors, all specifically identifying information will be available only to those persons for whom it is absolutely necessary for making contact. Each collection agency will develop its own plan for accomplishing this purpose, which must be based on its system of keeping records. No donor will be contacted until the patient manager or TSS-blood bank clerk has located the informed consent signed by the donor. Both will make all possible efforts to protect the identity of these persons.

From the blood unit number, the patient manager or data clerk will obtain the donor's name and other information necessary to make contact. When the donor is first contacted, he will be reminded of his previously expressed willingness to be contacted if further participation may be helpful in evaluating new tests related to transfusion safety. The donor will be asked to present himself at a convenient facility available to the blood service to discuss further participation. The site and time will be one of mutual convenience. If the donor states that he is not interested in further participation but wants to know his test result, he will be told that, to ensure privacy, test results cannot be given over the phone or by mail. To ensure confidentiality, it is essential that the person satisfactorily be identified as the person who made the donation. He must, therefore, provide adequate identification in person at a facility available to the blood service in order to learn his result.

When the donor presents himself to the blood service, he will be reminded again that he agreed to participate in a study of transfusion safety during a recent blood donation and will be asked to participate in the second phase of the study. A copy of his signed consent form for the pre-screening phase will be available. The donor will be asked to sign an informed consent agreeing to be interviewed, to have a

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simple physical examination, and to have a blood sample taken at specified intervals for a period of up to four years for HIV testing (antibody and possible virus isolation), tests of immune function (T-cell phenotypes and possible special tests), and for storage in a subject repository. The donor will be given the opportunity to ask the patient manager any questions about the study and the meaning of anti-HIV tests. If the donor does not want to participate in the study, he will be given his test result at this time and its meaning will be discussed.

The donor will be given a release form which would permit the blood center to release the test results to his personal physician in a fashion which will protect the donor's confidentiality. A brief description of the research study will also be provided to the donor's physician.

After obtaining informed consent, the patient manager will conduct the entry interview, which will contain questions about medical history, present health, and patterns of behavior, including sexual practices (specifically homosexual contacts) and recreational intravenous drug use. The patient manager will then conduct a partial physical examination, including height, weight, temperature, and examination of the mouth, exposed skin, and lymph nodes in the head and neck. If any abnormalities are found, a physician on the study staff will conduct a more complete physical examination, which will include checking for hepatosplenomegaly and a more extensive examination of the skin and lymph nodes, including axillary and inguinal nodes. The patient manager will also draw a sample of blood. The blood collection procedure, including the amount of blood to be drawn, is specified in the manual of operations.

The procedure for handling the completed questionnaires, physical exam records, and blood specimens is described in the manual of operations.

All subjects entered into the study will be placed into one of three categories based on symptoms, signs on physical exam, and laboratory findings: asymptomatic, AIDS-related findings, or AIDS. These categories are defined in the manual of operations and will be consistent with CDC criteria. Subjects who are asymptomatic will be followed at six-month intervals. Subjects with AIDS-related findings or AIDS at entry into the study will be followed every three months. Subjects who are asymptomatic at entry but who subsequently present with symptoms will be reclassified and followed at three-month intervals. Symptoms and/or findings related to conditions other than AIDS may also make subjects eligible for special follow-up.

For each donor participating in the study, a schedule of dates for follow-up visits will be kept in the central record. One month before the scheduled visit the patient manager will contact the donor to arrange the time and site for the visit. The follow-up visit should occur within one month of the date scheduled in the record.

Each follow-up visit will include a shorter version of the entry interview, a physical examination, and drawing a sample of blood, as described above.

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Recipient Follow-up

Recipients of blood or components from Group C or D donors will be observed. Adult recipients will be identified by reviewing blood center records and then contacting the blood bank director of the hospital that received the blood component. Information will be obtained from the blood bank director concerning the patient who received the unit. The physician who ordered the blood component will also be identified from blood bank records and will be contacted by a physician on the study staff or the patient manager assigned to recipient follow-up (depending upon the written procedures of each clinical center). If he is not the physician currently responsible for the recipient, he will be asked to identify the physician who is responsible.

The responsible physician will be told about the study and asked to give permission for his patient to be recruited for the study. If the physician agrees, the recipient will be contacted according to the procedures established by each clinical center. These procedures include the following options:

1. The physician will be asked to contact his patient, inform the patient of the study, and recommend that the patient participate in the study. If the recipient is willing to participate in the study, the physician will inform the patient manager, who will then contact the recipient.
2. The patient manager will make the initial contact by telephone. Using a standardized script, an attempt will be made to be certain that the person on the telephone is actually the recipient. The patient manager will explain that a national study of transfusion safety is being conducted and that the recipient was selected to participate because he recently received blood or blood components.
3. The recipient will be informed of the study jointly by the physician and patient manager in the physician's office.

If the physician refuses permission to contact his patient, the recipient will not be contacted.

When the recipient presents himself to the study staff member, he will be told by the appropriate person (the personal physician, a physician member of the study team, or patient manager) about his exposure to blood for which anti-HIV testing yielded equivocal results and about the study. It will be emphasized that his participation is being sought to better understand the interpretation of equivocal results, and these results do not mean that he was exposed to HIV. He will be given an opportunity to ask the staff member about the meaning of the possible exposure and the study, and will be asked to sign an informed consent to be interviewed, to have a simple physical examination, and to have a blood sample taken for HIV testing (antibody and possible virus isolation), immune function testing, and storage in a subject repository. The patient will be told that these studies will be repeated at specified intervals for a period of up to four years.

If the recipient does not want to participate, there will be no attempts at further contact by any member of the study team. If the recipient signs the consent form, the patient manager responsible for recipient follow-up will conduct the interview and physical examination and will draw the blood sample as described above for donors.

Minors. The recruitment procedures described for adults will be followed, with the legal guardian(s) acting as the recipient's representative(s). The legal guardian(s) will be asked to sign the informed

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consent form specifically designed for minor recipients. In addition, children over age 12 will be asked to sign an assent form. If the appropriate form(s) are signed, the patient manager will conduct the interview and physical examination, and will draw the blood sample. The parent/legal guardian will be interviewed. Minors over a certain age, specified by each clinical center, will also be interviewed.

Adult recipients for whom there is a legally appointed guardian will be approached in the same way as minors.

Follow-up visits for recipients will be handled in the same way as described above for donors.