RESEARCH TRIANGLE INSTITUTE

APPENDIX 12

PROSPECTIVE STUDY OF PULMONARY COMPLICATIONS OF HUMAN IMMUNODEFICIENCY VIRUS INFECTION

Manual of Operations for Data Collection

3040 Cornwallis Road Research Triangle Park, NC 27709 (919)541-6000

MANUAL OF OPERATIONS FOR DATA COLLECTION

OUTLINE

OUTLINE
(continued)

OUTLINE
(continued)

PREFACE

The Manual of Operations is concerned solely with the collection of the data specified in the study protocol. Procedures to be followed in systematically collecting and recording study data are discussed in this manual. The design of the study is discussed in the study Protocol.

All personnel who are involved in this study are urged to read this manual and the study Protocol very carefully. Questions about the operational aspects of the study protocol and the procedures discussed here should be addressed to the Principal Investigator.

1. INTRODUCTION

It is increasingly apparent that the concept of the ***acquired immunodeficiency syndrome*** (AIDS) presents a very limited and incomplete picture of the manifestations of human immunodeficiency virus (HIV) infection. Rather, it is more valid from both clinical and epidemiologic points of view to think in terms of ***HIV disease***, a designation that encompasses the full range of both primary and secondary effects of HIV infection.

Current data indicate that infection with HIV, once established, persists. Although there is a several year period of latency between infection and the onset of symptoms that are clearly attributable to HIV, it appears that there are ongoing, progressive effects on host immune responsiveness. These effects are measurable in part as a decrease in the number of circulation helper T lymphocytes (T-4 or CD-4 cells). The reduction in immune response sets the stage for a variety of opportunistic diseases, some of which are currently considered to be AIDS-defining and mark the end-stages of HIV disease. The point at which opportunistic illnesses occur is probably determined by the interactions of a number of factors, including the status of non-immunologic host defenses, exposure to or previous infection with specific pathogens, the virulence of these pathogens, and the effects of HIV infection, to name a few. Because of the complexity of these interactions there is no ***threshold*** before which opportunistic diseases are not seen and beyond which they occur with predictable frequency. Thus, it is likely that, although the frequency and severity of disorders associated with HIV disease occur in crescendo fashion as the end-stages of the infection approach, HIV plays a role in the causation of disease all along the course of infection.

In addition to the secondary opportunistic processes associated with HIV disease, HIV itself may primarily cause symptoms and organ system dysfunction that contribute to the spectrum of illnesses. Examples of diseases resulting directly from HIV infection include acute ***seroconversion*** illness, dementia, myelopathy, peripheral neuropathy, and, perhaps, lymphadenopathy and gastrointestinal disease.

Because the lungs are the most frequent sites of AIDS-defining diseases, it is logical to assume that there may be pulmonary involvement, either primary or secondary, at any point in the course of HIV disease. Although much is known about the pulmonary disorders associated with AIDS, there are very important deficiencies in the available information. These deficiencies are due in large part to clinicians and investigators focusing on those disorders that serve to

define AIDS or that occur in patients who have AIDS. There is little or no information that systematically identifies the pulmonary disorders that occur at all stages of HIV disease or that describes the course patients follow after the diagnosis of a lung disease has been established.

2. STUDY OVERVIEW

The Pulmonary Aids Clinical Study (PACS) seeks to provide information concerning pulmonary diseases that occur as a result of infection involving the human immunodeficiency virus (HIV). The primary objective of the study is to determine the frequency and types of lung diseases that occur in persons with HIV infection and to describe the course and outcome of these disorders. These diseases or abnormalities may be either secondary to the immunologic abnormalities caused by the virus (opportunistic processes) or be the result of the HIV infection itself.

The Pulmonary Aids Clinical Study will be based in seven Clinical Centers (six in the United States and one in West Germany) and will be a prospective, longitudinal evaluation of a cohort of seropositive persons and seronegative control subjects. Seropositive subjects will be stratified into two groups defined by the number of circulating CD-4 cells ($\geq$ 400 per microliter, $<$ 400 per microliter) and the presence or absence of ***pre-AIDS*** conditions. Evaluations of these three groups will enable identification of the lung diseases that occur at various stages of HIV disease. The ***background*** prevalence/incidence of various diseases and abnormalities that are related to demographic, environmental, or lifestyle factors, rather than HIV infection, will be determined by evaluation of the seronegative group.

Evaluations of study participants will be conducted at regular intervals and when predefined symptoms occur. Screening and diagnostic studies that are of proven value in detecting AIDS-associated lung diseases will be used. Tests will be performed in a uniform manner using standardized techniques among all Clinical Centers. Subjects will be followed for between 3 and 4 years (one year recruitment period and 3 year period of observation) or until death.

As a substudy, the effect of attempting to detect ***P. carinii*** pneumonia at a ***pre-clinical stage*** was evaluated and found not to be of any value. Seropositive subjects were allocated at random to either ***routine*** (testing at 12-month intervals) or ***intense*** (testing at 3-month intervals) evaluations.

Data from all components of the study will be transmitted by all Clinical Centers to the Coordinating Center located at Research Triangle Institute. The Clinical Coordinating Center will maintain all study-wide data files, will monitor performance of each of the centers, and will coordinate a quality control program that will ensure uniformity of data quality.

Appropriate statistical techniques will be applied in the analysis of data relevant to the various questions being asked.

A variety of carefully designed safeguards will be implemented at both the Clinical Centers and at the Clinical Coordinating Center to ensure the confidentiality of subject records.

3. STUDY ORGANIZATION

3.1 Study Officers

The ***Project Officer*** for the Pulmonary Aids Clinical Study is Dr. Tony Kalica from the Division of Lung Diseases at the National Heart, Lung and Blood Institute. Dr. Kalica is responsible for providing NHLBI's views on the organizational, scientific and statistical direction of the study. His is also responsible for all administrative and fiscal matters related to the award and conduct of the contract.

The ***Chairman*** for the Pulmonary Aids Clinical Study is Dr. Phil Hopewell from the University of California, San Francisco. As Chairman, Dr. Hopewell provides leadership for the study, sets agendas for study meetings and presides over all Committee meetings.

The ***Vice-Chairman*** for the study is Dr. Jeff Glassroth, from Northwestern University in Chicago, Illinois. Dr. Glassroth assumes all Chairman duties in the absence of Dr. Hopewell.

3.2 Steering and Planning Committee

The Steering Committee meets a minimum of twice yearly to provide scientific direction to the study on an operational level. The membership is comprised of NHLBI project officers, principal investigators and clinic coordinators from each of the study clinics, and key members from the Clinical Coordinating Center. The special functions of the Steering Committee are:

- To see that the program policy and protocol is carried out under the guidance of the DLD and AP Project Officers.
- To review and analyze the progress of the program.
- To make recommendations to the Policy and Data Safety Monitoring Board concerning changes in the protocol and Manual of Operations.
- To review all proposed ancillary studies and to report all recommendations to the Policy and Data Safety Monitoring Board (the major criterion being the possible effect on accomplishing the objectives of the main study).
- To monitor the performance of the individual Clinical Centers with regard to patient recruitment and patient follow-up studies.
- To monitor the quality of data collected.

- To be responsible for the presentation of the program result to the biomedical community.

3.3 Clinical Coordinating Center

The Clinical Coordinating Center (CCC) is under the direction of the Steering Committee. The CCC's role in the study is in the design, implementation, and execution of the study. Coordinating Center staff is responsible for collection, editing, storing and analyzing all data from the study clinical centers and outside review consultants. Among the specific functions of the Clinical Coordinating Center are:

- To participate with the investigators in the development of the study protocol, forms, data reporting procedures, and the Manual of Operations.
- To pretest the procedures for data recording, processing, and reporting.
- To make random assignment of patient entry into the study.
- To review and edit all data transmitted to the Coordinating Center.
- To participate in the establishment and monitoring of quality control procedures.
- To provide statistical analyses of all study data.
- To check the completeness of records and periodically prepare performance reports to the Division of Lung Diseases and the participating Clinical Centers.
- To analyze periodically the frequency of adverse side effects of the diagnostic procedures and to report these data to the Policy and Data Safety Monitoring Board.
- To prepare interim technical and statistical reports for the Steering Committee.
- To monitor patient recruitment at each Clinical Center.
- To assist in the preparation of reports of the study for publication.
- To provide administrative support to the program office in arranging the meetings of the study investigators.
- To interact with the program office on all aspects of the study: design, conduct, monitoring progress and scientific inference.

The Clinical Coordinating Center staff meet on a weekly basis to review the current work being done and to plan for work to be done in the future. The Clinical Coordinating Center for the project is:

Research Triangle Institute
Post Office Box 12194
Research Triangle Park, NC 27709-2194

Dr. Kenneth Poole is the Principal Investigator at the Coordinating Center.

3.4 Clinical Centers

The clinical centers for the Pulmonary Aids Clinical Study are responsible for recruiting study patients, performing clinical evaluation and testing as required by the protocol, and collecting, recording and forwarding these data to the Coordinating Center. The Principal Investigator at each Clinical Center will be responsible for overseeing study operations at their clinic. The institutions participating in the study as Clinical Centers and their respective Principal Investigators are listed below.

University of California, San Francisco
San Francisco, California
Philip Hopewell, MD, Principal Investigator, Study Chairman

Northwestern University
Evanston, Illinois
Jeffrey Glassroth, MD, Principal Investigator, Vice Chairman

Mount Sinai School of Medicine
New York, New York
Mark Rosen, MD, Principal Investigator

University of Medicine and Dentistry of New Jersey
New Jersey Medical School
Lee B. Reichman, MD, Principal Investigator

University of California, Los Angeles
Los Angeles, California
Jeanne M. Wallace, MD, Principal Investigator

Henry Ford Hospital
Detroit, Michigan
Paul A. Kvale, MD, Principal Investigator

Klinikum J.W. Goethe University
Federal Republic of Germany
Michael Rust, MD, Principal Investigator

3.5 Project Offices

The Division of Lung Diseases (DLD), National Heart, Lung and Blood Institute, and the AIDS Program (AP), National Institute of Allergy and Infectious Diseases, as sponsors of this study, are responsible for providing organizational, scientific, and statistical direction to the study. Representatives from NHLBI are members of the Steering Committee and the Policy and Data Safety Monitoring Board.

3.6 Data Safety and Monitoring Board

The Policy and Data Safety Monitoring Board acts in a senior advisory capacity to the DLD and AP on policy matters throughout the duration of the study. In addition, it periodically reviews study results and evaluates the study diagnostic procedures for beneficial and adverse effects. This board meets twice a year. Specific functions of the Policy and Data Safety Monitoring Board are:

- To review and approve the study protocol, forms and Manual of Operations.
- To review and analyze the progress of the study, and to evaluate its relevance to the program goals.
- To monitor the study diagnostic procedures for beneficial and adverse effects on the patient.
- To make recommendations to the DLD and AP on major changes in the protocol, forms, or Manual of Operations.
- To review and advise DLD and AP ancillary studies(with the possible effect on the main study being the major criterion).
- To review the quality of the data.
- To assist the DLD and AP in resolution of problems referred by the Steering Committee.
- To make recommendations to the DLD and AP on any proposed early termination of the study because of failure to achieve recruitment goals or adverse or beneficial effects of any study procedure.
- To recommend remedial measures or discontinuation of individual Clinical Centers which perform unsatisfactorily.

3.7 Executive Subcommittee

The Executive Subcommittee meets as necessary between Steering Committee meetings to review interdisciplinary issues on the Steering Committee agenda. Specific functions of the Executive Subcommittee are: to make recommendations to the Steering Committee; to assign working groups; to review clinical science procedures for consistency; to review data collection and interpretation; to promote communication within centers.

4. PATIENT SELECTION AND RECRUITMENT

4.1 Composition of Study Cohort

The study cohort will consist of three groups of subjects (Table 1): Group A, HIV seropositive subjects with ≥ 400 CD-4 cells and no clinical manifestations of HIV infection; Group B, a second HIV seropositive group with < 400 CD-4 cells or clinical manifestations of HIV infection; and Group C, an HIV seronegative control group drawn from the same HIV transmission categories as the seropositive subjects. The relative proportions of these three groups will be the same for each clinical center. Each Clinical Center will recruit a study population that is representative of the HIV transmission categories in their study area. The transmission categories include homosexual/bisexual men, intravenous drug users, and seropositive women who have no risk factors other than sexual contact with presumed HIV infected men.

Table 1. Definition of Study Groups

Group A:	HIV seropositive. No symptoms or findings attributable to HIV. CD-4 cells ≥ 400 per microliter.
Group B:	HIV seropositive. Either clinical manifestations attributable to HIV within the past 6 months (thrush, hairy leukoplakia, fever > 38° C persisting > 2 weeks, involuntary weight loss > 10% of baseline, or diarrhea persisting > 1 month) or CD-4 cells < 400/microliter.
Group C:	HIV seronegative homosexual/bisexual men and IVDUs.

Each of the seven Clinical Centers enrolled approximately 200 subjects in the first year of the study. All drop-outs during the first year were replaced, hence, the study population totaled 1497 by the end of recruitment and of the first year of the study. Table 2 shows the actual number of study subjects by clinical group and transmission category that were recruited in the first year of the study. The relative transmission categories for study subjects include

homosexual/bisexual men, intravenous drug users, and seropositive women who have no risk factors other than sexual contact with presumed HIV infected men.

Table 2. Number of Study Subjects By Clinical Status and Transmission Category

	A	B	C	Total	
H/B[1]	431	509	134	1074	
IVDU[2]	132	167	60	359	
Partners[3]	34	30	0	64	
Total	597	706	194	1497	recruited Yr 1

1 H/B - Homosexual/Bisexual males
2 IVDU - Intravenous Drug Users
3 Partners - Seropositive female sexual partners of HIV infected men

Target quotas have also been established for each Clinical Center. Table 3 shows the transmission category quotas for each of the seven centers. The apportioning of subjects among the three groups (A, B, C) and transmission categories were monitored by the Clinical Coordinating Center and monthly quotas were established to guide the Clinical Centers in recruitment.

Table 3. Target Quotas for HIV Transmission Categories By Center

	H/B[1]	IVDU[2]	Partners[3]	Total
New York	170	29	16	215
Newark	3	198	2	203
Detroit	175	38	12	225
Chicago	186	24	16	226
San Francisco	201	44	8	253
Los Angeles	233	7	7	247
Frankfurt	106	19	3	128
Total	1074	359	64	1497

1 H/B - Homosexual/Bisexual males
2 IVDU - Intravenous Drug Users
3 Partners - Seropositive female sexual partners of HIV infected men

4.2 Eligibility Criteria

Eligibility criteria were established to help screen participants for enrollment into the study. Inclusion criteria were established to help ensure optimal compliance with the study protocol and with quality control. Exclusion criteria were established to assure that subjects would be likely to complete the study, and that meaningful data relative to the course of HIV-associated disease could be collected.

Inclusion Criteria:

- Membership in at least one of the study defined HIV transmission groups: homosexual/bisexual men, intravenous drug users, seropositive female partners of presumed HIV infected men
- Willing to have HIV antibody status determined
- Willing to have evaluations for symptoms or findings suggestive of pulmonary disease performed in one of the Clinical Centers, or allow the data from such evaluations to be retrieved by the Clinical Centers
- Willing and able to provide informed consent
- Willing to be informed of HIV antibody status as required by Federal legislation

Exclusion Criteria:

- Younger than 18 years
- Presence of a non-HIV-related process likely to interfere with survival
- Judged unwilling or unable to cooperate with the study protocol
- Use of immunosuppressive therapy (Table 4) within the last six months
- Presence of an underlying disorder that makes future use of immunosuppressive therapy likely
- Lung disease that may interfere with evaluations (Table 4)
- Presence of pulmonary symptoms at baseline evaluation (Table 4)
- Current or previous AIDS-defining diseases (Table 4)
- (Applies only to female partners of HIV infected men)
  Receipt of blood or blood products or intraveneous drug use in the past 10 years

4.3 Recruitment Strategies

Study subjects were recruited from various sources, depending on the Clinical Center and the transmission category to be recruited. Based on the potential sources as delineated below, flyers, brochures, posters, and contact with study personnel publicized the program. Each clinical center developed their own strategies to recruit patients. Each center's particular strategy has been outlined in Appendix 4 of the study protocol.

Sources of Subjects:

The following is a composite listing of possible sources of subjects compiled from input from various Clinical Centers:

- Existing hospital clinics, services and programs: Hemophilia Clinic, Sexually Transmitted Disease Clinic, Tuberculosis or Chest Clinic, Infectious Diseases Clinic, Pediatric Clinic (for mothers of AIDS children), AIDS Clinic (for sexual contacts of AIDS patients)
- Hospital Consultation Services: Pulmonary Disease Service, Infectious Disease Service
- Existing cohorts: ATEU, CSG, pediatric CSG (parents of AIDS children) MACS, other established cohorts (having the advantage of demonstrated motivation and commitment already generated by the subject)

Table 4. Excluding Conditions and Therapies

Immunosuppressive therapy within previous 6 months:

- Corticosteroids (systemic administration of > 15mg prednisone or equivalent amount of other preparation for > 14 days)
- Cytotoxic agents
- Antimetabolites Cyclosporin

Lung diseases that may interfere with evaluation:

- Pneumonia, any cause, within 3 months
- Invasive pulmonary fungal infection diagnosed within the past 12 months or continuing to receive treatment for such infection
- Pulmonary mycobacterial disease diagnosed within the past 12 months or continuing to receive therapy for the disease
- Diagnosed diffuse fibrotic lung disease or sarcoidosis

Respiratory symptoms

- Unexplained cough that persists for more than five days
- Unexplained breathlessness progressive over five days or severe one day

AIDS-defining diseases (MMWR 1987;37:45) (Appendix 12)

- Items 1-12 Section I-B CDC definition
- Items 1-12 Section II-A CDC definition
- Items 1-7 Section II-B CDC definition

Note: Potential study subjects who at the time of initial evaluation are excluded because of any of the exclusions listed above (1-3), except fibrotic lung disease or sarcoidosis may be reevaluated for study entry after an appropriate time interval.

- Gay events: Various gay events may be held by Gay Men's Health Associations and similar groups. Support is often requested from local agencies and personnel can be assigned to participate in the gay event and impart information of the PACS study at the encounter

- The private practices of physicians who see large numbers of HIV infected persons

- Methadone Maintenance Treatment Programs and other drug programs (advantage in commitment of subjects to continued care in drug treatment program)

- Individuals responding to word of mouth or media-generated publicity about the project (advantage of subject generated interest in the program).

4.4 Informed Consent

Informed consent was obtained from all study subjects prior to enrollment into the study. A patient consent form describing the study was prepared for this purpose. Personnel enrolling patients into the study explained the study and obtained consent in the form of a signature on the patient consent form. Those who refused consent were not entered into the study. The following information was provided to each subject:

- A statement that the study involves research, an explanation of the purposes of the research, the expected duration of the subject's participation, and a description of the procedures to be followed

- A description of any reasonably foreseeable risks or discomforts to the subject

- A description of any benefits to the subject or to others which may be expected from the research

- A disclosure of appropriate alternative procedures or courses of treatment, if any, that might be advantageous to the subject

- A statement describing the extent, if any, to which confidentiality of records identifying the subject will be maintained

- An explanation as to whether any compensation and an explanation as to whether any medical treatments are available if injury occurs and, if so, what they consist of, or where further information may be obtained

- An explanation of whom to contact for answers to pertinent questions about the research and research subject's rights, and whom to contact in the event of a research-related injury to the subject

- A statement that participation is voluntary, that refusal to participate will involve no penalty or loss of benefits to which the subject is otherwise entitled, and that the subject may discontinue participation at any time without penalty or loss of benefits to which the subject is otherwise entitled

- A statement that randomization may be involved.

5. ENROLLMENT AND INITIAL VISIT

5.1 Enrollment

Potential study participants were approached and introduced to the study by the principal investigator and/or program coordinator either at the study center or at the place of referral. The introduction entailed a detailed explanation of the study, its objectives and the procedures to be used in the study.

After appropriate description of the study, the eligibility of the patient was determined. This was done using the inclusion and exclusion criteria that was discussed earlier in this manual. Patients that passed all eligibility requirements were required to sign an informed consent form before being officially enrolled into the study. The informed consent form was reviewed with the participant and then signed by the participant if he/she desired to be enrolled into the study. Subjects who refused to give consent were not entered into the study.

After the informed consent was obtained, a patient ID number was assigned to the study participant. The next available ID from the list generated by RTI for the specific clinic was assigned to the participant. The initial visit and assessment was performed by the investigators and/or coordinator. The initial visit and assessment required an initial interview, physical examination, and initial laboratory examinations (chest films, blood studies, pulmonary function tests and sputum induction and skin tests) to be performed at the Clinical Center. Once all examinations were completed and results were returned from the study labs to the Clinical Centers, the participant's study classification was determined. The investigator/coordinator reviewed the patient's initial interview, laboratory examinations and CD-4 count and used guidelines set up in the study protocol to assign the patient to his/her classification group. Note that if information was found during the initial assessment that would exclude the patient from the study, the patient was dropped from the study at that time.

The randomization of patients into either routine (A1, B1) or intense (A2, B2) follow-up group was done at the Clinical Center via the randomization computer program set up by RTI on the clinic's computer. This allowed for immediate randomization at the center once the

patient's study classification (Group A or B) had been determined. The patient's ID and study classification was entered into the program and the patient's randomization and randomization sequence number was shown on the screen. The randomization sequence number kept up with how many randomizations were made in each classification group. This information was kept in a log by the coordinator at the Clinical Center as a backup to the computer system. Once the randomization was assigned, the study participant was advised of his/her schedule of follow-up visits.

After the patient was officially enrolled and participating in the study, the patient's care provider was contacted, informed of the study, and his/her cooperation sought to ensure that all diagnostic evaluations are performed in the Clinical Center or that such data were made available to personnel at the Clinical Center.

5.2 Initial Visit and Assessment

The initial visit and assessment mentioned above, was conducted as soon as possible after the study participant was deemed eligible for the study and had signed the necessary consent forms. Details of the study procedures were again reviewed with the participant and any questions the participant had were addressed at that time. Documentation detailing vital study information was given to the participant. The documentation included information on the patient's follow up schedule and a symptom list, detailing the symptoms that would initiate an early follow up visit. This documentation also included the phone number of the Clinical Center and the names of the contact person, Principal Investigator, Study Coordinator and other relevant personnel.

The initial visit and assessment was used to collect all baseline data for a participant. Medical history information was collected and physical examination, and initial laboratory examinations (chest x-ray, blood studies, pulmonary function and sputum induction and skin tests) were performed. Further evaluation of the patient's eligibility was completed based on the data collected at the initial visit. The specific evaluations performed at the initial assessment included the following:

- History and physical examination
- Performance status
- Frontal and lateral view chest films
- Pulmonary function tests
- Hemoglobin, hematocrit, white blood cell, differential and platelet counts, erythrocyte sedimentation rate
- SMA 20 automated blood chemistry panel including serum lactate dehydrogenase
- HIV antibody test (previously known positive results from a qualified laboratory will be accepted)
- Serum specimens to be frozen at -70° C for subsequent analysis, possibly to include P-24 antigen and Beta-2 microglobulin
- Determination of lymphocyte subsets (helper/inducer, cytotoxic/suppressor and mature circulating T cells)
- Skin testing using tuberculin, and mumps antigens (read by study staff at 48 to 72 hours)
- Induced sputum examined for **P. carinni** and mycobacteria. Abnormalities detected during the initial assessment were evaluated as described in the diagnostic algorithms, Section 7.2.7 of the study protocol.

6. FOLLOW-UP EVALUATIONS

6.1 Frequency of Follow-up Evaluations

Subjects in Groups A and B were randomly assigned at the center using the computer program set up by the Clinical Coordinating Center to receive either routine (subgroups A-1 and B-1 and Group C) or intensive (subgroups A-2 and B-2) follow-up evaluations. Routine patients will be seen, at a minimum, every 6 months while intense patients will be evaluated, at a minimum, every 3 months.

6.2 Scheduled Follow-up Evaluations

During scheduled follow-up evaluations, a modified history and physical examination will be used and the same series of laboratory tests as performed in the initial evaluation will be obtained. For purposes of data analysis, subjects will be considered as belonging to the group to which they were originally assigned; however, subjects in Groups A-1, B-1 and C who develop an HIV-related pulmonary process will continue to be followed at the same intervals, except that pulmonary function tests will be performed at 6 month intervals. HIV seronegative controls will be followed at the same interval as Groups A-1 and B-1. The schedule for Group C subjects who become HIV seropositive will not change.

6.3 Diagnostic Evaluations

All diagnostic evaluations will be performed in the Clinical Centers and should follow the algorithms devised for purposes of the protocol. These algorithms are outlined in Section 7.2.7 of the study protocol.

A diagnostic evaluation may be triggered by the occurrence of symptoms (noted either at the time of a scheduled evaluation or between evaluations) or by chest radiograph detected on scheduled screening examinations. Standard methods to be used for each of the laboratory studies are described in the Manual For Diagnostic Procedures. All data will be recorded on standard forms. The symptoms that will trigger an evaluation include: (1) unexplained cough that persists for more than five days; (2) unexplained breathlessness (progressive over $\geq$ 5

days or severe ≥ 1 day); or (3) unexplained new onset of documented fever (≥ 38 °C oral ≥ 5 days). As part of the orientation and at each routine clinic visit, each study subject should be alerted to these symptoms and instructed to contact the study office should they develop. For each of these symptoms, if a reason other than a possible HIV-related pulmonary process can be found (e.g., bronchitis or asthma), the evaluation will not proceed. If the subject has fever with localizing extrapulmonary symptoms (e.g., diarrhea), and no pulmonary symptoms the evaluation will not proceed, and the subject will be referred to the primary physician. Any diagnoses resulting from such an episode will be entered at the subsequent visit on the Interval Visit Questionnaire.

6.4 Symptom Evaluation

When symptoms are reported the subject will be scheduled for a Symptom Evaluation Visit. These evaluations may be performed either on an inpatient or outpatient basis depending on the severity of symptoms and logistics of the evaluation.

The symptom evaluation visit will include:

- ***Interval Visit Questionnaire***

- ***Physical Examination***
  The physical examination will be the same as that performed for the routine visits.

The methods for procedures 3 through 7 are described in Appendix 5 of the study protocol.

- ***Chest Radiograph*** (frontal and lateral views)
  If a radiographic abnormality is found, further evaluation will proceed as outlined in Section 7.2.7 of the study protocol. Patients with cough and/or shortness of breath and no radiographic findings will have pulmonary function tests performed. Patients with fever but no specific pulmonary symptoms and normal chest films will be referred to their primary physicians.

NOTE: Based on the results of the history, physical examination, and chestradiograph, the physician investigator may make the clinical decision that the patient has acute bronchitis and elect to empirically treat him with antibiotics or simply observe him without proceeding to the items listed below. Otherwise, the following sequence of studies will be done:

- ***Pulmonary Function Tests*** (lung volumes, expiratory flows and the single breath diffusing capacity for carbon monoxide (DLCO)).
  In some instances, other abnormalities in pulmonary function such as airways obstruction or bronchitis may be found and account for the patient's symptoms.

- *67 Gallium Citrate Imaging*
  Lung imaging using ^{67}Ga citrate will be performed in symptomatic subjects with normal chest radiographs in whom pulmonary function abnormalities do not account for the symptoms. Subjects having pulmonary parenchymal uptake of the isotope will be evaluated with sputum induction. If there is no pulmonary ^{67}Ga uptake, the diagnostic evaluation will not proceed further, and the subject will be scheduled for a one month follow-up visit.

- ***Sputum Induction***
  All patients who have symptoms and an abnormality on chest film (as specified in Section 7.2.7 of the study protocol), or ^{67}Ga scan will have sputum induced.

- ***Bronchoscopic Procedures***
  All patients who have sputum induced and who do not have a pathogen identified will undergo bronchoscopy. Bronchoscopy will be performed within two weeks of the symptom evaluation visit.

- ***Blood Studies***
  All subjects who require additional evaluation because of chest radiographic abnormality, or abnormal GA scans will have the following blood tests:

  - T-cell subset determination (if not done within 3 months)
  - Complete blood count and differential, ESR SMAC (with LDH)
  - Serum to be frozen at -70°.

6.5 Chest Radiograph Abnormalities

Chest radiograph abnormalities may be discovered during symptom evaluation visits or scheduled clinic visits in subjects with or without associated symptoms. Radiographic abnormalities will be categorized as follows:

- diffuse process (including diffuse infiltration of any pattern, multiple ill-defined densities, multiple nodules or masses)
- focal interstitial pattern
- focal consolidation
- pleural effusion
- intrathoracic adenopathy
- focal cavity lesions
- solitary masses or nodules.

If a chest radiograph abnormality is noted, comparison with any available previous chest radiographs will be made. If the abnormality can be documented on radiographs performed ≥ 6 months previously, and is currently unchanged or has been evaluated previously and is unchanged, the evaluation will not proceed unless there are new symptoms.

The specific evaluations to be performed will depend on the kind of abnormality noted and on the clinical circumstances. The diagnostic approaches to be used for the most common kinds of abnormalities are shown in Section 7.2.7 of the study protocol.

6.6 Pulmonary Function Abnormalities

Pulmonary function abnormalities may be identified during scheduled or symptom evaluation visits in symptomatic and asymptomatic subjects with or without chest radiograph abnormalities. Asymptomatic subjects with abnormal spirometry will not require further diagnostic evaluation because airways obstruction or restriction have not been features of AIDS-related lung disease. The subject and the primary physician will be notified so that treatment (e.g., bronchodilators) can be started if appropriate. Any treatment that has been started will be recorded on the Subsequent Interval Visit Questionnaire.

6.7 Follow-up After a Non-Diagnostic Evaluation

If the evaluation does not provide a diagnosis and the clinical course is stable or improving, the patient will be observed. If he/she is severely ill or rapidly deteriorating further evaluation will be undertaken as defined by the clinical circumstances. All subjects who undergo a diagnostic evaluation for suspected pulmonary complications will return for a follow-up one month after hospital discharge for inpatients or one month after the symptom evaluation visit for outpatients.

Patients will be instructed to call the study office if they feel there is no symptomatic improvement after one week or sooner if they feel the symptoms are worsening. Patients who appear unreliable or ill will be telephoned by a member of the study team as frequently as needed to determine their progress until stable or improving.

Subjects who do not have symptoms or whose symptoms are improving will return for a one month follow-up visit. If the chest radiograph and pulmonary function tests demonstrate no deterioration the subject will revert to the routine protocol.

If the symptoms worsen or do not improve, the subject will return before the one month follow-up visit for a repeat physical examination, and chest radiograph. If the chest radiograph abnormality is worse, the patient will be considered to have deteriorated. Patients with documented deterioration will undergo repeat sputum induction, bronchoscopy and/or open lung biopsy. If no deterioration is documented, the subject will continue to be monitored by telephone calls and reevaluated as needed until deterioration, stability or improvement is noted. After the one month follow-up clinic visit, stable and improved patients will revert to the routine study protocol.

6.8 One Month Follow-up Visit

In addition to follow-up after evaluations for suspected pulmonary complications, one month follow-up visits will be scheduled for subjects with: (1) fever, no respiratory symptoms, and a normal chest radiograph; (2) respiratory symptoms and normal ^{67}Ga scan; (3) respiratory symptoms and other abnormalities in pulmonary function that account for the symptoms; and (4) abnormal chest film at initial visit.

For numbers 1-3 above the one month follow-up visit will include:

- ***Interval Visit Questionnaire.*** In addition, information from relevant medical records will be abstracted.
- ***Physical Examination.*** The physical examination will be the same as that performed for routine visits and will be recorded on the same form.
- ***Pulmonary Function Tests*** (lung volumes, expiratory flows and DLCO).
- ***Chest Radiographs*** (standard frontal and lateral views).
- ***Blood Tests.*** All subjects who had blood tests performed at the symptom evaluation visit will have the following blood tests:
    - T-cell subset determination (if not done within 3 months of the visit)
    - Complete blood count and differential, ESR, SMAC panel (including LDH)
    - Serum to be frozen at -70°.

For patients having abnormal chest films the evaluation will include only a repeat of the chest film. If the abnormality is stable or improved, no further evaluation will be performed. If it is worse, the evaluation will proceed.

A one month follow-up visit can substitute for a routine assessment if it is done ± 3 months for Groups A-1 and B-1 or ± 1 month for Groups A-2 and B-2 from the date of scheduled routine investigation, provided all routinely scheduled studies and tests are completed. This may in some cases require that in addition to the usual one month follow-up studies HIV antibody and skin test.

6.9 Diagnostic Algorithms

The following algorithms will be used in the diagnostic evaluations of patients with symptoms or findings:

Evaluations of Radiographic Abnormalities

Figure 2: Focal Consolidation

Figure 3: Pleural Effusion

Figure 4: Intrathoracic Adenopathy

Figure 5: Cavitary Lesions

Figure 6: Mass Lesions or Solitary Pulmonary Nodule

Figure 7: Diffuse Process of Focal Interstitial Pattern

6.10 Nonresponse

During the first (recruitment) year, subjects who did not appear for evaluation in spite of at least three telephone or mail reminders during a three month period were replaced by new enrollees. For any subject who does not return for scheduled evaluation, efforts will be made to ascertain his/her status at each of the originally scheduled intervals so long as he/she is not confirmed to be dead.

6.11 Management and Retention of the Cohort

Each Clinical Center developed its own administrative structure for managing the cohort and for retaining study subjects. The major means of ensuring the continued cooperation of the study subjects is personal contact with an individual from the study staff. This contact person should have frequent telephone communication with the subjects to inform and remind them of scheduled visits. The contact person is easily accessible to the subjects by telephone and

serves to facilitate their interactions with other study personnel and, where possible, with the subjects' sources of care.

Patients are sent reminders by mail before their scheduled follow up visits are to occur and notes after each visit. Other forms of contact such as newsletters and social gatherings may also be implemented to foster compliance and to provide the study with a ***face.*** Each center's plan for fostering compliance will be reviewed periodically by the Executive Committee. Data on missed visits is monitored throughout the study by the Clinical Coordinating Center as a control measure.

Compliance is also achieved by maintaining regular contact with the subject's primary physicians. At the time of enrollment in the study the subject's primary physician was provided with a written summary of the protocol and a schedule of follow-up visits. After each visit a summary of the results of the evaluation is sent to the physician (with the subject's authorization).

7. GENERAL INSTRUCTIONS ABOUT RECORDING DATA ON FORMS

7.1 General Instructions

Record all data in a legible manner. All names and words other than the signatures of the clinical center coordinators and examiners should be printed or typed in capital letters with one letter per box. Numbers should also be clearly written or typed with one number per box.

7.2 Patient ID and Identifying Information

Every subject enrolled in this study from each of the six U.S. centers and the 1 European center was assigned a unique 5 digit study identification number by RTI. RTI will not use the identification number assigned by the clinic. RTI's assigned identification number has the advantages of eliminating duplicate ID numbers between clinics, identifying the clinic from which the data came by adding a clinic digit to the ID number, and safeguarding against many types of errors including single digit transcription errors by adding a check digit.

The first digit of the patient identification number will be a center identification number. This digit will uniquely identify the center from which the data came. The center identification numbers are given below.

University of California, San Francisco San Francisco, California	1
Northwestern University Evanston, Illinois	2
Mount Sinai School of Medicine New York, New York	3
University of Medicine and Dentistry New Jersey Medical School Newark, New Jersey	4
University of California, Los Angeles Los Angeles, California	5
Henry Ford Hospital Detroit, Michigan	6

As an example of the RTI identification number assignment scheme, the very first patient enrolled at Henry Ford Hospital will have an ***ID Number*** of 6001 plus the check digit.

The next three digits of the identification number will be the sequence number for the subject within each center. In general, these numbers will be in the ascending order in which the subjects are enrolled in the study. However, certain combinations of these digits have been eliminated because they present problems in checking certain types of keying errors.

The last digit, the fifth in the identification number, is a check digit. The sole purpose of the check digit is to aid the Coordinating Center in checking for keying/transcription errors in the ID number. The check digit represents a letter(a thru I) that is generated by a computer algorithm at RTI. The algorithm uses the first four digits in the ID number as a basis for assigning the letter that is used as the check digit. When an ID is entered by a keyer, the algorithm computes the appropriate check digit and compares it to the check digit that is entered. If the ID was entered incorrectly, the check digit computed and the one in the ID number will not match and a flag will be raised indicating an error in the ID number.

7.3 Computer Generated Labels

The Clinical Coordinating Center will provide computer generated labels to the clinics, with the study subject's identification number printed on them. These labels will be organized on sheets and will be grouped together as a set. They may be peeled off to be used as needed. The labels are for use in the following contexts:

- for labelling on each of the forms
- for labelling the patient log
- for labelling biologic specimens.

Other unique patient identifying information such as name, address, social security number and physician information will be stored on a computer file to be maintained at the clinic. This computer file will also contain the RTI assigned identification number which will act as a link between the more confidential clinic file and the RTI data file.

7.4 Recording Dates

- ***Recording Complete Dates:*** Most forms have a question for dates. All complete dates that are recorded on any forms should be recorded in this manner. Numeric responses for day and year should be right justified and leading Ø's should be used when appropriate. Months should be recorded with three alpha characters. Valid abbreviations are: JAN, FEB, MAR, APR, MAY, JUN, JUL, AUG, SEP, OCT, NOV, and DEC.

Correct:	0 9	J U L	8 9	
Incorrect:	9	J U L	8 9	(no leading zero)
	9	J U L	8 9	(not right justified)
	0 7	9	8 9	(month/date switched)
	0 9	7	8 9	(month not alpha)

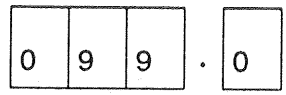
- ***Recording Incomplete Dates.*** In some cases, a complete date (day, month, year) will not be remembered by the study subject. In this case, record data for all fields that are known as specified above and leave all unknown fields blank.

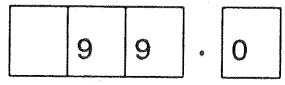
Examples

	J A N	8 9	Month and Year Known
		8 9	Year Known Only

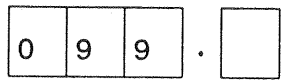
7.5 Numeric Responses

As with dates, numbers should be recorded using leading Ø's when necessary and should also have Ø's in the tenths and hundredths column. All numeric responses should also be right justified. For example, a subject's weight of 99kg would be correctly and incorrectly recorded as follows:

Correct: 

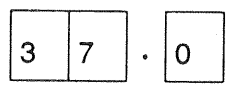
Incorrect: 

(no leading zero)

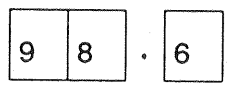
Incorrect: 

(no trailing zero in decimal spot)

- ***Measurement Units:*** Pay particular attention to the units in which all numeric responses are to be recorded. Always use the indicated units when measurements are requested and make sure the decimal is in the correct place. Be sure to use leading and trailing Ø's where appropriate. For example, when recording temperature, record as follows:

Correct: 

Celcius

Incorrect: 

Celsius (not recorded in Celsius)

- ***Rounding:*** All numeric responses will be rounded using the following rounding conventions.

  Responses with a fractional component should be rounded to the nearest integer.

  ex. 32 1/4 would be rounded to 32

  32 3/4 would be rounded to 33

  If the fractional component of the response is 1/2, round up for integer components that are even and round down for integer components that are odd.

  ex. 32 1/2 would be rounded to 33.

  31 1/2 would be rounded to 31.

  If the situation arises where a numeric response is less than 1, it should be recorded as a Ø.

- ***Missing Values:*** If a value can not be determined for a numeric response, it should be left blank.

7.6 Multiple Choice Responses

To record the answers to multiple choice type questions, place either a check (√) or a cross (X) (whichever method is chosen should be used consistently) in the box corresponding to the appropriate answer. For example, when answering a yes or no question such as:

IV Drug Use ☐ yes ☐ no

This question may be recorded as follows:

☒ yes ☐ no

or

☑ yes ☐ no

But, only one way of recording the data should be used.

7.7 Severity Scores

To record the severity score place a circle around the appropriate score. Be careful to circle only one response.

7.8 Specify Responses

Print clearly and legibly all entries that go in the places marked ***specify.*** These must be read and coded by people who do not necessarily have a medical background.

8. INSTRUCTIONS FOR COMPLETING STUDY FORMS

Study forms should be administered in quiet and comfortable environment where the interviewer and study participant's conversation cannot be overhead. Speaking in a comfortable, soft tone, the interviewer should offer some general information about the interviewing process and the type of information to be asked (e.g., I will ask you a series of questions about your background and your health. Some may not seem relevant to you however, all questions are relevant to the study.) The study subject should be instructed to respond to the best of their knowledge to all questions and to ask for clarification if they don't fully understand what is being asked by the interviewer. It is extremely important to remind the study subject that all information obtained from them will be kept strictly confidential.

When the study subject seems comfortable to being, ***the questionnaire should be administered by the interviewer.*** Ample time should be taken to ensure that the information collected is complete and accurate and to ensure that skip patterns and the logical flow of the questionnaire are followed. Questions or terminology within the forms that may be hard for the study participant to understand should be thoroughly explained.

Once the interview is completed, the questionnaire should be reviewed by the clinic coordinator for completeness and correctness. This should be done before the subject leaves the clinic so that any problems that arise can be corrected while the study subject is still present.

Once the questionnaire is deemed complete, the study subject should be thanked for their participation and reminded when their next scheduled visit is and about criteria that should warrant a visit before the next scheduled one (i.e., a symptom visit). A record of all completed interviews and forms should be kept to ensure that data is not misplaced and to keep inventory on who has been interviewed and what data has been collected. All completed forms should be kept in a specified secure place to ensure that data is not misplaced and to ensure that confidentiality will be maintained.

9. DATA ENTRY AND TRANSMISSION

9.1 Data Entry

Data entry programs for each of the study data forms have been developed at RTI using the Research Triangle Institute Data Entry system (RTIDE). This system allows for easy data entry and initial editing of study forms on microcomputers located at the clinical centers. Each study form's data entry program is stored on the hard disk of the center's microcomputer with a unique file name for each form which is used to invoke the screen entry program for the specific form. At the convenience of the data coordinator at the clinic, data that has been collected on paper forms is keyed using the (RTIDE) screen entry programs. As data is keyed, it is stored in form specific data files where it is later retrieved via data transfer programs that are executed by RTI's VAX computer. **NOTE: Detailed instructions on how to use the (RTIDE) data entry software can be found in the Pulmonary Aids Clinical Study Computer Reference Manual that has been provided to each Clinical Center.**

Some of the basic features of the RTIDE data entry software are given below.

- Field checks (only numeric data in numeric fields allowed)
- Required data item checks (data entry cannot proceed until a legitimated value is entered into that field)
- Range checks and/or valid value checks
- Checks for legitimate date values
- Within form logical consistency checks (e.g., proper data sequence for procedures reported in one form)
- Logical branching algorithms (e.g., if a procedure that is optional is not done, the data entry system will automatically skip over that section of the form)
- Check digit verification checks
- Log of keying errors.

In addition to the features above, all skip logic, range checks, and data consistency checks inherent in the form are included in the form program. The complexities that can be

programmed into the RTIDE package by CCC staff allow for tight control over the keying process.

Data can be keyed at the convenience of the clinical center staff, under the control of the data entry software. Although keyed at the convenience of the Clinical Center, keying should be completed as soon as possible after a subject's visit. Any errors detected by the data entry software or by the more complex editing which will be done at the CCC subsequently, will be easier to resolve when needed information is more readily available.

Each completed data form should be retained at the clinic in a subject folder and kept current with any revisions to the database. These files should be kept in a locked cabinet with access allowed to Clinical Center staff only.

9.2 Transmission of Data to the Clinical Coordinating Center

The transmission of data from the clinics to the Clinical Coordination Center is accomplished using the BLAST communications software package. Each clinic microcomputer is outfitted with BLAST communications software and a 1200 baud modem. Data files to be transferred reside in single form specific files on the microcomputer's hard disk at each center. At the end of the day, Friday of each week, the clinic staff will set their microcomputer up for polling. Following this, all subsequent transmission activities will be automatic. **NOTE: The procedures for setting the microcomputer up for polling and for using the BLAST communications software are documented in the Pulmonary Aids Clinical Study Computer Reference Manual.**

During the evening hours, the BLAST communications package located on RTI's in-house VAX executes a program which continuously reads the VAX's system clock and calender and at predetermined times, dials phone numbers attached to the Clinical Center's microcomputers. On connection to a specific Clinical Center's microcomputer, the communications package logs into the microprocessor, locates the screen entry form data files and transmits the data to the CCC VAX computer.

At the end of the transmission, the communications package terminates the phone connection and posts a message on the screen for the data coordinator to read, giving the results of the transmission, including any instructions for retransmission of failed transmissions.

Session transmission, including dialup, requires less than 3 minutes per Clinical Center. The communication process is an ***error-free*** process. After every 128- or 256-character record transmitted, the sending and receiving computers will compare the record. If they do not agree, the record will be resent until it is correct or until a 10 minute window for transmission is surpassed. In the latter case, the Clinical Center will be notified in the form of a computer message, before the phone connection is terminated that transmission did not proceed properly.

9.3 Backup Procedures for Data Entry Files

Backup files of all newly keyed data is automatically produced during the transmission session. After the transmission is complete, the original copy of the keyed data is deleted. These backups are then be used if problems are discovered in the transmission of the original files.

10. EDITING PROCEDURES FOR STUDY DATA

10.1 Data Editing Procedures

After study data has passed the extensive processing edits of the RTIDE data entry software, more complex and longitudinal edits are performed. ***(NOTE: For a complete list of all edits, refer to the Pulmonary Aids Clinical Study Edit Specification Users Manual.)*** As part of the editing process, edit error resolution forms are produced by staff at the CCC documenting all suspected errors to be resolved by Clinical Center staff. These forms are generated on a monthly basis and are sent to the centers for resolution. Center staff are to indicate on the form either a change in the data specified or that data are uncorrectable or need no correction. The resolved forms are then sent to the Clinical Coordinating Center where changes are made to the study database by CCC programming staff. Data that are determined to be correct but still fail study edit checks remain flagged in the database and are not removed from the study data files. Subsequent analysis of flagged data frequently points to the need to modify data collection procedures and/or edit specifications. Later analyses of edit resolution data provides a means of assessing Clinical Center performance with respect to proper data collection.

10.2 EDIT MONITORING PROCEDURES

A complex computer based monitoring system is used by the CCC staff to account for outstanding error resolutions. All resulting data changes are recorded at the Clinical Centers on ***edit error resolution forms*** and are sent to the CCC for processing and implementation into the database. These forms along with an edit data computer file, provide an audit trail that includes clinic ID, study participant ID, date, time, variable name, old field value, and new value. In addition, monitoring files are maintained that indicate the inventory of all edit resolutions that are sent to the centers, the response time to return the edits, and an indication of whether the edit instituted a change in the study database. This approach not only documents all changes to the database, but also enables an ongoing analysis of the success/failure of data collection and data processing.

11. DATA COLLECTION MONITORING AND REPORTS

11.1 Data Collection Monitoring

The Clinical Coordinating Center(CCC) monitors both the data collection activities within the Clinical Centers and the data processing tasks at the CCC. Monitoring is accomplished by establishing a schedule for data collection for each participant enrolled in the study. This schedule allows the CCC to track a patients visits and thus to determine when the CCC should expect to receive baseline data on a newly enrolled patient and follow-up data on currently enrolled patients. In addition, the CCC monitors the receipt of study forms after each patient visit to ensure that all proper forms were received. Various monitoring reports are generated in a timely fashion to keep the center apprised of the current status of all patients.

All editing and data correction activities at the CCC will be monitored so that the status of any record in the CCC database can be obtained immediately.

11.2 Data Collection Reports

A variety of reports required by NHLBI/NIAID are prepared on a periodic basis for the duration of the study. These required reports include financial reports, subcontracting reports, reports showing the progress of recruitment, quarterly reports describing current study data, and various other reports. In addition, in-depth Steering Committee reports and Data Safety and Monitoring Board reports are produced twice each year. These reports describe current study data in great detail and act as an informative document for the preparation of abstracts and manuscripts.

In addition, the Clinical Coordinating Center assumes the responsibility for several other types of periodic (e.g., monthly) computer-generated reports. These reports are adaptations of data monitoring system reports developed at RTI and used successfully in other studies in which RTI has served as a coordinating center.

The following is a current list of the reports being prepared by the CCC and the time frames on which they are delivered. A brief description of each of these reports follows.

Periodic Reports Sent by Coordinating Center

Polling	Weekly polling on Friday
Polling Forms Received List	Weekly - Monday
Delinquency Report	Monthly (1st Tues or Wed)
Termination	Monthly (1st Tues or Wed)
% of Completed Visits	Monthly (1st Tues or Wed)
Key Stroke Error	Part of DSMB Report, beginning in March
DMB/Steering Committee	As Needed
Compliance Report	Monthly (1st Tues. or Wed.)

The periodic reports will be designed to assist the data coordinators in the collection of data outlined in the protocol. Additionally, the reports will summarize the successes and failure of data collection for the Clinical Coordinating Center, the Program Offices at NHLBI and NIAID, and principal investigators.

12. QUALITY ASSURANCE PROCEDURES

12.1 Medical Procedures

Many of the medical procedures performed in this study will be subjected to some degree of quality assurance (QA).

There will be two kinds of QA; one (described in Section A) in which a sample of chest radiographs, induced sputum slides and pathology slides will be circulated to all participating centers to determine any variability in interpretation among the readers at each center. In the other kind (Section B), samples of chest radiographs, induced sputum slides and pathology slides from each center will be sent to a specified QA specialist for interpretation. Discordant interpretations will be summarized and presented to the appropriate forum.

As described below in Section A (Reader Variability), representative samples of slides and chest radiographs will be selected by a QA specialist and sent to each center's Data Coordinator (DC) to pass on to the designated specialist for evaluation. For these procedures, a center order listing (01 is San Francisco General Hospital (SFGH), 02 is Northwestern, etc.) will be used so the samples can be forwarded directly to the next center on the list. The QA specialist will send the sample(s) to the first center, and that center should complete its evaluation on the specific QA Forms. QA x-ray forms should be speedily sent to the QA Coordinator at RTI for keying; QA pathology Forms should be returned to the designated microbiologist at San Francisco General Hospital (SFGH), and the sample(s) should be forwarded to the next center on the list. ***All of this should be done within a one week time period.*** When the last center has completed its interpretation, the specimen(s) should be returned to the designated QA person. The DC should keep track of all specimens that come to the center and make certain that (1) incoming samples are interpreted on the QA forms and forwarded to the next indicated center and (2) that the QA Forms are sent to RTI, within ***a week***.

For procedures described in Section B (Discordant Interpretations), in which each center sends samples to the QA specialist for interpretation, the DC should be the person responsible for keeping track of all the specimens that were selected by the CC for review. She must be certain that the patient ID is the only identifying characteristic on a sample before it is sent out

for QA review. If there are any problems (shipping, labeling, etc.) it will be the DC who will be contacted.

The QA specialist should be able to complete the evaluation, send the specimens back to the originating center, and send the QA Form to the QA Coordinator within one week.

Induced Sputum

- ***Reader Variability:*** Every year SFGH's Microbiology Laboratory will select ten representative Induced Sputum slides (positive and negative) and send them to each participating center's laboratory for interpretation. These slides will be labeled with an identification number and the type of stain used. The center's microbiologist will record his/her interpretation of the slides on the Quality Assurance Specimen Evaluation Form (QA-V) and send the completed form back to SFGH for interpretation agreement. The slides should be forwarded to the next center on the list, and when the last center has completed their interpretation, the slides should be returned to SFGH. All evaluations will be compared for variability among interpretations.
- ***Discordant Evaluations:*** At the same time, each clinical center will send one stained (each center should use their customary staining method) PCP positive slide to SFGH. The slide should be labeled with the patient's ID Number, the date the specimen was obtained and the stain used on the slide; no patient's name should ever appear on a slide. Slides may be packaged and shipped to SFGH in the manner customarily used at each center. Microbiologists at SFGH will report their agreement (or lack of agreement) on the same QA_V Forms, and return the slide to the center from which they came. The completed forms should be sent to the QA coordinator at RTI. The forms will be keyed and a record of all discordant interpretations will be presented to the appropriate forum.

Radiology

- ***Reader Variability:*** Once a year the QA Radiologist will select 25 representative chest radiographs (positive and negative) for circulation among the clinical centers. If at all possible the original film should be used and a duplicate kept in

the patient's file. If the film has the patient's name imprinted on it, it should be covered with tape, and only an identification number and the date of x-ray should be seen on the film. The center's radiologist will record his/her interpretation of the film on a Quality Assurance Radiology Form (QA-R), and then send the form to RTI for keying. All interpretations will be compared for variability among readers.

- ***Discordant Interpretations:*** Ten percent of all chest radiographs taken at specific visits (positive and negative) will be selected by the CC for QA. X-rays from two specified consecutive visits for each ID selected by the CC will be to the QA radiologist. The ***most recent*** x-ray will be interpreted and then compared to the previous x-ray for changes. These QA readings will be done on a quarterly basis. Centers should send the films that best represents the results of their findings from the specified visit to the designated QA radiologist. Whenever possible the original films should be sent and a duplicate maintained in the patient's file. Films must be labeled with the patient's ID number and the date the films were taken; any evidence of the patient's name on the film must be completely obliterated with tape. The radiographs should be packaged and mailed in the center's usual way and sent to the QA radiologist. The QA radiologist will review the films, record his/her findings on the QA-R form, and send the form to the QA coordinator at RTI. The films should be returned to the originating center. Forms will be keyed at the CC and a record of all discordant interpretations as well as problems noted in the quality of technique will be presented to the proper forum.

Pathology

- ***Reader Variability:*** The selected QA pathologist will select and send a representative sample of 10 histologic, and 10 cytologic slides per year to the designated study pathologists at each center. The slides should be labeled with an identification number, the type and site of the specimen and the stain used. The pathologist will ***review the materials according to the center's usual procedure and*** record his/her slide interpretation on Form QA-V and then send the form to the CC for keying. Slides should be sent to the next center on the list and, after they are reviewed by the last center, they should be returned to the QA

pathologist. All interpretations will be compared for variability among interpretations.

- ***Discordant Interpretations:*** Ten percent of all histologic, and cytologic slides ***from specific visits*** will be selected by the CC for quality assurance. All requested slides (or duplicates) clearly labeled with the the patient ID, date specimen was obtained, type and site of specimen, and the stain used (but with no patient name marking) should be packaged and sent in the center's usual manner to the QA pathologist. The QA pathologist will record his/her interpretation on the QA-V form and send it to RTI for keying. The slides will be returned to the originating center. The QA reading will be compared to the original reading and a record of all discordant interpretations as well as notations of problems noted in the quality of specimens will be presented to the appropriate forum.

Pulmonary Function Tests

- Pulmonary function tests will be performed on the same two members of the site visit team at every site visit. The original tracings and the Quality Assurance Pulmonary Test Forms (QA-F) should be sent to RTI; copies should be made and maintained at the clinical center. DLCO measurements will be evaluated for consistency.
- DLCO measurements on a healthy staff member will be done on a quarterly basis at each center on all equipment used. The results should be sent to RTI. Copies of the tracing should be kept at the center.

Gallium Scans

Fifty percent of all 67 Gallium Scans performed will be selected by the CC for QA. The centers will send copies (not to be returned) of the scan labeled only with the patient's ID number to the QA nuclear medicine specialist. He/she will do an independent qualitative reading and record the findings on the QA Gallium Form (QA-G) and also check the quality of the scan. The form will be sent to the CC for keying and a record of all discordant readings will be maintained by the QA coordinator.

12.6 Rekeying of Study Data

On a quarterly basis, a sample of 3% of the forms received from each center will be selected to be QA rekeyed. A list of these forms is sent to the Clinical Centers to be re-keyed using a special re-key option of the RTIDE data entry system. This re-key option keeps regular study data and re-key data separated in the study data files. The QA data files are then compared against the data on the study database. From this comparison, keystroke error rates are computed by center and for all centers overall. Any discrepancies found between the two files are followed up by CCC staff to determine which value should reside in the database. This data is then reported at the Steering Committee meetings.

13. SERUM BANKING PROCEDURES

13.1 General Information

For the Pulmonary Aids Clinical Study, serum should be collected at the initial visit, all regularly scheduled follow-up visits, and during symptom generated visits in which additional evaluation is required because of chest radiographic abnormalities, reduced DLCO values or abnormal GA scans.

The mechanics of how serum is processed is up to the individual center. Procedures for collecting and storing serum at the centers should be documented in a memo and sent to RTI. This memo should include information on who collects the serum, where it is collected, where it is temporarily stored before shipping, who is responsible for filling out data forms and who should be contacted for questions or problems.

The serum repository for the Pulmonary Aids Clinical Study is ERC Bioservices Corporation. The repository will be responsible for sending you the necessary equipment for serum shipping. A shipping container, containing serum storage boxes filled with empty cryotubes will be sent to each center as needed. Each serum storage box will contain 81 empty cryotubes laid out in a 9x9 grid. Each cryotube is designed to hold 1 ml of serum. A postcard will be included for you to return to Flow Laboratories acknowledging receipt of the container in good condition.

Enough blood should be drawn at each specified visit so that four 1 ml cryotubes of serum can be obtained and saved by the center for shipment to the repository each time the patient comes to the center for one of the visits described above. These cryotubes should be labeled using the pink study ID label (to avoid confusion with other ID labels) provided to the centers by RTI. The date the blood was taken should be written on the label.

13.2 Shipping Forms

A ***SERUM SAMPLES LOG*** and a ***SERUM SHIPPING FORM*** have been devised to keep track of serum storage and shipment. The serum samples log is designed to keep track of the location of individual serum cryotubes within a serum storage box and is to be completed at the center. The serum shipping form is designed for the repository to identify the serum storage

boxes in the shipping container, to identify the condition of the serum shipped, and to identify the storage location of the serum storage boxes at the repository. The serum shipping form is to be completed by both the center and the repository. Examples of these two forms are attached and instructions on completing the forms will follow.

13.3 Box Labeling

It is very important that the serum storage box be labeled in the same manner as the serum samples log. This will help ensure that mistakes are not made when a serum cryotube location in a particular slot of the serum storage box is recorded on the serum samples log. Any writing that is done on the serum storage box or on the cryotube labels should be with a specialized freezer pen.

To label the serum storage box, remove the top and set it aside. All labeling of the serum storage box will be done on the sides of the bottom part of the box and not on the lid of the box. To begin, label one of the sides as FRONT (Figure 1). This will act as a reference point to keep the orientation of the box the same while it's being filled with cryotubes. On the side that you labeled FRONT, the columns of the grid should be labeled alphabetically, A thru J omitting the letter I. Begin with A in the front left corner of the box and proceed to J in the front right corner of the box (Figure 2). With the FRONT side of the box still facing you, locate the left side of the box. This side will be used to label the rows of the box. Number the rows of the grid from 1 to 9 starting in the upper left corner of the box and proceeding to the lower left corner (Figure 3). This labeling should now match that of the serum samples log. The slot in the upper left corner of the box should be slot A1. The lower right slot should be J9. This labeling should be done immediately upon receipt of the serum storage boxes from the repository.

The four samples for each patient should be kept together and the cryotubes placed in the storage box such that A1 is inserted first, then A2, A3 etc.

13.4 Data Form Completion at the Center

For each serum storage box to be shipped, a separate serum samples log must be completed that identifies the ID number and date of collection of all the serum specimens

located within the serum storage boxes to be shipped. Please note that no more than six serum storage boxes may be sent at one time to the repository.

The serum samples log contains 81 cells laid out in 9 rows and 9 columns that match the 9x9 grid of the serum storage box you will be using. The FRONT label on the serum storage box should be oriented to correspond to the FRONT label on the serum samples log. This is done to ensure that the serum cryotube location in the box matches its location on the log sheet. Check the box to ensure that the grid labeling is on the box as specified in the Box Labeling section of this document. If the labeling has not been completed, refer to the Box Labeling section and complete the labeling before proceeding.

To complete the serum samples log, place the pink ID label corresponding to the ID of the serum specimen located in it's particular cell of the serum storage box on it's corresponding cell on the serum samples log and write below it the date the serum was collected. Be sure that the IDs entered in the squares of the serum samples log match the IDs of the cryotubes in their respective slots of the serum storage box.

Once all serum samples logs are completed, the center can complete it's section of the serum shipping form and the serum box numbers can be assigned. To complete the serum shipping form, enter the appropriate center name and the shipping container number in the space provided on the form. The serum shipping container number is a number used to link the individual serum storage boxes with their shipping container. The shipping container number should be placed on the front and top of the shipping container as well as in the space provided on the serum shipping form. The number is a 4 digit number that is assigned at the clinic. The first digit is the center number and the last three digits are a sequential box number (i.e., 1001 is the first shipping container at San Francisco). The shipping container numbers will be printed on labels provided by RTI and should be used whenever possible.

Serum storage boxes that are shipped together in a particular shipping container should be labeled with a five digit number where the first four digits are the shipping container number and the last digit is a sequence number from 1 to 6 depending on how many serum storage boxes are sent in the shipment (i.e. 1001-1, 1001-2,1001-3, ...). This number is used to uniquely identify the individual serum storage boxes within their shipping container. One of these labels should go on the top of the serum storage box and the other should go on the front

bottom right side of the serum storage box. A third label should go on the serum storage log in the upper right corner to denote the box number. Figures 4 and 5 provide illustrations of the placement of these labels on their respective boxes and forms. Once the box numbers are assigned and labels are affixed, enter the date the serum was shipped to the repository and the number of serum storage boxes that were shipped.

13.5 Shipping Procedures

Shipping Forms

When serum is shipped to the repository, a copy of the serum samples log should be enclosed in its corresponding serum storage box. One copy of the serum samples log and a copy of the serum shipping form should be stapled together, placed in an envelope, and taped to the outside of the shipping container. Keep one copy of these forms for your records and send another copy to RTI and to the Program Office, NHLBI.

Shipping Storage Boxes

It is recommended that you accumulate five or six storage boxes filled with cryotubes before sending a shipment to the repository. Specimen boxes should be wrapped in an absorbent and cushioned material such as a Pampers diaper. This helps in conforming to the requirement that there be enough absorbency in the package to handle the entire fluid volume. Then place the wrapped box in a heavy plastic bag that can be heat sealed or made water-tight in some manner. Zip-lock plastic bags are proven and convenient.

Shipping Containers

Dry ice should be placed in the bottom of the shipping container. The wrapped and sealed specimen boxes should then be placed on the dry ice, and more dry ice should be added around the sides of the sealed specimen boxes. Enough cushioning or padding material should be added to prevent drastic shifting as the ice evaporates. On the outside of the shipping container, securely place the following labels:

1. A return address label containing your name, address and phone number.

2. An address label for the repository:

Dr. Susan Stern
ERC Bioservices Corp.
685 Losstrand Lane
Rockville, Maryland 20850
(301) 340-0245

* Ms. Stern's phone number should be put on the lower left corner of the address label as a backup in case of problems.

SHIPMENTS ARE TO BE MADE ONLY ON MONDAY OR TUESDAY. Containers must be shipped prepaid for overnight delivery. Carriers suggested are Federal Express, Airbourne Express, Emery or Burlington Air Express. Labeling must be in accordance with the carrier's requirements. ***ONCE THE SERUM HAS BEEN SHIPPED, DR. SUSAN STERN AT THE REPOSITORY SHOULD BE NOTIFIED THAT THE SHIPMENT IS IN ROUTE.***

13.6 Repository Responsibilities

Once the shipment arrives at the repository, Dr. Stern will alert the center that it has arrived. Upon receipt, Dr. Stern will inspect the shipment to insure that no damage has occurred during shipping and complete the repository section of the serum shipping form. The date the samples were received at the repository should be recorded in the space provided on the shipping form. Each individual serum storage box should then be checked to ensure that no damage has occurred to the serum, that the serum was received intact and frozen, and that all samples on the serum samples log are present and in their correct cell within the serum storage box. The storage location of each serum storage box at the repository should also be recorded on the shipping form. Any damaged serum samples and samples recorded on the serum samples log that don't match their location within the serum storage box should be noted on the serum samples log by circling the entry and noting the problem with that entry on the form.

Once all serum storage boxes have been inspected and stored, the original copy of the shipping form and it's corresponding serum samples logs should be sent to Judy Katzin at Research Triangle Institute. A copy of the forms should also be sent to Dr. Tony Kalica at the NHLBI offices and a copy should be kept at the repository.

PULMONARY AIDS CLINICAL STUDY
SERUM SHIPPING FORM

****

FOR RTI USE ONLY

Day Month Year

****

CENTER NAME: _____ SHIPPING LABEL NUMBER | | | | |

Day Month Year

DATE SHIPPED: ..... | | | | | | | |

NUMER OF SERUM STORAGE BOXES SHIPPED: ..... | |

****

FOR ERC BIOSERVICES USE ONLY

~~TO BE COMPLETED BY FLOW LABORATORIES, INC. ONLY~~

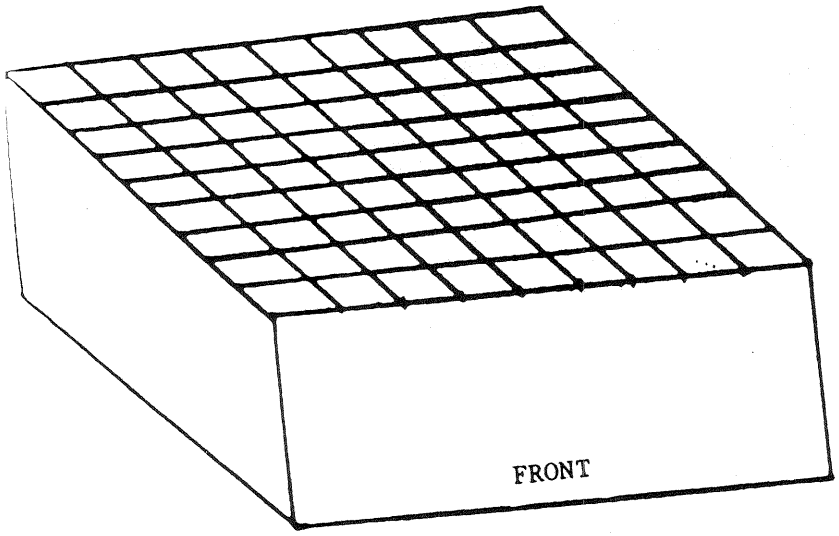
DAY MONTH YEAR

DATE RECEIVED: | | | | | | | |

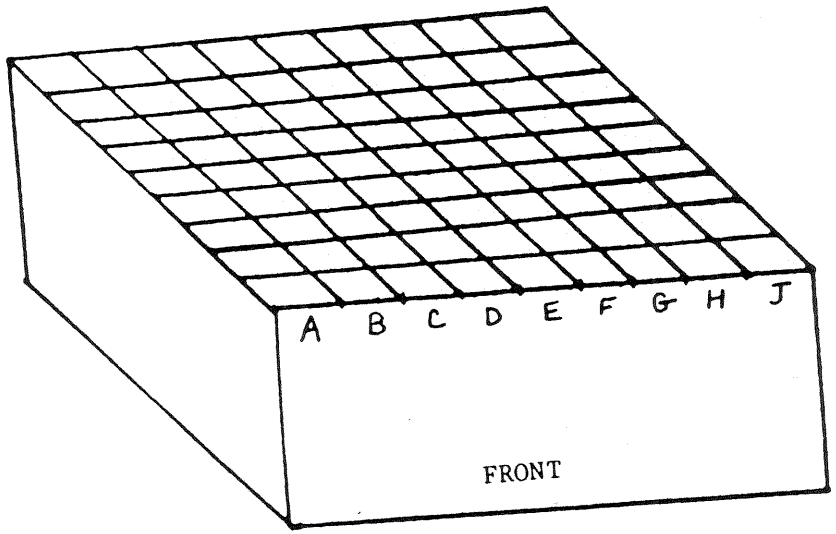
BOX NUMBER IN SHIPMENT	RECEIVED INTACT AND FROZEN		ALL SAMPLES PRESENT		STORAGE LOCATION		
	YES	NO	YES	NO	FREEZER	RACK	BOX
1	☐ 01	☐ 02	☐ 01	☐ 02	☐☐	☐☐	☐☐
2	☐ 01	☐ 02	☐ 01	☐ 02	☐☐	☐☐	☐☐
3	☐ 01	☐ 02	☐ 01	☐ 02	☐☐	☐☐	☐☐
4	☐ 01	☐ 02	☐ 01	☐ 02	☐☐	☐☐	☐☐
5	☐ 01	☐ 02	☐ 01	☐ 02	☐☐	☐☐	☐☐
6	☐ 01	☐ 02	☐ 01	☐ 02	☐☐	☐☐	☐☐

****

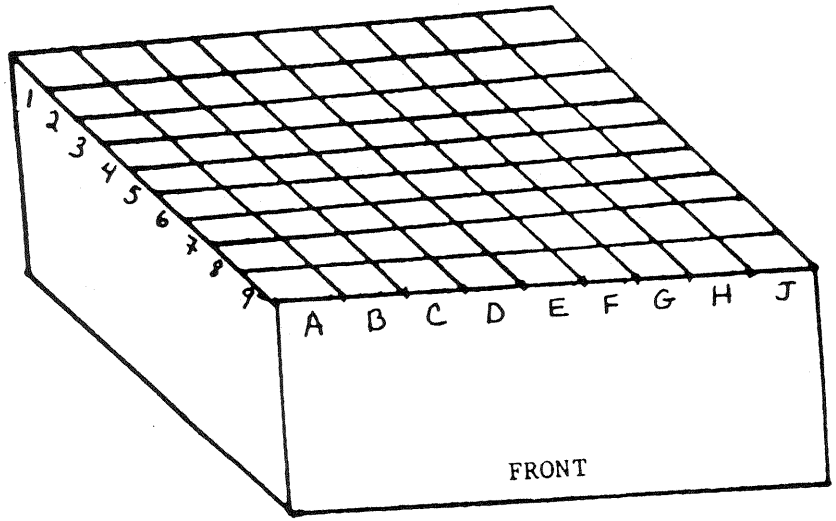
SERUM STORAGE BOX
LABELING PROCEDURES



(Figure 1)

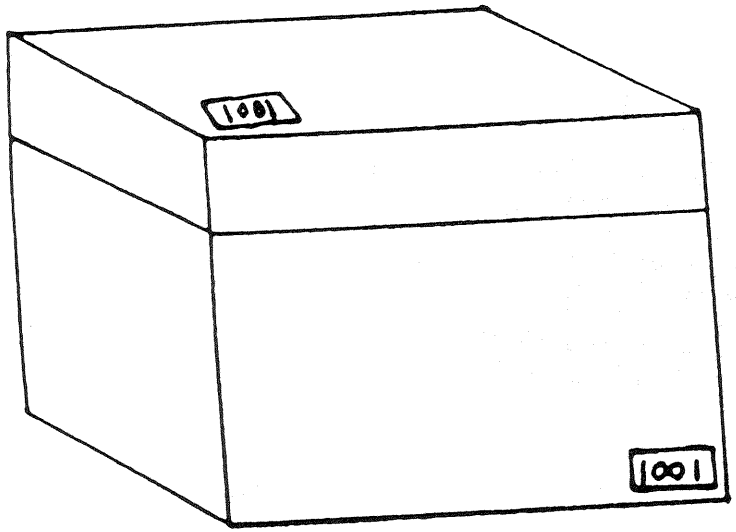


(Figure 2)



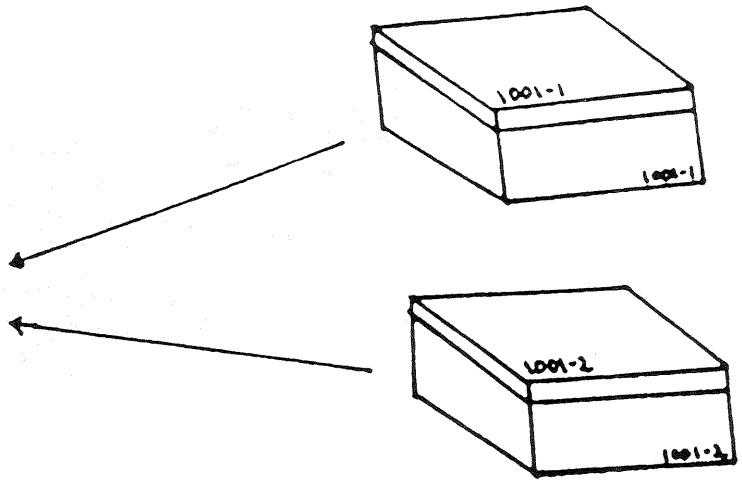
(Figure 3)
COMPLETED BOX

SHIPPING CONTAINER



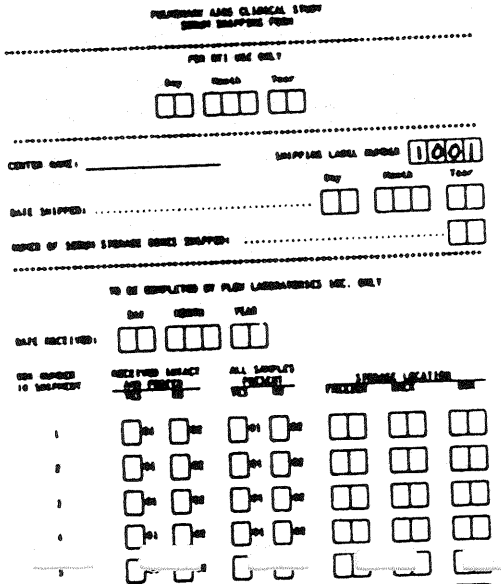
(Figure 4)

SERUM STORAGE BOXES

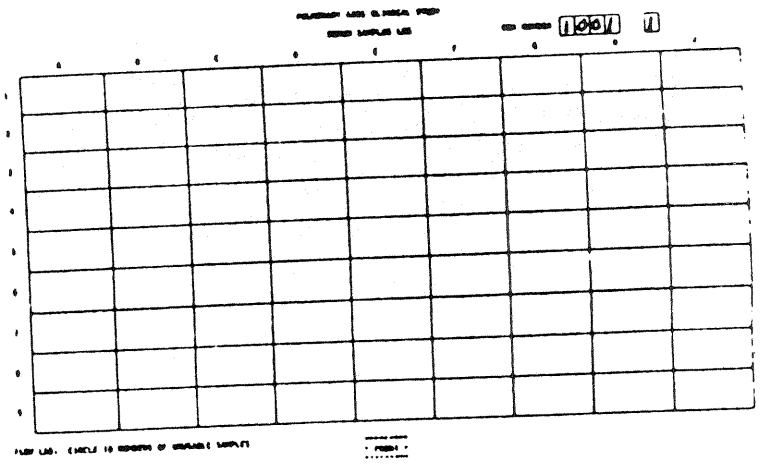


(Figure 5)

SERUM SHIPPING FORM



SERUM SAMPLES LOG



PULMONARY AIDS CLINICAL STUDY

SERUM SAMPLES LOG

BOX NUMBER ☐☐☐☐ - ☐

	A	B	C	D	E	F	G	H	J
1									
2									
3									
4									
5									
6									
7									
8									
9									

FLOW LAB: CIRCLE ID NUMBERS OF UNUSABLE SAMPLES

****
* FRONT *
****