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PROTOCOL

PROJECT RANCH HAND II

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EPIDEMIOLOGIC INVESTIGATION OF HEALTH EFFECTS IN AIR FORCE PERSONNEL FOLLOWING EXPOSURE TO "HERBICIDE ORANGE"

MATCHED COHORT DESIGN

PREPARED BY
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PREPARED FOR
PEER REVIEW AGENCIES
NATIONAL ACADEMY OF SCIENCES

AIR FORCE WORKING PAPER

PROJECT RANCH HAND II

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Glossary of Abbreviations

ABBREVIATION	DEFINITION
AFSC	Air Force Specialty Code
ALK PHOS	Alkaline Phosphatase
AV-GAS	Leaded Aviation Fuel (Reciprocating Engine)
BUN	Blood Urea Nitrogen
C-7	USAF Cargo Aircraft, 2 engine, Propeller, Reciprocating
C-123	USAF Cargo Aircraft, 2 engine, Propeller, Reciprocating
C-130	USAF Cargo Aircraft, 4 engines,Turbo-Propeller-Jet
CBC	Complete Blood Count
CPK	Creatine Phosphokinase
CSF	Cerebrospinal Fluid
DNA	Deoxyribonucleic acid
DOD	Department of Defense
2,4-D	2,4-dichlorophenoxyacetic acid
ECG	Electrocardiogram
EPA	Environmental Protection Agency
FBS	Fasting Blood Suger
FSH	Follicle Stimulating Hormone
G.I.	Gastrointestinal
GAO	General Accounting Office
GGTP	Glutaryl-glutamic Transpeptidase
HDL	High Density Lipid
Herbicide Orange	Mixture of 2,4-D and 2,4,5-T contaminated with TCDD
Herbicide Pink, Purple, Green	Other 2,4,5-T/TCDD-containing herbicides
JP-4	Jet Fuel
LDH	Lactose Dehydrogenase
LD_{50}	(Median) Lethal Dose for 50% of Tested Animals
LH	Luteinizing Hormone
NCI	National Cancer Institute
MMPI	Minnesota Multiphasic Personality Inventory
PACER HO	Code Name for the Herbicide Incineration Project
PACER IVY	Code Name for the Movement and Storage of Herbicides at Johnston Island
RANCH HAND	USAF Organizational Code Name for the Defoliation Operations
RBC	Red Blood Cell
RIA	Radio-immuno Assay
RVN	Republic of Vietnam
SGOT	Serum Glutamic Oxaloacetic Transaminase
SGPT	Serum Glutamic Pyruvic Transaminase

ABBREVIATION	DEFINITION
SMR	Standardized Mortality Ratio
SSS	Sensation Seeking Scale
TCDD	2,3,7,8-tetrachlorodibenzo-p-dioxin
TLV	Threshold Limit Value
2,4,5-T	2,4,5-trichlorophenoxyacetic acid
USAF	United States Air Force
USAFSAM	United States Air Force School of Aerospace Medicine
USSR	Union of Soviet Socialist Republics
VA	Veterans Administration
VDRL/FTA	Serological Tests for Syphilis
WAIS	Wechsler Adult Intelligence Scale
WRAT	Wide Range Achievement Test

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PROJECT RANCH HAND II

Executive Summary of Protocol

The Air Force has made the commitment to Congress and to the White House to conduct an epidemiologic study of possible health effects in Air Force personnel (RANCH HAND) who conducted aerial herbicide missions in Vietnam. The purpose of this investigation is to determine whether long-term health effects exist and can be attributed to occupational exposure to Herbicide Orange. The extensive use of herbicides in Vietnam between 1962 and 1970 was terminated when it became known that a contaminant, TCDD, was present in 2,4,5-T-containing herbicides and that this contaminant caused congenital abnormalities when administered to pregnant rodents. Subsequent extensive research into the toxicity of TCDD in animals remains equivocal. Presently, the potential for teratogenicity and carcinogenicity of TCDD is significant but appears to be species specific. The scientific literature on the toxicity of the components of Herbicide Orange reveals that the two main ingredients, 2,4-D and 2,4,5-T, have extremely low toxicity, distinctly different in nature than TCDD. TCDD has been shown to be embryotoxic at markedly lower doses in animals. Only recently have comprehensive prospective studies in humans been undertaken. Most previous epidemiological studies dealing with TCDD exposure in humans have suffered from weaknesses in design and statistical power. These studies have only validated a link between TCDD exposure and the subsequent development of chloracne. The public's perception of the toxicity of Herbicide Orange/TCDD is generally different than that of the scientific community. A review of 625 veteran claims submitted to the Veterans Administration supports this fact and reveals an awesome spectrum of alleged symptoms and diseases.

This study design incorporates a matched cohort design placed in a non-concurrent prospective setting (incorporating mortality, morbidity, and follow-up studies). Detailed computer searches of Air Force personnel records, with several cross-referencing techniques, will ensure total ascertainment of the RANCH HAND population. Since there was a documented higher concentration of TCDD contamination prior to 1965, this factor will be incorporated in the development of an exposure index. A group of C-130 crewmembers and support personnel will form the control group. They will be matched to RANCH HAND personnel for the variables of age, AFSC, length of time in Vietnam, and race. The ten best matches for each exposed subject will form a control set and they will be randomly ordered. In the analysis of mortality, each exposed subject and a 50% random selection from each control set will be followed yearly. The first of the randomized mortality controls will be selected and entered into the questionnaire and physical examination phases of the study. If this primary control drops out, for reasons other than death, the next control will be selected from the control set so that both statistical power and loss to study bias in the follow-up

study may be improved. All RANCH HAND personnel and their primary controls will be asked to complete a telephone questionnaire and will be offered a comprehensive physical examination, with special emphasis being placed on dermatologic, neuropsychiatric, hepatic, reproductive and neoplastic conditions. An adaptive physical examination and questionnaire will be developed to be used in years three and five of the follow-up study, which is set for a minimum of five years duration. The unusual capabilities of the USAF in the collection, storage, and retrieval of data will allow integration of the various data collection methods used throughout the study. Expected biases and study difficulties include differences in group accountability, risk taking behavior bias in the volunteer RANCH HAND group, response bias, interviewer bias, loss to study difficulties, and variability of procedures performed, as well as the political overlay of this effort.

Since this study is dealing with unknown clinical endpoints, determination of a disease state by statistical methodology is a prime thrust of the investigation. Inferences about a disease state will be developed by identifying symptom complexes or physical findings which in themselves may represent disease. By comparison of symptoms, signs and laboratory tests within and between groups, a logical decision-making scheme can be utilized to calculate relative risks from baseline data. If appropriate, these results will be used to sharpen adaptive approaches in the follow-up study. By the use of combinational and correlational analysis, statements about the probability of a disease state, a subclinical state, and/or over-reporting bias will be attempted. In addition, the application of regression techniques to a normalized exposure index among exposed individuals exhibiting symptoms and/or signs will also assist in the determination of a disease state. Beyond these pair-wise and group comparisons, newer techniques of pattern recognition, such as Factor Analysis and Cluster Theory, are being considered in order to achieve a more automatic and objective analysis. Mortality data will be analyzed using several different approaches, including age and age-disease specific rates, standardized mortality rates and modified life table approaches, as well as more sophisticated logistic and multiplicative models (survival analysis). Analysis of questionnaire and physical examination data will utilize log-linear models to verify the appropriateness of the standard statistical methodologies (e.g., McNemar's test for dichotomous rates). Polytomous or categorical findings will be analyzed by the use of log-linear models. Continuous variables will undergo covariance analysis to remove non-controlled effects, followed by the use of a paired difference statistic. Some data will naturally fall into groups (e.g., fertility/reproduction, liver function tests) in which case, multiway contingency table analysis or extensions of the generalized linear model analyses will be used.

PROJECT RANCH HAND II

EPIDEMIOLOGIC INVESTIGATION OF HEALTH EFFECTS
IN AIR FORCE PERSONNEL FOLLOWING EXPOSURE TO HERBICIDE ORANGE

MATCHED COHORT DESIGN

I. Purpose of the Investigation

The purpose of this investigation is to determine whether long-term health effects exist and can be attributed to occupational exposure to Herbicide Orange through the use of epidemiologic techniques.

II. Synopsis of Background

A. Current

News media presentations have recently focused medical, political and lay attention on possible adverse health effects in military personnel, allegedly due to Herbicide Orange [a mixture of 2,4-dichlorophenoxyacetic acid (2,4-D) and 2,4,5-trichlorophenoxyacetic acid (2,4,5-T)] which was used as a defoliant during the Vietnam Conflict. Other herbicides containing 2,4,5-T were also used extensively, and as commonly used by the news media, the term "Herbicide Orange" refers to all of these 2,4,5-T products (a convention used throughout this protocol). This defoliant was later found to have been contaminated with the toxin 2,3,7,8-tetrachlorodibenzo-p-dioxin (TCDD) (Figure A-1, Section XII). Claims for compensation have been filed against the Veterans Administration (VA), by 650 veterans. In response to Congress, the General Accounting Office (GAO) investigated the issue and subsequently recommended that the Department of Defense (DOD) conduct a long-term epidemiologic study of the problem. The Department of the Air Force has made a formal commitment to the Congress and the White House to conduct such a study.

B. Use of Herbicides

Research and development on phenoxy herbicides began in the early 1940s. Most of the initial phytotoxic screening programs and the development of application technologies were sponsored by the DOD. The herbicide, 2,4,5-T, was first commercially produced in the United States in 1944. During the years from 1961 through 1969, the DOD procured approximately 34 percent of the total US production (53 million pounds) for use in the Republic of Vietnam (RVN). However, 8.9 million pounds of that amount were not sprayed in Vietnam, but were destroyed by at-sea incineration in 1977. The first sustained DOD operational use of herbicides was initiated during the Vietnam Conflict (Operation RANCH HAND) and the first shipment of herbicides used in RANCH HAND was received at Tan Son Nhut Air Base, (RVN), on 9 January 1962. The use of these compounds was intended to accomplish two objectives: (1) the defoliation of vegetation to improve visibility and thus decrease the risk of ambush, and (2) the destruction of enemy crops.

Four 2,4,5-T-containing herbicides were used by the military during the period 1962-1970. These four included:

(1) Herbicide Purple (used from 1962 through 1964)

n-butyl	2,4-D	50%
n-butyl	2,4,5-T	30%
iso-butyl	2,4,5-T	20%

(2) Herbicide Pink (used from 1962 through 1964)

n-butyl	2,4,5-T	60%
iso-butyl	2,4,5-T	40%

(3) Herbicide Green (used from 1962 through 1964)

n-butyl	2,4,5-T	100%

(4) Herbicide Orange (used from early 1965 through 15 April 1970)

n-butyl	2,4-D	50%
n-butyl	2,4,5-T	50%

Analyses of archived samples of Herbicide Purple suggest that the mean concentration of TCDD may have been approximately 33 ppm (Range: 17 to 47 ppm TCDD) while archived samples of Herbicide Orange may have had a mean concentration of approximately 2 ppm (Range: <0.02 to 15 ppm TCDD).

In addition, two other herbicides were widely used in RVN. These were Herbicide Blue (an organic arsenical formulated from the sodium salt of cacodylic acid), and Herbicide White (a water soluble triisopropanolamine salt formulation of 2,4-D and picloram). The amounts of the various herbicides used in RVN from January 1962 through February 1972 are shown in Table 1.

Table 1. Estimated Quantities of Herbicides and TCDD Sprayed in RVN, Jan 1962-Feb 1972

CHEMICAL	POUNDS
2,4-D	55,940,150
2,4,5-T	44,232,600
TCDD	368
Picloram	3,041,800
Cacodylic Acid	3,548,710
Herbicide Total	106,763,260

Ninety-six percent of the 2,4,5-T disseminated in RVN was contained in Herbicide Orange; the remaining 4 percent in Herbicides Green, Pink, and Purple. However, Herbicides Green, Pink and Purple contained approximately 40 percent of the estimated amount of TCDD disseminated in RVN. Green, Pink and Purple were sprayed as defoliants on less than 90,000 acres from 1962 through 1964, a period when only a small force of U.S. military personnel were in RVN. Ninety percent of all the Herbicide Orange (containing 38.3 million pounds of 2,4,5-T and 203 lb of TCDD) was used in defoliation operations on 2.9 million acres of inland forests and mangrove forests of RVN.

Most of the herbicide used in RVN was sprayed from aircraft. RANCH HAND aircraft, the C-123, disseminated 88 percent of all herbicide. Helicopters and ground application equipment used by personnel from all branches of the U.S. Armed Forces applied the remaining 12 percent (primarily Herbicide Blue).

Concurrent with the change to Herbicide Orange, the scope of aerial use shifted from four rotating aircrews to 30 permanently assigned aircrews and additional support personnel. Following the announcement in October 1969 that the administration of 2,4,5-T to pregnant rodents caused an increase in the rate of congenital abnormalities, the DOD confined Herbicide Orange spray operations to non-populated areas and in April 1970, all uses of the herbicide were halted. In March 1972, all remaining stocks of 2,4,5-T-containing herbicides were removed from RVN, and transported to Johnston Island, Pacific Ocean, for open storage (Project PACER IVY), and incinerated at sea in 1977 (Project PACER HO). In 1979, the Environmental Protection Agency (EPA) suspended the use of herbicides containing 2,4,5-T because an epidemiologic study in the United States attributed abortogenic effects to its use.

III. Goals of the Investigation

From the above background, three interdependent study goals emerge:

A. Health

(1) To identify veteran and active duty individuals with adverse health effects (physical and psychological) if any, which are attributable to herbicide exposure, and

(2) To identify other individuals at risk of developing future adverse health effects, if such exist.

B. Political

To satisfy the social concern for proper investigation voiced by lay and scientific communities, both national and international.

C. Legal

To clarify the question of compensation awards to the VA claimants.

With regard to the goal of legal clarification, it is apparent that data and conclusions arising from this investigation, positive, negative, or indeterminant, will probably be used to better assess the issue of long-term health effects and resultant compensation. The operational assumption of this study, therefore, is: Air Force Operation RANCH HAND personnel probably received a greater average occupational exposure to 2,4,5-T and TCDD than US Army ground personnel, implying that if there are any adverse long-term health effects, RANCH HAND personnel should develop greater numbers of acute and chronic clinical signs/symptoms from the exposure, and should manifest them sooner than US Army personnel. This dose-response notion suggests that although the Air Force population is not the best one to study, it is probably more appropriate than the Army population.

The overall scientific thrust of this investigation is to define the natural history of disease, if any, and its spectrum of illness, by direct and indirect methodology.

IV. Synopsis and Discussion of Literature

A. Overview

More than 20,000 scientific articles relating to the phenoxy herbicides have been published since the 1940's. Many of the articles cite herbicide-caused health effects in a variety of animal species. Most early studies used a myriad of herbicide formulations and unknowingly dealt with physically and chemically impure compounds, and the assay technology was far short of today's state-of-the-art. Many human studies have ascribed cause and effect relationships but have suffered from problems of clinical empiricism or questionable methodology. The only consistent chronic clinical finding associated with exposure to 2,4,5-T herbicide has been chloracne, recognized by most workers as the herald sign of overexposure to the herbicide. It is now recognized that the chloracne was caused by the presence of TCDD rather than the 2,4,5-T. Sequelae from chloracne, localized or systemic, appear to be unusual according to the preponderance of the literature. It is appropriate to note that sustained worldwide usage of herbicides for 30 years has not evoked a readily identifiable disease state. It is clear from the literature and the usage history of herbicides that if there are significant attributable long-term health effects, they are either reasonably rare, or of such nonspecific commonality that they blend unnoticeably into the symptoms, syndromes, or diseases associated with increasing age or other such factors.

B. Pharmacokinetics of 2,4-D, 2,4,5-T and TCDD

(1) 2,4-D

The pharmocokinetics of 2,4-D have been well studied in animals. 2,4-D is readily absorbed on oral administration, and is initially distributed in high concentrations to the central nervous system and liver. Eventually, all tissues are involved, with the kidneys accumulating twenty times the concentration of the other tissues. The plasma half-life of 2,4-D is approximately 3 to 12 hours, with elimination from the body through the kidneys at a dose-dependent rate. Generally, high doses or repeated lower doses result in tissue accumulation. The majority of 2,4-D is eliminated unmetabolized; however, esters of 2,4-D have been shown to undergo hydrolysis prior to excretion. Muscle and fat show the lowest accumulation of 2,4-D on repeated exposure, whereas the kidneys and liver show the highest accumulations. Within 24 hours of a single-dose administration of 2,4-D, 16.8% was present in the uterus, placenta, fetus and amniotic fluid in gravid rats. In addition, 2,4-D was found in the milk of lactating rats for up to six days following single-dose exposure.

(2) 2,4,5-T

The pharmacokinetics of 2,4,5-T have been well studied in animals. In all animals, 2,4,5-T has been shown to be readily absorbed upon oral administration. However, beyond this point, 2,4,5-T has shown marked variations in its pharmacokinetics in various animal species. These differences are thought to be due to variations in species, age, dosage levels, routes of administration and chemical formulations used in the various studies. The distribution is generally ubiquitous throughout the body except in hamsters, which show no placental passage, and in mice, which show placental passage only in late gestation. Clearance from plasma and the body varies greatly among animals with rats showing faster clearance than dogs, mice and man. In addition, this clearance appears to be generally dose-dependent. The biological half-life of 2,4,5-T in rats, as estimated by tissue analyses and urinary clearance at administered dosages of 5 mg/kg, is 4.7 hours. However, at 200 mg/kg, the half-life in rats is prolonged to 25 hours. Excretion of 2,4,5-T is primarily via the kidneys. The elimination of 2,4,5-T at low doses is essentially achieved in an unmetabolized form. However, at higher or more chronic doses, elimination involves the liver in a more active role (i.e., conjugation). Higher doses and repeated lower doses appear to result in accumulation in animal tissues.

(3) TCDD

The information on the absorption, distribution and excretion of TCDD has been mostly derived from animal models. The only reported human study dealing with pharmacokinetics of TCDD dealt with the analysis of TCDD in tissues at necropsy of one case of confirmed exposure subsequent to the accidental release of TCDD in Seveso, Italy in July 1976. Studies in rats, mice and guinea pigs generally show that intestinal absorption of TCDD is relatively complete, with a large proportion remaining unmetabolized in the liver. The majority of this TCDD is assumed to be localized in the liver microsomes (centrifugation techniques). Initially, adipose tissue accumulates TCDD, followed later by accumulation in the liver, adrenals, kidneys and lungs. The level of TCDD in the liver and adipose tissue is about ten-fold greater than in other body tissues; however, significant species variability has been observed. The biological half-life of TCDD varies by species, but is reported to range from 12 to 50 days. The major route of excretion is via the feces with urinary excretion occurring at a much reduced rate.

(4) Phenoxy Herbicides in Humans

Relatively few studies have dealt with the pharmacokinetics of 2,4-D and 2,4,5-T in humans. Numerous reports of occupational exposures in industry and in commercial and private herbicide applications have supported percutaneous entry as a major route of

exposure. Rapid absorption of 2,4-D and 2,4,5-T has been observed after oral administration. The primary mode of excretion of the phenoxy herbicides is via the urine with 74% of 2,4-D and 63%-72% of 2,4,5-T being cleared from the body within the first 96 hours. The majority of the herbicide is unmetabolized prior to excretion and the biological half-life of 2,4-D and 2,4,5-T in humans (as estimated by tissue analyses and urinary excretion) is 33 hours and 18 hours, respectively. Tissue analysis has revealed an ubiquitous distribution of the herbicides after absorption. Limited studies on the accumulation of the phenoxy herbicides following repeated doses suggest that such accumulation in humans is unlikely. This is in contrast to numerous animal studies on 2,4-D and 2,4,5-T which show that such accumulation does occur.

No specific data are available on the odor threshold of Herbicide Orange. Data are available however, on the odor threshold of a butyl ester formulation of 2,4,5-T. The odor threshold was found to be about 0.3 ppb (the taste threshold was 1.3 ppb). A Threshold Limit Value (TLV) of 10 mg/m^3 for both 2,4-D or 2,4,5-T has been adopted by the American Conference of Governmental Industrial Hygienists. The TLV is a time-weighted average concentration for a normal 8-hour workday/40-hour workweek to which workers may be repeatedly exposed, day after day, without adverse effect. Analysis of ambient air samples collected adjacent to and downwind from actual dedrumming operations involving Herbicide Orange were at least two orders of magnitude below the TLVs.

C. Proposed Cellular Mechanisms of Action for TCDD

TCDD has, in general, three proposed mechanisms of action by which its variety of effects, both documented and suspected, can be understood. All currently available information in this area is derived from animal, plant, and bacterial models. The few human studies dealing with mechanisms are limited to the clinical manifestation of chloracne.

(1) Microsomal Enzyme Induction

TCDD's ability to induce a variety of microsomal enzymes is well documented. The induction of aryl hydrocarbon hydroxylase, delta-aminolevulinic acid synthetase and cytochrome P-448/P-450 associated enzymes are implicated in the development of cutaneous porphyria. The induction of aryl hydrocarbon hydroxylase and other mixed-function oxygenases/oxidases has been associated with carcinogenesis and tumorogenesis. In addition, TCDD has been shown to be a possible promoter or cocarcinogen of known carcinogens. In some nonhuman studies, TCDD produced a protective effect against endocrine tumors (e.g., pituitary, uterine, pancreatic, adrenal and mammary tumors). TCDD's induction of UDP-glucuronyl transferase, an important enzyme in steroid metabolism, may explain this peculiar effect. The induction of DT-diaphorase and lysosomal acid proteinases have been implicated in TCDD's neuropathic effects.

These and other biochemical alterations may account for TCDD's clinical manifestation of chloracne resulting from an over production of keratin in the sebaceous ducts.

(2) DNA/TCDD Interaction.

Alterations in the structure and fidelity of transcription of DNA due to TCDD have been indirectly demonstrated. TCDD, because of its planar ring structure, is felt to "intercalate" with DNA causing "frame-shift" mutations in a manner similar to that seen with the acridine family of compounds. A few laboratory studies with bacterial systems (Escherichia coli and Salmonella typhimurium) and one plant system (the African Blood Lily) have implicated TCDD as being capable of producing chromosomal aberrations and perhaps a weak dominant lethal effect. This hypothesized DNA/TCDD interaction could explain the development of chloracne, as well as the suggested mutagenic and carcinogenic effects, if similar mechanisms occur in mammalian species.

(3) Toxicity.

A nonspecific or as yet unspecified toxicity continues to serve as a reasonable mechanism for TCDD's hepatic and thymus toxicity. TCDD has been described by some as "one of the most potent, low molecular weight toxins known", with extremely low concentrations producing severe liver damage and death in various animal studies. The immune suppression effect of TCDD has been shown to result specifically from its T-cell (thymus) toxicity. In addition, TCDD's concentration in adipose tissue suggests the possibility that under situations of weight loss (e.g., life style, medical indications, or disease), TCDD may be released into the circulation. Such hypothesized reemergence of the agent could result in low doses being either detectable and/or toxic at some later point in time. If TCDD's primary toxicity results from low doses (e.g., a mutagenic/carcinogenic effect) rather than high doses (e.g., cellular poisoning and cell death), then the deposition of TCDD in the adipose tissue may have greater significance with respect to delayed effects on the long-term health of the exposed individual. This possibility raises a theoretical dose-response paradox which might "explain" the prevailing preponderance of symptoms in populations which may have been exposed to relatively low doses of TCDD (see Section IV D).

D. Animal Studies

A comparison of animal toxicity studies is difficult due to variations in experimental designs which include differences in (1) the species, age, and sex of animals used; (2) the level, route, and length of exposure to chemicals; (3) the purity of the chemicals used; and (4) the criteria measured and the time sequence of data

collection. Animals have shown a wide range of toxic effects. This range may serve as a guide to anticipate the potential toxic effects in humans following exposure to Herbicide Orange.

A summarization of the literature is presented in Table A-1 of the Appendix, Section XIII. It is apparent that the toxic effects of 2,4-D and 2,4,5-T are markedly different than the effects of TCDD. TCDD is approximately 1000 times more toxic in acute studies. In addition, the slower clearance time of TCDD may account for the significantly lower daily doses required to elicit chronic toxicity. A consistent finding in TCDD toxicity is depletion of the lymphoid tissues throughout the host. This is readily characterized by involution of the thymus in all species studied. In relation to the chronic maternal toxic dose, the embryotoxic dose is markedly lower for TCDD than for 2,4-D and 2,4,5-T. Both 2,4,5-T and 2,4-D appear to be very weak teratogens and/or carcinogens at best, but these evaluations are complicated by varying levels of contamination by various dibenzo-p-dioxins. TCDD appears to have significant teratogenic and carcinogenic potentials which appear to be species specific.

The most striking observation noted in the literature is a marked variation in response among species. Examples of these variations are in the areas of acute toxicity (TCDD's LD_{50} in the guinea pig is 1 µg/kg compared to 1000 µg/kg in the dog), excretion (2,4,5-T plasma half-life in rats is 4.7 hrs compared to 77 hrs in dog), and oncogenicity (TCDD is oncogenic in rats but has not been shown to be oncogenic in mice under similar conditions). Even among strains of the same species (rats) variations in oncogenicity were noted following 2,4,5-T exposures. As noted earlier, this high variability between species is an important consideration in designing human studies.

A second area of interest noted in the literature is a possible dose-response paradox in nonhuman primates (rhesus monkey) following exposure to TCDD. Animals in chronic exposure studies fed a low level of TCDD in feed [e.g., 50-500 parts per trillion (ppt)] have shown signs of disease only after several months when total TCDD consumption was approximately 1 µg/kg body weight. Unfortunately, animals receiving comparable amounts of TCDD in single-dose acute toxicity studies (LD_{50} determinations) have not been observed for the emergence of chronic effects. Therefore, it remains unclear whether the toxicity demonstrated in chronic exposure studies is dependent upon repetitive exposure accumulated to 1 µg/kg or whether similar toxicity would also be demonstrated following a single dose of 1 µg/kg after a comparable observation period.

E. Case Reports

Much of the medical literature on 2,4-D, 2,4,5-T and TCDD exposures in humans is based on individual case reports. Most of the patients discussed in these reports were exposed to multiple chemical agents and, therefore, it is difficult to determine which

agents were responsible for specific symptoms. Nevertheless, the general areas of dermatologic and neuropsychiatric disease have been of primary interest to most investigations. Since the neuropsychiatric symptoms of herbicide exposure are numerous and largely subjective in nature, they have been extremely difficult to assess from a clinical standpoint. In addition, hepatic dysfunction, and renal, gastrointestinal and cardiac disturbances are "linked" to exposures to these chlorophenolic compounds.

(1) 2,4-D

A multitude of symptoms have been attributed to 2,4-D, and the ones reported most consistently are listed in the Appendix, Table A-2. Components of some of these selected symptoms/signs are described in Table A-3 of the Appendix. The asthenic syndrome, peripheral neuropathy and hepatic dysfunction are of particular interest. Other symptoms of systemic toxicity occur, but usually resolve within 4-6 weeks. The acute peripheral neuropathy associated with 2,4-D exposure has been extensively described. It has an early onset, causes prolonged disability of variable degree, and recovery has been incomplete in many cases. Electromyography in some patients has demonstrated denervation, and some studies have detected decreases in nerve conduction velocities. One autopsy study demonstrated a demyelination process within the brain of a 76-year-old male who committed suicide by ingesting 2,4-D in kerosene.

(2) 2,4,5-T/TCDD

The human effects of 2,4,5-T are difficult to evaluate since the chemical is contaminated with TCDD in the manufacturing process. The effects of TCDD have been determined from studies of trichlorophenol workers, and from laboratory workers using TCDD. Symptom/sign complexes attributable to exposure to 2,4,5-T and TCDD are listed in Tables A-2 and A-3 of the Appendix. Chloracne usually begins in the zygomatic/temporal region and is often found on and behind the pinna of the ear. This is an oily acne-like skin condition characterized by comedones and inclusion cysts which may result in extensive scarring. In severe cases spread of lesions to the throat, back and inguinal areas has been noted. This skin condition is frequently preceded by erythema and blepharoconjunctivitis. Active lesions usually disappear within two years, but have been found 30 years after exposure. Porphyria cutanea tarda and hypothyroidism have also been linked to 2,4,5-T/TCDD exposure. Other symptoms such as asthenia, liver and renal dysfunction, neuropathy, and gastrointestinal and cardiac disturbances are probably due to mechanisms similar or identical to those of 2,4-D.

Numerous instances of alleged disease due to 2,4-D/2,4,5-T exposure have been the subject of heavy media attention, particularly an episode of alleged 2,4,5-T exposure in Globe, Arizona,

in 1969. Despite extensive scientific review and analysis with negative findings, the Globe incident continues to be cited in current news media presentations. An incident in Missouri in 1971 in which six children, two adults and numerous animals were exposed to TCDD contaminated oil is frequently described as well. Many of the animals died and the humans developed chloracne and other acute toxic effects; however, all humans were healthy after five years of follow-up study. A final prospective assessment of fertility, teratogenesis and carcinogenesis will probably be made in the future.

F. Veteran Complaints

The Veterans Administration Compensation and Pension Service, Washington, DC, provided the USAF with data on the first 361 claims filed by veterans alleging altered health status due to exposure to Herbicide Orange. Numerous media presentations emphasizing both military and civilian herbicide exposures have described a remarkably wide spectrum of health effects being claimed by the veterans. Based on current guidelines established by the Veterans Administration (Program Guide 21-1, Section 0-18 and Title 38 USC), none of the symptoms cited in these claims could be shown to be secondary to exposure to Herbicide Orange. Based on a review of military personnel and medical records, the vast majority of the exposure claims remained unsubstantiated. The guidelines state that the only chronic residual of defoliant exposure ever incriminated by clinical history has been chloracne. Furthermore, chloracne has been associated with prolonged intensive exposure and all other toxic effects of the herbicide were viewed to be rapid in onset and to run a brief course followed by recovery without residual disease. In fact, the vast majority of the claims alleging exposure to Herbicide Orange were not for chloracne and, as a result, did not satisfy the criteria set forth for compensation. Three of the first 361 claims cited chloracne, but none could be confirmed by physical examination. Table 2 summarizes the descriptive characteristics of the first 361 claimants, while Table 3 summarizes the distribution of these complaints by symptom category. Appendix Table A-4 displays similar data from a VA review of 625 claims received by 30 June 1979.

Table 2

SUMMARY OF DESCRIPTIVE CHARACTERISTICS OF HERBICIDE RELATED CLAIMS SUBMITTED TO THE VETERANS ADMINISTRATION AS OF 30 APRIL 1979*

Total Number of Claims: 361
Sex: 100% Male
Mean Age: 34 years
Mean Number of Alleged Symptoms per Veteran: 2.3
Branch of Service: (Service history identified in 66.8% of claims)

US Army	66.4%
US Marine Corp	17.4%
US Air Force	11.2%
US Navy	5.0%

*Exact racial distribution unknown; anecdotal information suggests the majority of claimants are non-Caucasian.

Table 3

HERBICIDE RELATED CLAIMS SUBMITTED TO THE VETERANS ADMINISTRATION BY SYMPTOM CATEGORY AS OF 30 APRIL 1979

Total Number of Claims: 361-13 = 348*

		PERCENT
DERMATOLOGIC	(hairloss; chloracne; tinea, eczema, contact dermatitis, keloid, vitiligo, tumors, porphyria)	48.9
PSYCHIATRIC	(personality disorders, anxiety neurosis, depression, psychoses, pedophilia, alcoholism, adjustment reactions)	27.6
EAR, NOSE, & THROAT	(hearing loss, tinnitus, voice loss, sinusitis)	14.4
CANCER	(lung, bone, pancreas, brain, thyroid, larynx, colon, skin, soft palate, leukemia, lymphomas, Hodgkins Disease)	13.8
PERIPHERAL NEUROPATHY	(numbness, paresthesia, weakness, tingling, Guillan-Barre Syndrome, Multiple Sclerosis, Amyotrophic Lateral Sclerosis)	12.1
ASTHENIA	(headache, weight loss/gain, dizziness, fainting/blackouts, fatigue, lethargy)	11.2
GASTRO-INTESTINAL	(pain, ulcers, diarrhea, bleeding, hemorrhoids, colitis, achalasia, regional enteritis)	10.9
REPRODUCTIVE	(decreased sex drive, impotence, decreased fertility, miscarriages, sterility; genetic defects in offspring)	10.1
PULMONARY	(asthma, shortness of breath, infiltrates, chest pain, bronchitis, pulmonary hypertension, lung disease)	9.2

Table 3 (CONTINUED)

		PERCENT
OPHTHALMOLOGIC	(conjunctivitis, visual loss, pterygium, blurred vision, light sensitivity, optic atrophy)	8.9
MUSCULO-SKELETAL	(arthritis, gout, fractures, stiffness, spasm, hernia, bone disease, strains)	8.1
CARDIO-VASCULAR	(hypertension, arrhythmias, myocardial infarction, peripheral vascular disease, heart problems)	7.5
GENITO-URINARY	(urethritis, stones, renal disease, prostatitis, epididymitis, testicular mass)	4.0
CENTRAL NERVOUS SYSTEM	(strokes, loss of memory, seizures, tremors, meningoencephalitis,speech impairment)	3.7
HEPATIC	(hepatitis, liver disease, gall-bladder disease, jaundice)	3.5
PANCREATIC	(Diabetes Mellitus, pancreatitis, reactive hypoglycemia, increased amylase levels)	2.3
HEMATOLOGIC	(Pernicious Anemia, blood disorders, lymphnode disease, spleen disease, Polycythemia Vera)	1.4
COLLAGEN-VASCULAR	(Systemic Lupus Erythematosis, Rheumatoid Arthritis, Polymyositis, Sarcoidosis)	1.2
ALLERGIC	(allergic reactions)	0.9
FEVER	(low-grade fever, fever of unknown origin)	0.9
POISONING*	(lateritic soils)	0.3
PERIODONTITIS		0.3
AMYLOIDOSIS		0.3
HYPERTHYROIDISM		0.3

*NOTE: 13 CLAIMS ALLEGED EXPOSURE ONLY (WITHOUT SYMPTOMS) AS BASIS FOR COMPENSATION

Study design implications that can be drawn from these tables are limited due to the lack of knowledge concerning denominator data. Overall, the group of claimants exhibited a high frequency of readily identifiable disorders (e.g., dermatologic, psychiatric, and cancer). Further evaluation of the claims revealed that of the total number of claimants, 16.3%, had previous diagnoses of psychiatric disorders (20% of these diagnosed with schizophrenia).

Table 4 summarizes information from those claims submitted by USAF veterans.

Table 4

HERBICIDE RELATED CLAIMS SUBMITTED BY USAF VETERANS BY SYMPTOM CATEGORY AS OF 30 APRIL 1979

Number of USAF Veterans: 28 (Mean age = 35.4 years)

Symptom	Percent
Psychiatric	50
Dermatologic	39
Reproductive	25
Peripheral Neuropathy	14
Cancer	7
Miscellaneous	7

The demonstrated lack of an easily identifiable symptom complex on review of the veteran claims clearly substantiates the need for a comprehensive evaluation of individual patients.

G. Epidemiologic Studies

Epidemiologic studies of occupational groups have validated links between exposure to TCDD and the development of chloracne. Associations between TCDD and psychological abnormalities have also been suggested. A 1978 study by Hardell and Sandstrom in Sweden evaluated occupational exposure to chlorophenolic compounds in soft tissue cancer patients by a case-control design. They found an association between cancer and exposure, but methodologic problems have raised questions concerning the value of these findings.

Tung (1973) reported an abnormal increase in the occurrence of primary carcinoma of the liver in Vietnam (26 cases per year during 1955-1961 versus 144 cases per year during 1962-1968). He attributed the increase to a suspected carcinogenic effect of TCDD. His published study, however, has been criticized for failure to contain sufficient data and descriptions of methodology to verify his conclusions. The role of aflatoxin as an alternative cause of liver cancer was not addressed. His study was largely an empiric clinical report. A study sponsored by the EPA in 1979 in Alsea, Oregon, found a statistically significant increase in spontaneous abortion in areas where 2,4,5-T herbicide was routinely used in reforestation programs. The EPA concluded, however, that "for all its complexity, this analysis is a correlation analysis, and correlation does not necessarily mean causation." This report has also been the subject of intense scientific criticism. Differences in the availability of specialty obstetrical care and in the patterns of health care delivery existed between the exposed and control areas; these differences were not taken into consideration by the researchers. Variations in the ascertainment of spontaneous abortions in each of the area severely limited the validity of the

data, and of the conclusions derived from them. A recent study conducted in Australia (1978) was unable to find an association between neural tube birth defects and the use of 2,4,5-T herbicide.

Epidemiologic studies are continuing in Seveso, Italy where a population of 220,000 was potentially exposed to TCDD following an industrial accident in July 1976. These studies have involved investigations of more than 30,000 children and detailed clinical examinations of 1,024 persons, including the most severely exposed children and adults. Recent data (Homberger, et al., 1979) indicated that most cases of chloracne from this incident cleared rapidly. To date, the growth and development of newborn infants and children, immunological response, chromosomal aberrations, the response to the challenges of infectious diseases, and the morbidity and mortality patterns of the study population have not been significantly altered by TCDD exposure. Thirty-eight cases of birth defects were reported in early 1977, approximately 6-8 months after the industrial accident. However, the authors ascribe this increase to an artifact of surveillance. The social pressures operating in the Seveso population prior to the accident fostered underreporting of birth defects, while the atmosphere after the accident made the occurrence of a birth defect more socially acceptable. The post accident congenital malformation rate is not significantly different than the rate in similar areas of Central Europe. Similarly, ascertainment and surveillance of spontaneous abortions after July 1976 is hampered by the lack of valid baselines for the pre-accident period.

Another progress report on the aftermath of the Seveso accident (Pocchiari, et al. 1979) has revealed: (1) a decrease in the prevalence and severity of chloracne in the exposed population; (2) an increase in idiopathic clinical and subclinical neurologic disease as demonstrated by delayed peripheral nerve conduction velocities; and (3) increases in the prevalence of idiopathic hepatomegaly (8%) and alterations in liver function tests which returned to normal over an 18 month period of follow-up. Thus far, immunologic, cytogenetic, and embryomorphologic analyses have been unable to detect significant differences between exposed and non-exposed individuals.

A 2,4,5-T Dispute Resolution Conference was held in Arlington, Virginia, from 3 to 7 June 1979. Fifty-six recognized experts from the United States and seven foreign nations were actively involved in the deliberations of the conference. Human Exposure, Carcinogenicity/Mutagenicity, and Teratogenicity Working Groups independently reached the conclusions that there was no valid scientific evidence linking fetotoxicity, teratogenicity or carcinogenicity in humans to 2,4,5-T/TCDD exposures. The Human Exposure Working Group also concluded that there were no epidemiologic data associating TCDD with any long-term health effect in humans other than persistent chloracne. While they did not find evidence of serious long-term health effects, neither could they find strong

evidence for lack of effect. Most previous epidemiologic studies have not had sufficient statistical power to detect increased risks of low incidence/prevalence conditions in the observed populations, and the period of observation in many prospective studies has been less than ideal.

Several potentially valuable epidemiologic studies are currently in progress. Two independent and comprehensive studies of workers exposed to TCDD at a Monsanto manufacturing plant in Nitro, West Virginia, are currently being conducted (Mt. Sinai Medical Center, New York, and the Kettering Laboratory, University of Cincinnati, Ohio). These chemical industry workers were exposed over long periods of time and were previously evaluated in 1953 and 1956, following an industrial accident which occurred in 1949. In a personal communication, Dr. Raymond Suskind of the Kettering Laboratory has reported a follow-up study of 122 workers 28 years after heavy exposures to TCDD. There were 32 deaths in the group, and the relative risks of death were 0.69 for all causes, and 1.0 for malignancy; however, no firm conclusions can be drawn due to the small numbers involved. The Dow Chemical Company is currently analyzing data from a reproductive survey of the spouses of 2,4,5-T/TCDD exposed workers. A Czechoslovakian study involving a 10 year followup of TCDD exposed workers, and a US National Cancer Institute (NCI) mortality study of 4,400 structural pest control workers are also underway. Preliminary reports of a larger study of long-term morbidity by Suskind at the Nitro site have failed to reveal significant abnormalities other than persistent mild chloracne and decreased nerve conduction velocities, possibly associated with alcohol intake.

These new studies, and the continuing evaluations of the Seveso, Italy, population, should provide valuable data. The large study groups involved in the Seveso and NCI studies should provide good statistical power, and the Nitro, West Virginia, and Czechoslovakian efforts will evaluate the effects of exposure after prolonged periods of time (10-30 years). The results of these studies should fill major gaps in the knowledge of 2,4,5-T/TCDD epidemiology, and should prove to be useful in evaluating the long-term effects of these compounds on health and reproductive outcomes.

V. Epidemiologic Study Design: Matched Cohort

A. Design Considerations

The proposed goals for this study clearly mandate a comprehensive epidemiologic approach, incorporating mortality, morbidity and follow-up studies. Exposure to herbicides during the 1962-1970 time period may have initiated long-term health effects that may or may not be progressive. If such effects are detectable by past history, and can be verified, direct links to the compensation issue can be made. Current health status, as mirrored by the large number of recent VA claims, becomes of major interest, because such claims may indicate medical conditions that might be confirmed by a comprehensive physical examination. If both the mortality and morbidity studies yield only indeterminant or weakly suggestive findings, it may be that sufficient time has not yet passed for substantial emergence of longterm health effects. This dictates a requirement for a follow-up element to the study.

Methodological shortcomings are inherent in each element of this comprehensive study. To some extent, the classical deficiencies of each particular epidemiologic approach are compensated by the concurrent use of the other elements. For example, the low chance of identifying a relatively uncommon disease solely by the use of a follow-up study is offset by the inclusion of the mortality and morbidity studies. The relatively quick feedback that can be attained from these studies will serve to better define the follow-up study and will help to alleviate problems that arise as a result of changes in diagnostic criteria and methods over time. Nevertheless, problems that can affect ascertainment of disease in all phases of the study will remain. Inaccurate patient recall of antecedent events, the distortion of information by knowledge of anticipated symptomatology and participant or observer knowledge of their exposure status can only be corrected to a limited extent by review of records for symptom validation and "blind" assessment protocols. In addition, fundamental problems dealing with adequate selection of a control group and limiting loss to study can influence any comprehensive epidemiologic investigation. These and other pitfalls in study design will be discussed in more detail in Section VIII.

Since the study has three elements and confronts a health issue with undefined endpoints, strong bias, and political pressure with severe time constraints, the following design represents the best overall framework for achieving validity. The design process is complex and in itself time dependent. The epidemiologic techniques used are time-compressed. Unique record searching systems within the Air Force, and computer and clinical capabilities, as well as bias and loss-to-study correctors, will work toward making this effort achievable.

B. Ascertainment of Exposed and Control Group Populations

(1) Exposed Group

Operation RANCH HAND personnel flew C-123 aircraft in RVN during 1962-1970. Data from hand-compiled lists obtained through the RANCH HAND Association (a reunion organization), Air Force personnel computer entries, historical records and actual C-123 flight orders, place the estimated study population at approximately 1200 individuals. Of those personnel confirmed by the USAF computer system, 25% are still on active duty, with the remainder being composed of retired or separated persons. An indepth search to identify all RANCH HAND participants is being conducted of all organizational records stored at the Military Records Division, National Personnel Records Center (NPRC), St. Louis, Missouri. Detailed advertisements in active/retired military trade journals, VA publications, and local newspapers may be pursued in the near future to insure maximal ascertainment/identification of the exposed group. Introductory letters will be sent to the last known address of all identified persons, and nonresponse will be pursued by cross-locator systems available within the government (e.g., Social Security Administration, VA, Internal Revenue Service). Significant efforts will be made to account for at least 99% of the total population (see Figure A-2, Section XII).Because of the limited number of estimated RANCH HAND personnel (1200), no subsampling of the exposed group is planned in any phase of the study. All members will be strongly encouraged to participate in all phases of the investigation.

(a) Known or Predicted Characteristics of the Exposed Group

All exposed aircrew personnel are males currently ranging in age from approximately 28-58 years. The normal C-123 crew composition was one pilot and one copilot/navigator (both officers) and one spray equipment console operator in the rear of the aircraft (enlisted); thus, the aircrew officer-enlisted ratio will be approximately 2:1. The inclusion of RANCH HAND support personnel (predominantly enlisted) in the study will make the overall officer-enlisted ratio approximately 1:2. While almost all officers were Caucasian, approximately 10-14% of the enlisted men were Black. Attempts will be made to identify all maintenance personnel assigned to the RANCH HAND units. Maintenance of the RANCH HAND aircraft was performed within a step-wise organizational structure. Routine daily maintenance (primary) was conducted by flight line support personnel who were often dedicated exclusively to RANCH HAND operations. More extensive maintenance (secondary) was carried out by consolidated support units at the base level, which were also responsible for non-RANCH HAND C-123s as well. Major aircraft overhauls and modification were conducted by maintenance units at Clark Air Base, Philippines. The maintenance personnel in these centralized units were not directly assigned to RANCH HAND, and their exposures to RANCH HAND C-123 aircraft and herbicide

cannot be validated. From 1962 through 1964, the primary flight line maintenance teams were dedicated to RANCH HAND aircraft and these individuals can be identified by the mechanisms described above. In 1965, flight line maintenance was performed by personnel of the centralized maintenance organization (secondary) and it may not be feasible to adequately identify these individuals from available records. After 1966, the RANCH HAND organization transferred their base of operations to a new location, and primary maintenance was once again performed by personnel assigned specifically to RANCH HAND. These individuals can again be readily identified. Thus, maintenance personnel directly assigned to RANCH HAND will be included in the study. These complexities are summarized in Table 5.

Table 5

FEASIBILITY OF IDENTIFYING AIRCRAFT MAINTENANCE PERSONNEL (TOTAL POPULATION) EXPOSED TO HERBICIDE ORANGE

Time	Primary Maint Personnel[1]	Secondary Maint Personnel[2]
Jan 1962-Jul 1964	Yes	No
Aug 1964-Dec 1966	Yes/No[3]	No
Jan 1967-Apr 1970	Yes	No

[1]individual assigned to RH; denominator known
[2]individual not assigned specifically to RH, although may have serviced the aircraft; denominator not ascertainable
[3]"Morning Reports" may permit ascertainment of this group.

Because of the significant combat hazard associated with low, slow flying missions, all early RANCH HAND crewmembers were elite volunteers (see Risk-Taking Bias, Section VIII). In fact, RANCH HAND crewmembers comprised one of the most highly decorated units during the RVN Conflict. Anecdotal stories reveal that most crew members were, on occasion, heavily exposed to Herbicide Orange due to normal or combat induced equipment malfunctions within the aircraft. Many former RANCH HAND personnel are expected to be currently employed in the aerospace industry as commercial airline pilots, airline managers, and flight mechanics. RANCH HAND personnel still on active duty are expected to be found in senior management positions.

(2) Ancillary Study Groups (Non-RANCH HAND personnel)

Air Force handlers of herbicide drums in RVN were exposed to herbicides because of drum leakage. Advertisements

similar to those proposed for the RANCH HAND personnel may be issued in attempts to define this population. As the drum handlers were ad lib participants, no personnel designator was assigned to these individuals, thus prohibiting computer tracking and identification. The population is unknown, but expected to be small (less than 200) as the majority of drum handlers were known to be Vietnamese. Additional groups such as US Army personnel (officer and enlisted) who flew as observers, US Army helicopter crews, as well as experimental fighter-bomber spray personnel, may be included in the study. Specific epidemiologic/clinical studies for these groups must be planned by a separate protocol following their ascertainment since control group selection will be difficult or moot. It is intended that all data derived from the ancillary study groups will be subsetted for separate analysis and these data will be treated as anecdotal to the primary study.

The members of these groups are expected to be males, ranging in age from 28-68 years. The officer/enlisted ratio is estimated at 1:10. Approximately 10-18% of these populations are expected to be Black. Low numbers of respondents are expected and population at risk ascertainments will not be possible.

(3) Control Group (Not exposed to Herbicide Orange)

A review of all specialized flight units present during the RVN conflict, reveals clearly that there is no absolutely ideal control group for the RANCH HAND population. C-130 aircrew members and support personnel were selected because of sufficient population size, similar training profiles, and psychologic similarities to the RANCH HAND Group.

Total ascertainment of the C-130 population is being conducted by computer selection for specific military flying organizations, foreign country service and years of service. Over 2.3 million personnel records have already been scanned and the approximate C-130 sample size is 25,000 individuals. Aircrew members who flew C-130 aircraft in RVN during 1962-1970 will be selected as controls for the RANCH HAND aircrew population and the C-130 flight line maintenance population will be ascertained from personnel records by similar mechanisms, and will serve as the specific control population for the RANCH HAND support personnel. The proportions on active duty, and non-active duty status are expected to parallel the patterns in the exposed group.

Another possible control group, the non-RANCH HAND C-123 population, is known to be too small (approximately 3000) to provide adequate sampling flexibility and replacement under the proposed matched variable concept (see below and Section VI, I). Many of the RANCH HAND aircraft were reconfigured for transport and insecticide missions and thus, non-RANCH HAND crews responsible for these other missions, may have been exposed to significant Herbicide Orange residues in these aircraft.

Therefore, this group may not have been truly unexposed to herbicides, and was discarded as an appropriate control population. Crewmembers of C-7 transport aircraft were also considered as a potential control group; however, because of small size (1000-1200) and the fact that they served in RVN only during the post 1967 era, they were also dropped from consideration.

(a) Known or Predicted Characteristics of the Control Group

The normal crew composition of a C-130 is three officers and two enlisted personnel. The control group will be "pure" from the standpoint of lack of occupational exposure to herbicide. The entire control group will be considered "nonvolunteer" with respect to abnormally high combat risk. While in general they will possess lifestyle characteristics and socio-economic backgrounds similar to the exposed group, their overall combat morbidity/mortality and the resultant stress influences upon general health may be slightly less than in the exposed group. For those separated and retired C-130 controls, similar proportions to the exposed group are expected to be employed in the aerospace industry. Known and estimated factors of the control and exposed populations are summarized in Table 6.

(4) Matching Procedures and Rationale

Each member of the exposed group will be computer matched to a set of C-130 controls comprised of at least 10 individuals using four variables. Since the two groups are highly selected and inherently similar with respect to many variables, very close matches are feasible. This epidemiologic design incorporates a matched concept because: (1) a matched cohort design will provide maximum test power throughout the entire study, (2) statistical intergroup comparisons may be made without normalization by four key variables known to effect symptom frequencies of interest, thus providing greater power for complex statistical testing, and (3) close matching is feasible and necessary for some of the anticipated analyses of the physical examination findings. Matches will not necessarily be rigidly maintained throughout the analytic phase, depending upon the particular analysis. It is apparent that following the match, both exposed and control populations will be very nearly identical with respect to the four influencing variables so that a replacement concept is feasible (see E below). In the event that frequent match breaks still occur, stratification techniques can be used.

Matching will be conducted for (1) age, by year of birth, and closest month possible, (2) Air Force Speciality Code (AFSC) as an absolute match, (3) length of time spent in Vietnam, to the closest six month period, and (4) race (Caucasian versus non-Caucasian) as an absolute match. These variables are listed in priority order of the match sequence. Specific rationale for these

Table 6

COMPARISON OF THE STUDY GROUP TO POSSIBLE CONTROL GROUPS BY KNOWN AND ESTIMATED FACTORS

KNOWN FACTORS	STUDY GROUP	POSSIBLE CONTROL GROUPS		
	RANCH HAND C-123	Non-RANCH HAND C-123	C-7	C-130
POPULATION SIZE	1200	3000	1200	25,000
OFFICER/ENLISTED RATIO	1:2	1:2	1:2	1:2
AIRCRAFT FUEL (AV-GAS)	YES (+JP-4)*	YES (+JP-4)*	YES	NO (JP-4 only)
OCCUPATIONAL HERBICIDE EXPOSURE	YES	YES/NO**	NO	NO
ESTIMATED FACTORS				
OCCUPATIONAL INSECTICIDE EXPOSURE	2+	1+ to 4+	0	0
COMBAT HAZARD	4+	3+	3+	2+
RVN-IN COUNTRY ASSIGNMENT	4+	4+	4+	2+

*In 1968, aircraft were modified with a JP-4 booster.
**Contaminated aircraft reconfigured for transport may have resulted in exposure to non-RANCH HAND personnel.

variables is as follows: (1) many clinical symptoms and signs allegedly attributed to herbicide exposure (see literature review) can also be attributed to an aging effect, or to collateral diseases more commonly associated with advancing age, (2) AFSC controls specifically for officer-enlisted status (as well as crewmember or noncrewmember status), a variable strongly linked to educational background, current socio-economic status, and moderately linked to age (5 year median difference) and socio-economic background, (3) total length of tour in RVN (measured in six month intervals, or actual flying hours, if feasible) will control for the generalized probability of combat morbidity, mortality, and for combat induced neuro-psychiatric disorders; additionally, length of tour may reflect effects related to intensity of alcohol consumption, drug consumption (chemoprophylactic or illicit), and degree of tropical disease acquisition and (4) race controls for differences in chronic disease development, socio-economic background, etc. (note: there is possible racial discordance for VA claimants).

(5) Study Group Selection Procedures

(a) Mortality Analysis

For the mortality analysis, ten controls will be selected for each exposed subject, regardless of current vital status (Figure 1). The control individuals will be randomly assigned to one of 10 cohorts, C_1 through C_{10}. All of the exposed individuals and a 50% random sample of each set of controls will be included in the mortality analysis. The current vital status of each exposed-control set will be determined, and their mortality experience will be followed throughout the duration of this study. In addition to providing a 1:5 mortality analysis, this technique will characterize the mortality experience of each control cohort.

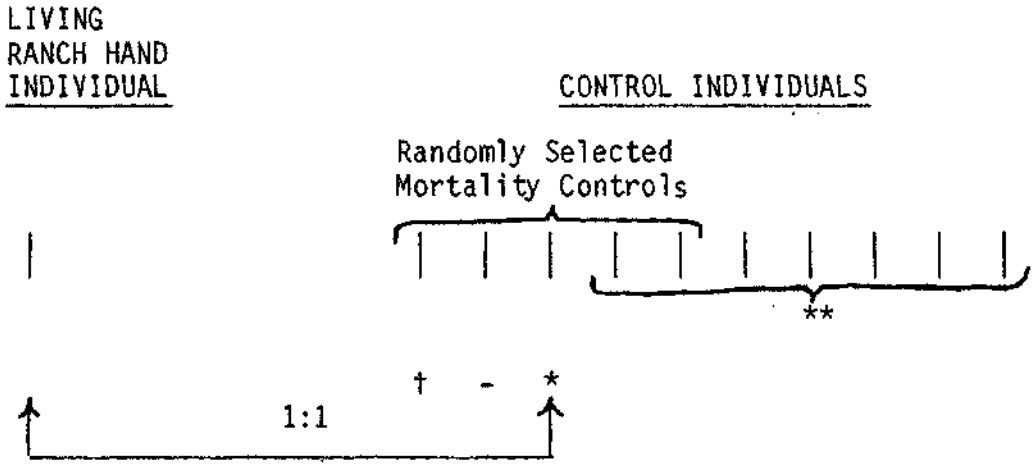
Figure 1. Mortality Analysis Cohorts

RH COHORT	CONTROL COHORTS					
E	C_1					C_{10}
E_1	$C_{1,1}$	$C_{1,2}$	$C_{1,3}$	$C_{1,4}$	.....	$C_{1,10}$
E_2	$C_{2,1}$	$C_{2,2}$	$C_{2,3}$	$C_{2,4}$	.....	$C_{2,10}$
E_3	$C_{3,1}$	$C_{3,2}$	$C_{3,3}$	$C_{3,4}$	.....	$C_{3,10}$
E_4	$C_{4,1}$	$C_{4,2}$	$C_{4,3}$	$C_{4,4}$	.....	$C_{4,10}$
.	.	.	.	.		.
.	.	.	.	.		.
.	.	.	.	.		.
E_j	$C_{j,1}$	$C_{j,2}$	$C_{j,3}$	$C_{j,4}$	.....	$C_{j,10}$

(b) Questionnaire, Physical Examination and Follow-up Study

Each living exposed subject and a randomly selected individual from his set of five mortality controls will be included in the questionnaire and physical examination phases of the study. If this primary control is deceased, unaccountable, or unwilling to participate in the morbidity and follow-up studies, another control will be selected and so on until a willing subject is found (Figure 2). Since the control's vital status and volunteerism should be independent of the matching sequence, many primary controls should enter the study. The remaining members of the control set will be used as replacement candidates for possible use later in the study (see section E below). All replacement controls will be clearly identified for the purposes of subset analysis so that population differences, if any, between the first randomly assigned selectees (noncompliant) and the replacements (compliant) can be assessed.

Figure 2. Selection Procedure for the Questionnaire, Physical Examination, and Follow-up Study



† Deceased
- Unwilling
* Volunteer
** Replacement Candidates

C. Mortality Study

(1) Introduction

The mortality, morbidity, and follow-up studies are components of a "non-concurrent" prospective study used in the observation of a specially exposed group or industrial population starting from some date in the past. The initial exposures occurred 8-17 years ago and varied in intensity and duration from one RANCH HAND member to another. Access to employment, medical and other

types of records is an obvious requisite for such a study. The classical "case-control", retrospective study is not operative in this protocol due to the lack of defined clinical endpoints. The mortality study will be conducted in two phases; a review of past mortality, and a continuing assessment of the death experience in the exposed and control cohorts over the duration of the RANCH HAND II project.

(2) Data Collection Methods

The mortality status of the exposed cohort and the randomly selected controls will be ascertained using multiple techniques including; telephone interviews with subjects or their families, a review of Death Benefits claims filed with the VA, acquisition of death certificates on all known deceased individuals and reviews of autopsy reports and medical records whenever possible.

D. Morbidity Study

(1) General Considerations

A vigorous attempt to determine the morbidity experience of all exposed subjects and their primary controls will be undertaken using questionnaires, indepth personal interviews, and physical examinations. The schedule and method of contact with the study subjects is depicted in the Appendix, Table A-5.

(2) Questionnaire Methods

All exposed members and their matched primary controls will be offered a comprehensive personal and family health questionnaire via telephone. The telephone questionnaire is an important part of this study because non-compliance rates for the physical examination and its face-to-face interview are expected to be substantially greater than non-compliance with the telephone questionnaire. As depicted in the Appendix, Figure A-2, only an estimated 40% of the RANCH HAND population will participate in the examination, while 65% will respond to the telephone questionnaire. The information collected by questionnaire from these additional 309 individuals and their controls will provide valuable morbidity data which would otherwise be lost. The questionnaire (see Section XIII) will emphasize identification data, RVN tour history, dermatologic conditions, neuropsychiatric conditions, fertility aberrations, genetic defects in offspring, sensory defects, and personality factors, including assessments of risk-taking behavior. A review of medical systems will be included in the questionnaire, and will inventory symptoms prior to, during, and after duty in RVN as well as those currently manifested. The questionnaire will be limited to a 30-45 minute telephonic interaction with participants, and it will take 10 to 12 months to complete all initial questionnaires on both groups. It may be necessary to conduct the questionnaire in two telephone sessions to minimize fatigue and maximize response. The

questionnaire will be "field-tested" on a group of 25 to 30 former Air Force pilots with RVN combat experience. Specific questions on the questionnaire will be directed to verifiable information, wherever possible. Specific response verification and bias indicator questions (nonsense symptoms), and indicators of risk-taking behavior are being developed. They will be added and appropriately sequenced immediately prior to the start of the study. Questionnaire data will be cross-linked and integrated with medical record information and physical examination findings. Questionnaire data from individuals not completing all phases of the study will not be discarded, but will be incorporated within the entire data base where statistically appropriate. Each participant will be asked to sign release forms so that all civilian health records, including those of dependents, can be obtained and reviewed as necessary. Federal health records on all family members on file in the NPRC will be retrieved. For retired members, and separated members with VA priviledges, all available VA medical records will be obtained. All retrieved medical records will be reviewed, scored, compared to questionnaire data for reliabililty, and then be entered into a repository system. Identified participants who are non-responsive to questionnaire will be pursued to determine status, disinterest, moribund state or death, etc. These individuals will be cross-referenced in other federal record systems in an attempt to achieve total ascertainment. Death certificates and autopsy reports will be retrieved on all dead exposed and matched control subjects for the mortality analysis. Birth/death certificates will be sought for all offspring, born subsequent to the study subject's RVN duty.

(3) Physical Examination

A voluntary comprehensive physical examination will be offered to all individuals in both the exposed and primary control groups. The condition for entry into the examination phase of the study will be the completion of the baseline questionnaire. In the event that the primary control does not complete both the questionnaire and the physical examination, a replacement will be selected from the control set (See Figure 2). Consequently, "person-months" of observation will be used in the statistical analysis. Statistical testing will be conducted by a variety of techniques on both questionnaire and examination findings (see VI, Statistical Methodology below). At the time of physical examination, an extensive face-to-face interview will be conducted. A standardized protocol will be used to insure comparability of interview data. This will provide cross-reference data to the initial questionnaire and to medical record data, if retrievable. Specific response verification and bias indicator questions will be included in the interview as well.

(4) Examination Parameters

A comprehensive physical examination will be conducted on all willing participants. The examination will be structured as outlined below and in Section XIV.

Physical Examination Profile

General Physical Examination	Hemoglobin	CPK
FBS, 2 Hr Post Prandial	Hematocrit	ECG
Urinalysis	White Blood Cell Count	Chest X-Ray
BUN/Creatinine	Platelet Count	VDRL/FTA
Cholesterol/HDL Cholesterol	RBC Indices	
Triglycerides	Sedimentation Rate	
Serum Protein Electrophoresis	Cortisol Differential	
	Thyroid Profile (RIA)	

Dermatologic Examination

- Urine Porphyrins
- Urine Porphobilinogen
- Delta-aminolevulenic Acid

Neuro-Psychiatric Examination

- General Neurologic Examination
- Psychological Battery
  - MMPI
  - WAIS
  - WRAT
  - Halstead-Reitan
  - Wechsler Memory Scale Subtests
  - Cornell Index
- Nerve Conduction Velocities

Reproductive Examination

- LH, FSH, Testosterone
- Semen Analysis

Neoplastic/Hepatic Examination

- SGOT
- SGPT
- GGTP
- Alkaline Phosphatase
- LDH (Isoenzymes if elevated)

Additional Studies (Individuals with abnormal history or examination)

- Karyotyping
- Hepatitis Antigens/ Antibodies (A and B)
- Additional Consultations as Required
- Anti-Nuclear Antibody
- Immunoelectrophoresis
- Monilia Skin Test
- Quantitative Immunoglobulins
- Bilateral profile and full-face photographs

Examinations will be performed at a single USAF medical facility having dermatologic, neurologic and electromyogram/ nerve conduction capabilities. Special Air Force authorization will be obtained to conduct such examinations on individuals separated from the service and informed consent forms will be obtained for nerve conduction tests. Physicians and technicians will handle

all participants without a knowledge of exposed or control status, and will conduct the examinations by standardized protocols to minimize variability. Medical students and interns will not perform these examinations, and specialty trained neurologists and dermatologists will perform the appropriate portions of the examination. An onsite monitor will insure that the examination protocol is followed. Clinical specimens will be forwarded to USAFSAM where most of the laboratory procedures will be conducted. The only laboratory procedures to be accomplished at the examining facility will be those which require immediate processing (see Section XIV, D(5)). All laboratory tests will thus be subject to the same technology and rigid quality control. Laboratory and physical examination data will be measured on a continuous scale whenever possible in order to improve statistical power in the analysis.

Special contingencies will be made for unusal laboratory testing. Karyotyping of the individual and his family members will be performed if clinical history or physical examination findings are suggestive of this need. Most well conducted studies have shown that, when present, chromosomal abnormalities due to TCDD are transient. If on detailed analysis of the baseline examination and questionnaire, reproductive areas are heavily affected, routine karyotyping may be included in the test battery for the follow-up study. TCDD analysis on blood and urine will be considered in the future provided that (1) strong cause and effect relationships can be ascribed to Herbicide Orange and (2) high resolution mass spectrometry technology achieves 10 femtogram sensitivity with high specificity. Appropriate specimens will be obtained from all participants, aliquoted, and preserved at -70°C for possible analysis in the future.

Physical examination and laboratory data will be placed in the member's coded master file for detailed cross-analysis to questionnaire data. Information identifiable to the subject will not be released without his consent in accordance with the Privacy Act. (Exceptions: In accordance with Air Force regulations, all active duty flying personnel and air traffic controllers found to have disqualifying defects will be temporarily "grounded" pending resolution; in accordance with federal regulations, all commercial airline pilots and air traffic controllers found to have disqualifying defects will be reported to the Federal Aviation Administration.)

E. Follow-up Study

(1) Study Adaptations

Following complete data analysis of the mortality and morbidity studies, an adaptive or restrictive health survey will be developed and administered to all follow-up study subjects three and five years after the initial questionnaire. Similarly, a condensed physical examination profile that will achieve adequate sensitivity and specificity for prospective diagnosis will be

developed. The adaptive physical examination will be offered to all follow-up participants, and will also be conducted in years three and five (see Appendix, Table A-5). An interim examination is essential in this study because the age group under study is approaching that portion of the mortality/illness incidence curve with the steepest slope. A lapse of five years between examinations would easily miss significant development of disease in the intervening years. Ample precedent for interim examinations can be found in the Framingham cardiovascular disease study and the follow-up evaluation of West Point graduates being conducted by the Air Force.

(2) Entry Criteria

All exposed or control individuals completing the baseline questionnaire and physical examination will be entered into the follow-up; further continuation will depend upon the member's willingness/ability to participate in additional health surveys and condensed examinations.

(3) Loss to Study

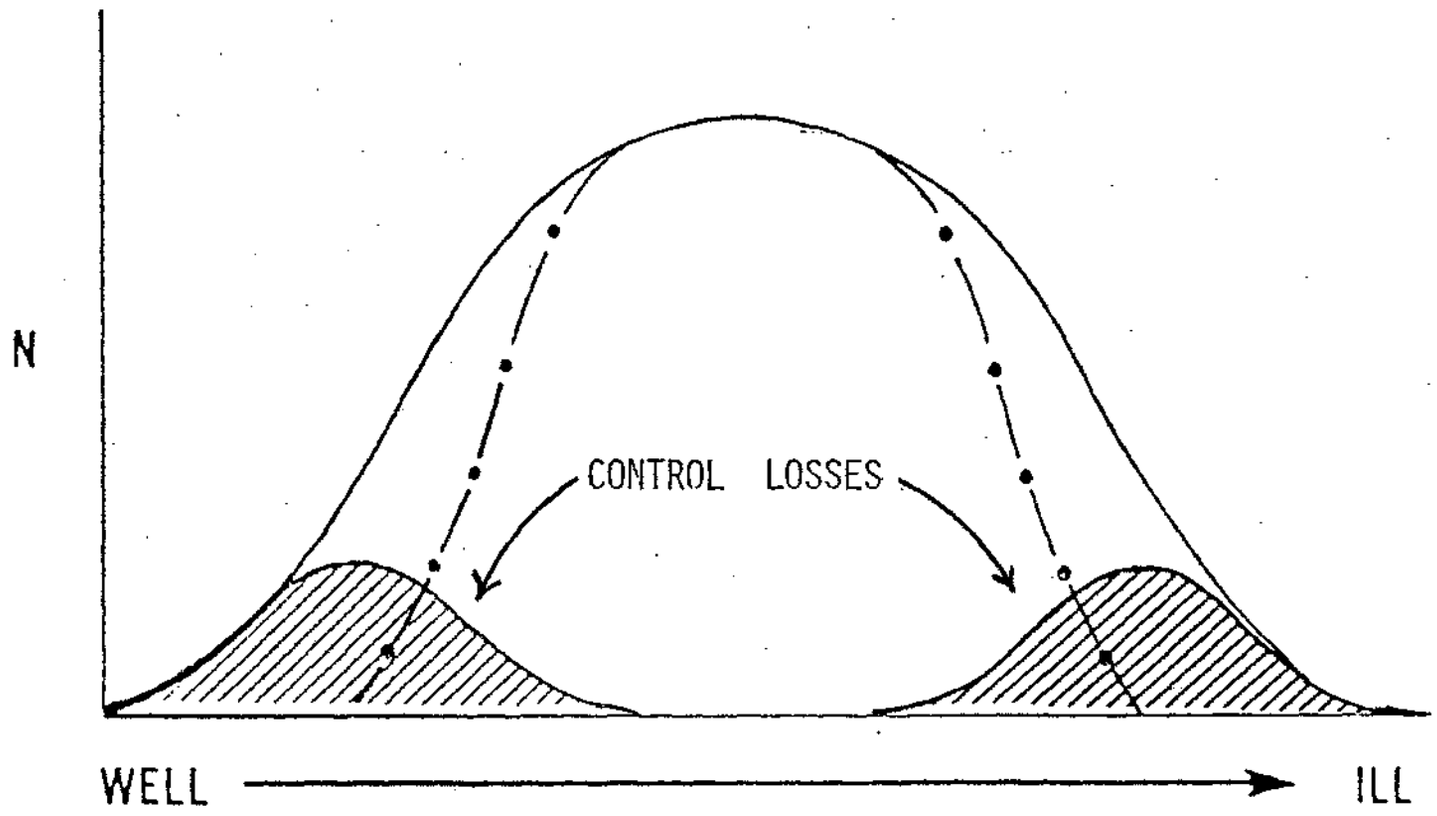
Loss of participants over time adversely affects any epidemologic study in two ways. As the sizes of the study groups decrease, statistical power also declines, and bias is injected into the study if losses are not randomly distributed in the study populations. It is reasonable to assume that in this study, losses will be non-random with greater non-compliance among individuals who perceive their health as "well," since there is less incentive for this group to continue participation. As shown in Figure 3, such a differential pattern of loss will alter the population and skew the frequency distribution curve.

Most previous epidemiologic studies have approached the problem of declining statistical power by beginning the study with multiple controls per exposed subject, and passively allowing attrition to occur throughout the study period. However, this approach does not address the problem of bias. This study will take an active approach to both of these problems by using a replacement concept. As a control is lost to study, a replacement will be chosen from the original set of 10 matched controls. The replacement will be selected from the control set, and will have a perception of health similar to that of the lost control (Figure 4). The replacement strategy will maintain statistical power and the integrity of the matched design despite loss to study in the control group, and will correct anticipated bias while minimizing the number of required physical examinations.

At the initiation of the follow-up study, loss of an exposed member will not be cause to cease surveillance of his primary matched control. In the event of a control loss (for reasons other than death), another control from the set will be brought to study (Figure 5), the comprehensive questionnaire will be administered, and a baseline physical examination performed. Medical data

FIGURE 3

EFFECT OF NON-RANDOM LOSS TO STUDY IN THE CONTROL POPULATION



- IF CONTROL LOSSES ARE ILL, A SPURIOUS EFFECT IS ATTRIBUTED TO HERBICIDE EXPOSURE.
- IF CONTROL LOSSES ARE WELL, A TRUE/VALID HEALTH EFFECT IS DILUTED.

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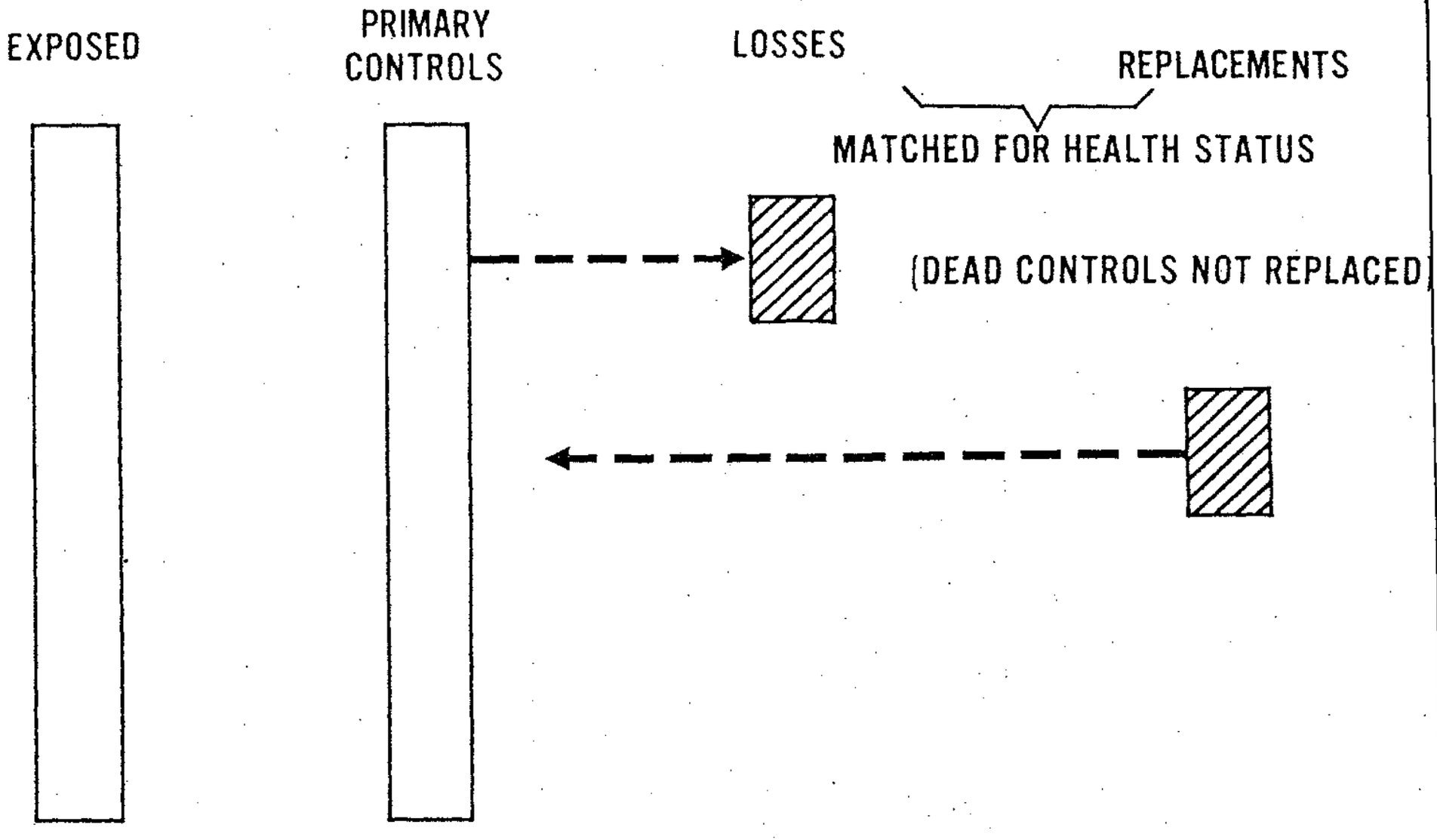
FIGURE 4

ANALYSIS OF THE REPLACEMENT STRATEGY



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for the intervening years will be reconstructed from the questionnaire and interview responses. IN ALL CASES OF LOSS-TO-LOSS STUDY, INTENSIVE EFFORTS WILL BE MADE TO DETERMINE THE SPECIFIC REASONS FOR NON-COMPLIANCE, AND DATA FROM REPLACEMENT CONTROLS WILL BE REVIEWED TO ASSESS COMPARABILITY WITH THE LOST INDIVIDUALS. Medical record reviews of new entrants will continue throughout the follow-up period.

(4) Study Length

The follow-up study is initially planned for five consecutive years. Results of the entire effort will be presented to a neutral scientific body. Their recommendation for continuance/discontinuance of the study will be forwarded to the Air Force Surgeon General for final decision.

F. Determination of "Disease"

(1) Introduction

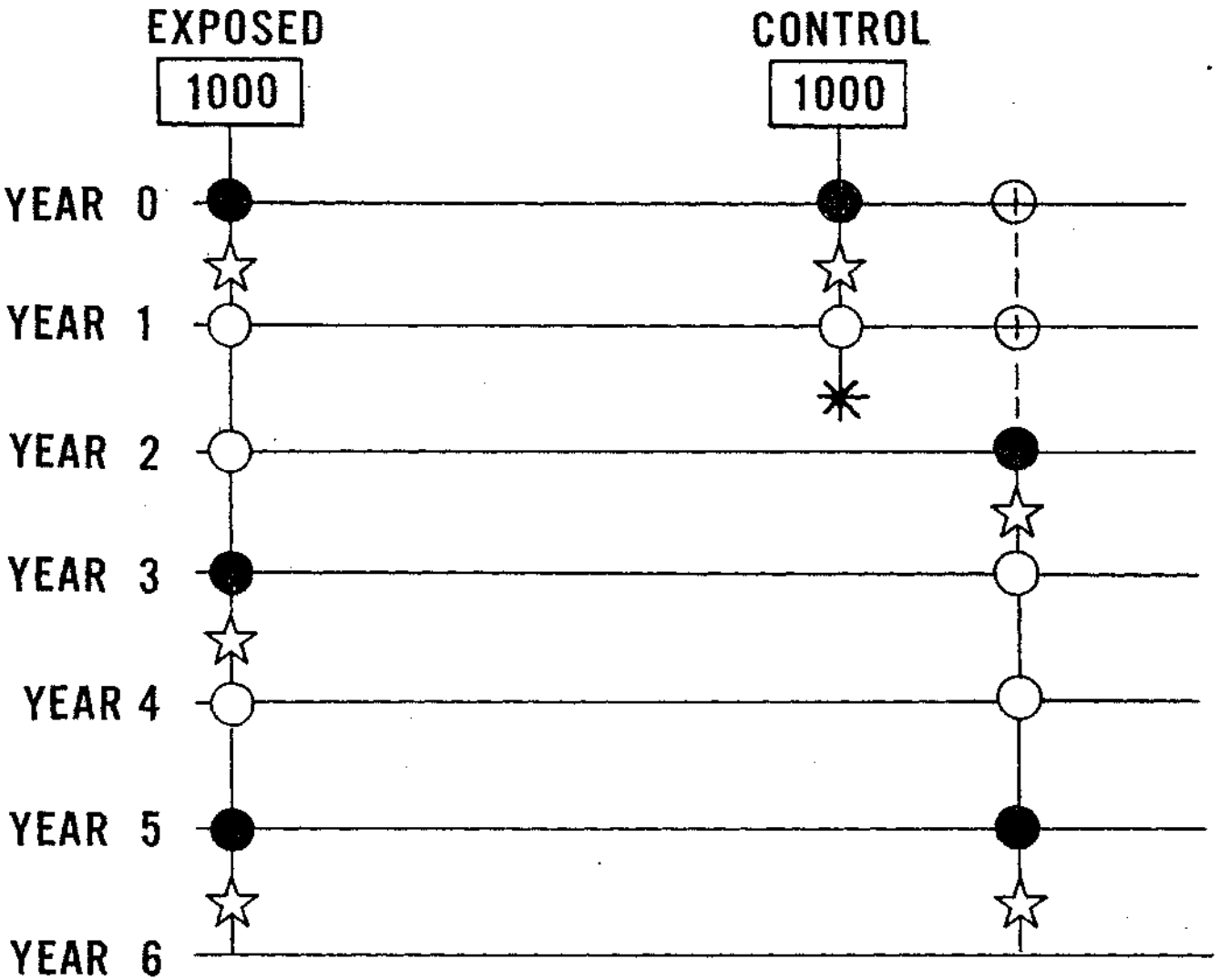
Since this study is dealing with an unknown clinical endpoint with unknown latency, determination of a disease state by statistical methodology is a prime scientific thrust of the investigation. From the literature, chloracne is the only recognized chronic disease associated with high exposure to dioxin. The questions of primary interest are: (1) Does a history of chloracne invariably lead to future disease? and (2) In the absence of chloracne, is there emergence of other attributable diseases? Under a broad concept of "spectrum of illness", either or both of these conditions are possible. The clarification of their respective contributions to the natural history of past or of subsequent "disease" is of significant interest.

(2) Discussion

Inferences about a disease state from this study can be derived from several logical approaches. These approaches can be grouped into two categories: (1) those dealing with symptoms which can be used to construct a symptom complex that may represent disease, and (2) those dealing with physical signs which in themselves represent disease. In the former, one can form a subset of individuals that have symptoms (e.g., infertility) and study them during the morbidity and follow-up studies. Focusing on the overall patterns of alleged symptoms and categorizing them into a symptom complex may identify those individuals with a disease syndrome, or those at higher risk of developing disease (e.g., genetic disorders, cancer). In the latter approach, data on abnormal physical signs (e.g., genetic defects in offspring) and laboratory results can be compared between exposed and non-exposed groups in an attempt to again establish the presence or absence of disease. By putting this array of data into a logical decision-making scheme, specific relative risks can be calculated in the follow-up study.

FIGURE 5

CONTROL REPLACEMENT FOR THE MORBIDITY AND FOLLOW UP STUDIES



● QUESTIONNAIRE DATA

○ RECONSTRUCTED DATA

✳ LOSS TO STUDY

☆ PHYSICAL EXAMINATION DATA

By the use of combinational and correlational analyses, statements about the probability of a disease state, a subclinical state, and over-reporting bias can be attempted. If the development of symptoms in the exposed group is positively correlated with physical findings, and this correlation is absent in the control group, a statement concerning the existence of a possible disease state can be made. By taking these possible combinations of observations and viewing them in the context of associated positive verifiers, negative bias indicators, and positive exposure index, the probability of over-reporting bias acting in these circumstances can be substantially reduced and, as a result, any statement concerning the existence of disease is strengthened. Similarly, if symptoms in the exposed group do not correlate with the development of findings, but are associated with positive laboratory results, a statement concerning the existence of a subclinical disease state can be made. However, if comparisons within the RANCH HAND group reveal a negative correlation between reported symptoms and the presence of abnormal physical signs, then an over-reporting bias and/or subclinical disease state is suggested.

Another method to assist in the determination of a disease state is the use of normalized exposure index and the application of regression techniques to the resulting curve. If there is a positive correlation between increased exposure and the presence of various abnormal physical signs and/or verifiable symptoms, then a symptom complex or disease syndrome is suggested. Factors suspected of altering the classical dose-response curve include cellular repair mechanisms and the hypothesized release of TCDD from adipose tissue following weight loss. The addition of multivariate techniques to the regression analyses will strengthen statements about the presence of disease. Beyond these pair-wise and group comparisons, newer techniques of pattern recognition, such as Factor Analysis and Cluster Theory, are being considered in order to achieve a more automatic and objective analysis.

The strength of any inferences made from these analyses is dependent upon the statistical power inherent in the study. In addition, due to the possibility of latency being a factor in this study, a negative analysis at any time within the study does not categorically imply lack of disease, since sufficient time for emergence may not have passed.

G. Determinations of Exposure Indices:

(1) Exposure Concepts

A major concern in conducting this study is the lack of accurate exposure data. Although most personnel assigned to RANCH HAND squadrons were undoubtedly exposed to Herbicide Orange and TCDD, the exposures within the group must have varied widely. Exposure to herbicides and TCDD by RANCH HAND personnel occurred almost daily. Anecdotal information suggests that many had direct skin contact which was repetitive over a long period of time

(one-year tour for most individuals). Further, it is also suggested that most RANCH HAND personnel felt that the herbicides employed in the operations were not toxic to animals and man and hence, they did not exercise the caution in handling these chemicals that is recommended today. Several individuals have, in fact, repetitively tasted or drunk Herbicide Orange to convince their colleagues or the press of its safety.

From a historical review of RANCH HAND operations, most individuals can be classified into one of three groups based on their likely potential for exposure to the herbicides:

(1)	Pilots, Co-pilots and Navigators:	low potential
(2)	Crew Chiefs, Aircraft Mechanic, and other Support Personnel:	moderate potential
(3)	Console Operators and Flight Engineers:	high potential

The "pilot" group probably received most of their exposure during pre-flight checks as well as during the actual dissemination missions. The crew chief group experienced contact with herbicides during dedrumming and aircraft loading operations as well as during on-site repair of the aircraft and spray equipment. The console operator group was exposed while supervising the loading of the aircraft, during ground testing the equipment, and by tank leakage during dissemination missions.

The available historical records on Operation RANCH HAND indicate that personnel assigned to the project seldom had a "routine" work schedule or environment, thus complicating estimates of the level of herbicide and dioxin exposure. Since actual exposure data (e.g., mg of herbicide/kg body wt) are not available, the establishment of an exposure index will be attempted. The exposure index will be calculated for each RANCH HAND individual to obtain a standardized frequency distribution. The exposure index will be calculated by evaluating the known factors that would have influenced exposure. For aircrew members, these will include such factors as:

(1) Date of tour with RANCH HAND in Vietnam.
(2) Number and lengths of tours in Vietnam with RANCH HAND.
(3) Number of herbicide dissemination missions (as reflected by flying hours and air medals).
(4) Herbicides employed (yearly records are available that reflect the amount of each herbicide sprayed).
(5) Crew position.
(6) Routes of exposure (the major route of exposure for most RANCH HAND personnel was probably percutaneous, although for the console operator, exposure through inhalation may have also been significant).

An exposure index may be derived from consideration of the above factors, i.e., it is a function of those factors:

$$E_I = f(D,T,P,C,R,H),$$

where E_I = Exposure Index

D = Dioxin Concentration Estimate During RANCH HAND Tour

The date of tour with RANCH HAND is important since mean dioxin concentrations in the herbicides changed by a factor of 16 between pre-1965 and post-1965. In 1965, both high and low dioxin formulations were available. Thus, a scale of values for TCDD are:

Pre-1965	.32
1965	.16
Post-1965	2

T = Duration of Exposure/Number of Missions Flown

P = Proportion of 2,4,5-T Herbicides Used

The ratio value of the amount of 2,4,5-T containing Herbicide applied for any given year against total amount of the herbicide applied during the same year. Data on herbicide expenditures by year are available. For example in 1962, Purple comprised 88 percent of all herbicides used that year, while in 1966, Herbicide Orange accounted for 66 percent of the total amount of herbicide applied.

C = Crew Position/AFSC

Relative exposure values for this factor will be determined by separate studies.

R = Route of Exposure

Relative values for exposures via ingestion, inhalation or percutaneous absorption will need to be evaluated from literature, computer modeling, and inflight determinations using a Herbicide Orange simulant.

H = Exposure History

A relative value based upon the subject's preception as to the magnitude of his own exposure. This will be obtained from the questionnaire and may reflect no exposure, a light exposure, a moderate exposure or a heavy exposure. A weighted value will be assigned to each exposure estimate, if possible.

(2) Proposed Studies

Within the exposure index equation, relative exposure values for two factors are lacking; crew position within the C-123 aircraft and the route of exposure. Approaches which will be used in obtaining estimates for these two factors include (1) use of a diffusion model which would consider the internal aircraft volume and configuration in computing specified source strengths and concentration levels; (2) use of quantitative data obtained at specified points within the aircraft following actual C-123A/A 45Y-1 dissemination missions using a Herbicide Orange Simulant; and (3) use of a combination of the above two methods. Discussion with personnel of the U.S. Army Environmental Hygiene Agency and the Chemical Systems Laboratory, Edgewood Area, Aberdeen Proving Grounds, Maryland, suggest that the dispersion of herbicide vapors and particles within the C-123 aircraft could be modeled provided data could be obtained for selected parameters. These parameters include:

(1) Air flow measurements within the aircraft under flying conditions simulating those encountered during actual RANCH HAND missions.
(2) Physical properties of Herbicide Orange components.
(3) Internal volume of aircraft and individual compartments.
(4) Temperatures of internal surfaces.
(5) Estimate of surface area on the floor of the aircraft.

Given a specific scenario, for example, a 25 sq ft spill involving two gallons of Herbicide Orange on the aircraft floor during a mission, the model could determine the magnitude and duration of the chemical concentration in the areas near the console operator versus the air concentration in the cockpit. They key to determining the magnitude and route of exposure of C-123 aircrew members is obviously selecting the most likely aircraft configuration used during RANCH HAND and determining the frequency and magnitude of spills within the aircraft. These observations will come from the recollection of personal observations by RANCH HAND personnel.

The 355th TAS/Spray Branch, Rickenbacker AFB OH is presently using the C-123 aircraft configured with the A/A 45 Y-1 Internal Dispenser. Air flow measurements, volume determinations, and surface temperature data will be obtained for modeling and simulant studies. A suitable Herbicide Orange Simulant was developed by the USAF in 1965 and consisted of glycerine, water, and sodium thiosulfate in a ratio of 68:16.8:15.2, respectively. Since physical and chemical data are available on the simulant, the addition of a sodium fluorescein tracer may permit particulate sizing and quantitative determinations of the extent of contamination in the air of the aircraft and on the surfaces of the aircraft and on aircrew members.

An estimate of relative exposure for non-flying personnel will be developed in a different manner. The Air Force conducted extensive industrial hygiene monitoring programs during the dedrumming and incineration of Herbicide Orange, Project PACER HO, (See Young, et al., 1978). These monitoring data (e.g., breathing zone data) and recently conducted, but as yet unpublished data on percutaneous absorption of 2,4,5-T in humans during actual spray operations in reforestation programs (Dow Chemical U.S.A., Midland MI, 1979), will permit more refined calculations of exposure estimates for this group of individuals. When these concepts are used to calculate an exposure estimate, there will be a group of RANCH HAND individuals with an exposure index of zero (clerical/administrative personnel with little or no flight line duties).

In the event that these more elaborate exposure index calculations are not feasible, two other approaches to the exposure estimate are available. A crude index can be constructed, based solely on the duration of RVN duty and the TCDD concentration of the herbicides used each year. While such an index would be less precise in defining an individual's estimated level of exposure, it would still permit valid comparisons within the RANCH HAND population. Stratification techniques could also be employed using AFSC and duration of exposure (RVN duty) as the grouping criteria.

VI. Statistical Methodology

A. Introduction

The design of the study is presented in schematic form in Figure 6. R' refers to the RANCH HAND personnel and C" refers to the collection of all possible control individuals. As defined, R' and C" will contain individuals who are deceased. Since C" may be 15 to 25 times larger than R', a randomized subsample C' of C" will be obtained. C' will be constructed from C" by computer selection of the ten best matched controls for each exposed study subject. As previously noted, close matches will be made for the variables of age, AFSC, RVN tour length, and race. The matched controls will form ten cohorts, C_1 through C_{10}, as previously shown in Figure 1. A 50% random sample from each of the matched control sets of 10 will be selected for inclusion in the mortality assessment so that a group, C' is obtained that consists of 5 matched controls for each exposed subject. These controls will be designated as initial replacement candidates for the morbidity and followup studies. The remaining individuals in the control set will be additional replacement candidates in the event that replacement must occur beyond the members of the mortality set (see Figure 2). C' will be constructed without regard to whether the individual is currently living or dead so that an assessment of mortality can be accomplished.

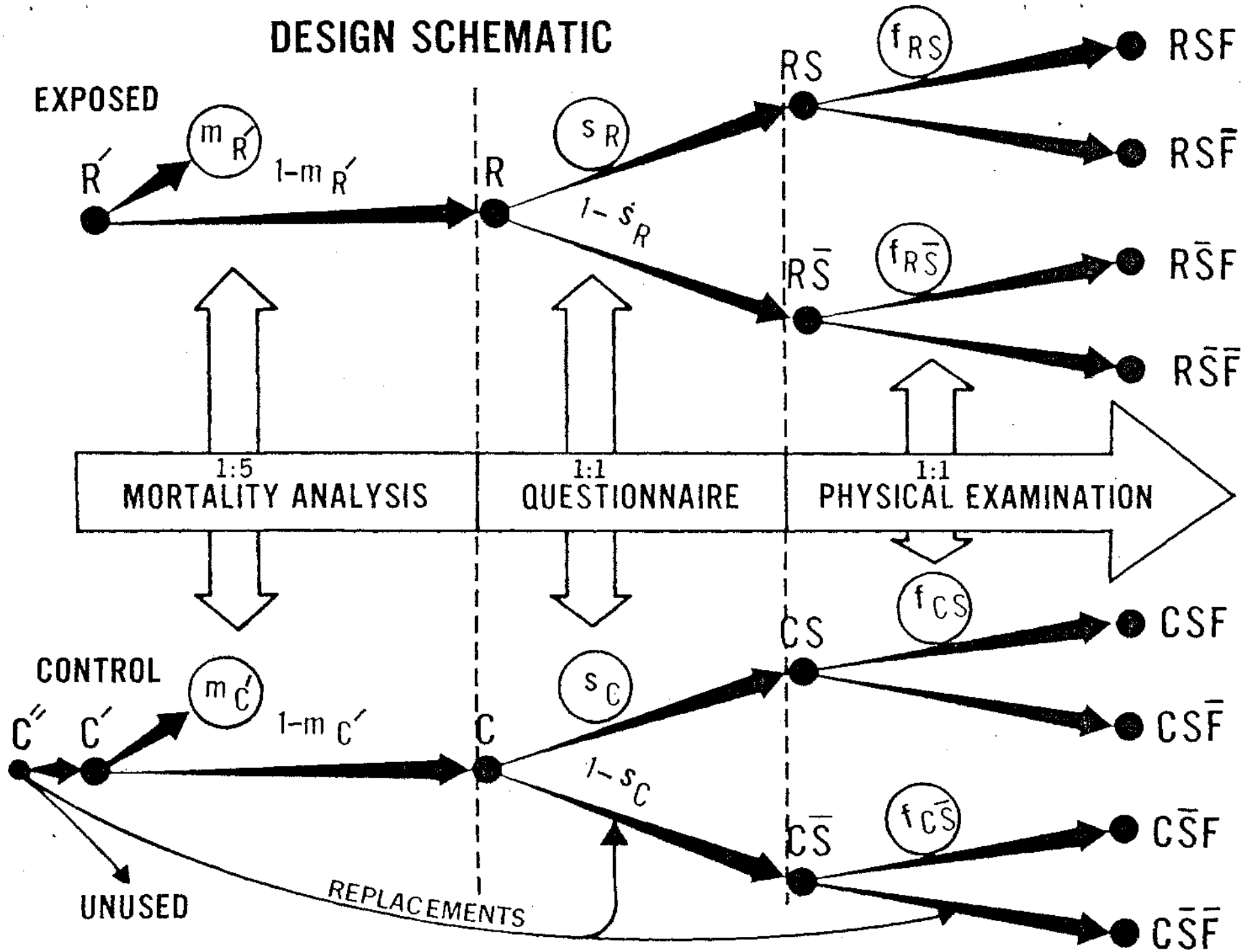
Referring again to Figure 6, R and C indicate living RANCH HAND members and primary matched controls. If $m_{R'}$ is the proportion of R' found to be deceased, then,

$$R = (1 - m_{R'})R'$$

The questionnaire will provide data concerning specific symptoms and other findings in the R and C groups. Thus, various questionnaire finding rates in R, s_R, will be calculated and compared with the corresponding rates in C, s_C.

The questionnaire will allow allocation of RANCH HAND personnel into those with symptoms on questionnaire, indicated by RS, and those without, $\overline{RS}$. Similarly, the control individuals will be placed into symptomatic, indicated CS, and asymptomatic, $\overline{CS}$ groups.

The physical examination performed on individuals from R and C will allow estimation and comparison of rates of physical findings in these groups. Rates of abnormal physical findings can be symbolically indicated as f_R and f_C for RANCH HAND and control groups respectively. Comparison of these rates is very important and details will be discussed below.


DESIGN SCHEMATIC
EXPOSED
R'
m R'
1-m R'
R
s R
1- s R
RS
f RS
RSF
RSF̄
RS̄
f RS̄
RS̄F
RS̄F̄
1:5
MORTALITY ANALYSIS
1:1
QUESTIONNAIRE
1:1
PHYSICAL EXAMINATION
CONTROL
C''
C'
m C'
1-m C'
C
s C
1- s C
CS
f CS
CSF
CSF̄
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f CS̄
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CS̄F̄
UNUSED
REPLACEMENTS

Let f_{RS} be the rate of physical findings among RANCH HAND personnel with findings by questionnaire and let $f_{R\bar{S}}$ be the rate of physical findings among RANCH HAND people with no findings on their questionnaire. For most disease processes it would be expected that f_{RS} should be a larger rate than $f_{R\bar{S}}$. If f_{RS} is observed to be equal to or less than $f_{R\bar{S}}$, an interpretation of over-reporting may be warranted, although the possibility of subclinical disease is recognized. Rates f_{CS} and $f_{C\bar{S}}$ will also be estimated, and comparisons between f_{RS}, f_{CS}, $f_{R\bar{S}}$ and $f_{C\bar{S}}$ will be accomplished.

The eight rates m_R', m_C', s_R, s_C, f_{RS}, $f_{R\bar{S}}$, f_{CS}, $f_{C\bar{S}}$ and their refinements fully characterize this study. As depicted in Figure 6, "vertical comparisons of these rates provide relative risks m_R'/m_C', s_R/s_C, f_R/f_C, f_{RS}/f_{CS} and $f_{R\bar{S}}/f_{C\bar{S}}$ which are of central importance in defining herbicide effects. "Horizontal comparisons" relate f_R to s_R, f_{RS} to $f_{R\bar{S}}$, f_C to s_C and f_{CS} to $f_{C\bar{S}}$. Specifically, the ratio f_R/s_R is the ratio of physical findings to reported symptoms in the RANCH HAND population. This ratio may be contrasted with the ratio f_C/s_C and if f_R/s_R is less than f_C/s_C overreporting can be inferred. Likewise, if f_{RS} is less than $f_{R\bar{S}}$, overreporting is further suggested. A comparison of $f_{RS}/f_{R\bar{S}}$ to $f_{CS}/f_{C\bar{S}}$ contrasts the odds of findings given symptoms in the RANCH HAND population with the odds of findings given symptoms in the control group. If these odds are lower in the RANCH HAND group, over-reporting is again implied. However, the existence of subclinical disease as the cause of these ratios cannot be excluded.

During the questionnaire and physical examination phases of this study, only one of the five randomly selected mortality study controls will be used for each RANCH HAND individual. If this control is unwilling to participate, another mortality study control will be used as indicated in Figures 2 and 5. These replacements will be carefully labelled for purposes of statistical comparison to the lost controls. Analysis without the replacements will also be accomplished, so that there will be no inferential loss from the use of replacements. A more detailed discussion of this replacement concept is found in section I below.

B. General Comments

Before proceeding to statistical details regarding this protocol, general areas of the overall design will be discussed.

(1) Adequacy of Sample Sizes

The size of R' is approximately 1000 individuals. It is clear that a lethal effect of herbicide which occurs in only 1 out of 2000 controls will be quite difficult to detect unless the herbicide effect is very strong. For example, at a rate of 1 in 2000, 0.5 affected controls are expected. If the basic rate is

doubled by herbicide to 2 per 2000, one affected RANCH HAND individual would be expected. At a rate of 1 per 2000 for controls and a rate of 2 per 2000 for RANCH HAND personnel, the probability of observing no affected individuals in both groups is

$$(1 - 1/2000)^{1000} (1 - 2/2000)^{1000} = .22$$

or, in other words, "there is a 22% chance" that no affected individuals will be found in this study. In a population of 100,000 exposed individuals, 100 cases would be expected, 50 of which would be due to herbicide. In short, since the size of the RANCH HAND group is fixed, this study has limited statistical power to define the relationship of herbicide to the rarer diseases.

(2) False Reporting/Misrepresentation

To understand the effect of misrepresentation on estimates of relative risk and the odds ratio, let S stand for presence of a symptom, and $\bar{S}$ denote its absence. This false reporting may be represented as in Figure 7.

FIGURE 7

FALSE REPORTING/MISREPRESENTATION

		TRUE STATUS: S	TRUE STATUS: $\bar{S}$	TOTAL
REPORTED STATUS	S	A	B	A + B
	$\bar{S}$	C	D	C + D
		A + C	B + D	

The proportion of correctly classified positives is defined by A/(A+C) and is called the sensitivity of the classification scheme; the proportion of correctly classified negatives D/(B+D) is called the specificity.

When there is non-differential misrepresentation, that is, when the sensitivity and the specificity are the same among the exposed and nonexposed, the bias induced in the estimate of relative risk will be toward the null value. The situation is summarized by Figure 8.

FIGURE 8

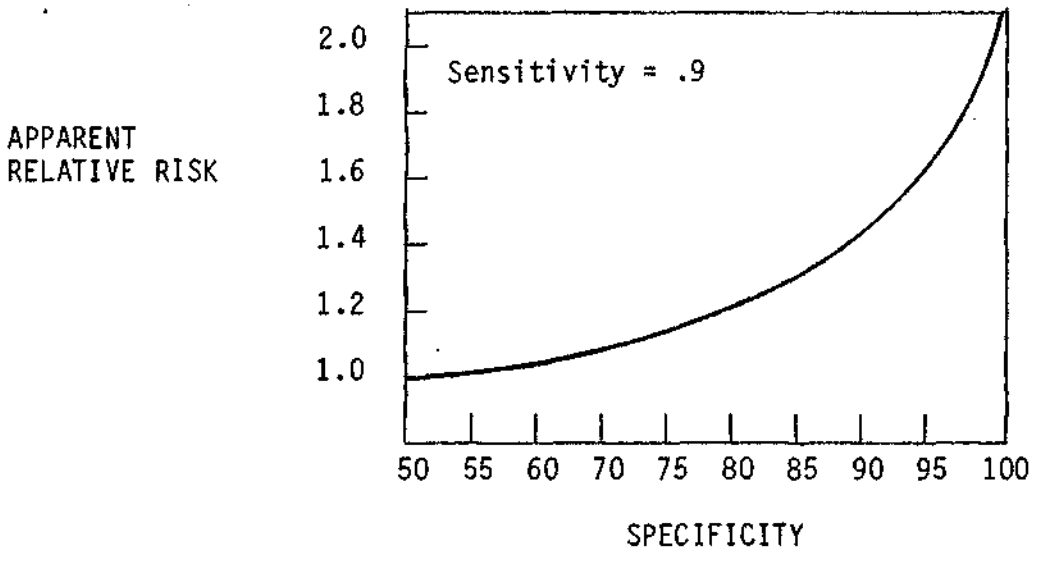
MISREPRESENTATION IN RANCH HAND II

		TRUE STATUS					
		EXPOSED			NONEXPOSED		
		S	$\bar{S}$	TOTAL	S	$\bar{S}$	TOTAL
REPORTED STATUS	S	a	b	a + b	e	f	e + f
	$\bar{S}$	c	d	c + d	g	h	g + h
		a + c	b + d	n	e + g	f + h	n

Using this representation, the true relative risk is $(a+c)/n \div (e+g)/n$, and the apparent relative risk is $(a+b)/n \div (e+f)/n$. Figure 9 provides a graphic representation of how apparent relative risk varies as a function of specificity. For this curve, the true relative risk is 2 with the exposed population having a symptom incidence of 0.1 and the nonexposed population having a symptom incidence of 0.05 (Copeland et. al. 1977). The effect of nondifferential false reporting on the odds ratio is nearly as severe as that shown in Figure 9 for relative risk. A technique does exist for correcting the estimate of relative risk to account for false reporting, but the technique requires knowledge of the sensitivity and specificity of the classification scheme, knowledge that may not exist in this study. It should be noted that since the above remarks are concerned with relative risk, the number n of subjects in each group is irrelevant, as the results shown are independent of n.

If the false reporting is differential, an estimate of relative risk that is biased away from the null value can result. This will occur in situations in which the RANCH HAND personnel and controls do not misrepresent their symptoms in the same manner (Copeland et. al. 1977). Thus the "true" outcomes of herbicide exposure may be distorted depending upon the degree and direction of misrepresentation.

FIGURE 9
APPARENT RELATIVE RISK VERSUS
SPECIFICITY



C. Analysis of Mortality Data

Considering the basic groups R' and C', individuals will be classified into three categories: alive, dead, unaccounted. If a large number of individuals of each group are unaccounted for, the study can obviously be severely biased. Thus, significant effort will be expended to reduce the unaccounted category as much as possible. At most 1 to 3 percent of both groups can be allowed to remain unaccounted, with a 1% rate being preferred. If for example, the mortality rate in C' is 0.10, then an unaccountability rate of 0.01 could alter the mortality rate by as much as 10%. Whatever the unaccountability rates, the pattern of unaccountability must also be compared between groups R' and C'. For example, the possibility of age differences or RVN tour length differences must be examined, particularly if the unaccountability rates are high. The following will discuss analysis of mortality under the assumption that low unaccountability rates have rendered the mortality analysis meaningful.

Two mortality analyses will be accomplished, one at the beginning of the study, using available mortality data on the basic cohorts C' and R'(1:5 ratio), and one several years later, using mortality data on R' and all controls used in the study (both C' and replacements) as it accumulates prospectively. The procedures described here and in Section E below can be used in both

analyses. Henceforth, within the protocol, the term "mortality data" does not distinguish between that data collected initially and that data collected in the future.

The mortality data will be analyzed using several different approaches. Crude age-specific death rates will first be calculated and tabulated. Age will be divided into k strata, and person-years will be observed for each strata as will be the number of deaths in each strata. In this manner a tabular display will be developed as shown in Table 7.

TABLE 7

STRATIFIED FORMAT OF AGE-SPECIFIC DEATH RATES

	Ranch Hand			Controls		
Age Group	Person Years	Deaths	Death Rate	Person Years	Deaths	Death Rate
1	P_{11}	m_{11}	r_{11}	P_{21}	m_{21}	r_{21}
2	P_{12}	m_{12}	r_{12}	P_{22}	m_{22}	r_{22}
3	P_{13}	m_{13}	r_{13}	P_{23}	m_{23}	r_{23}
.	.	.	.	.	.	.
.	.	.	.	.	.	.
.	.	.	.	.	.	.
k	P_{1k}	m_{1k}	r_{1k}	P_{2k}	m_{2k}	r_{2k}

Since the death rates r_{1j} and r_{2j} are Poisson variables, they can be contrasted directly. If the relationship of r_{1j} to r_{2j} is found to be consistent between age strata (within statistical variability), a summary mortality index may be calculated. One summary index that will be calculated is Standardized Mortality Ratio (SMR) which is (Armitage, 1971):

$$SMR = M \times 100$$

$$M = \frac{\sum_{j=1}^{k} m_{1j}}{\sum_{j=1}^{k} P_{1j}\, r_{2j}}$$

"Classical" standardized mortality ratios using national mortality data as the reference will not be calculated for RANCH HAND II due to the effects of the healthy worker phenomenon. The term $\Sigma\, m_{1j}$ is the total number of deaths observed in the RANCH HAND group while $\Sigma\, p_{1j}\, r_{2j}$ is the number of deaths that would be expected were the age-specific RANCH HAND death rates the same as the age-specific control death rates. Thus the concern is for an SMR greater than 100%. If a crude death rate for controls, d_c, is calculated as

$$d_c = \frac{\sum_{j=1}^{k} p_{2j}\, r_{2j}}{\sum_{j=1}^{k} p_{2j}}$$

then the standardized crude rate for the RANCH HAND group d_{RH} is

$$d_{RH} = M d_c .$$

An approximate statistical test would regard r_{RH} as a Poisson random variable with mean d_c.

An alternative approach to the provision of a proportionate mortality ratio is that of Breslow and Day (1975). In this treatment, a multiplicative model is employed, for example:

$$\lambda_{ijk} = \theta_i \phi_j \psi_k$$

where λ_{ijk} is the mortality rate, θ_i is the contribution due to population differences (RANCH HAND versus Control), ϕ_j is the contribution due to age group, and ψ_k is the contribution due to tour length, etc. The statistical approach here is via maximum likelihood.

Logistic models (Walker and Duncan, 1967) have been extensively studied at USAFSAM for application in cardiovascular disease. These models, in the herbicide context would have the form

$$P = [1 + \exp(\alpha + \beta_1 A + \beta_2 T + \beta_3 R + \beta_4 E + \beta_5 AE + \ldots)]^{-1}$$

where

P = probability of death
A = age in years
T = Vietnam tour length
R = indicator variable for race
E = exposure variable

and where α_1, β_i, i=1,2,... are coefficients to be estimated from the data. Testing for a group difference can be accomplished by estimating β_4 and interaction coefficients such as β_5. If all interaction coefficients involving the exposure variable E are zero and E is treated as a 0/1 variable, Cox (1958a, 1958b) has shown that the most powerful test for non-zero β_4, in the setting of matched pairs, is McNemar's test. This latter test makes full use of the paired design of the study. For McNemar's test, the data are cast into a 2 x 2 table as shown in Table 8. In this table,"a" is the number of pairs in which both members have died, "b" is the number of pairs in which only the RANCH HAND person has died, etc. Using McNemar's test, the test statistic

$$\chi^2 = \frac{|b - c|^2}{b + c}$$

is calculated and referred to the chi-square distribution with one degree of freedom. Cox (1966) and Meittinen (1969) provide extensions of McNemar's test for R controls per exposed (R-to-1 matching). Of course the above analyses will be accomplished considering all deaths, and deaths by specific cause.

TABLE 8

FORMAT OF McNEMAR'S TEST

CONTROLS

RANCH HAND PERSONNEL	DEAD	ALIVE	TOTAL
Dead	a	b	a+b
Alive	c	d	c+d
Total	a+c	b+d	n

As discussed in section V above, RANCH HAND personnel may be characterized as risk takers. This risk taking behavior may be associated with increased mortality from a variety of causes. If herbicide exposure has caused neuropathy in the RANCH HAND personnel, one could anticipate that this disability would increase the probability

of accidental death. Therefore, accidental death rates among RANCH HAND participants will be corrected for risk taking. This can be accomplished by including assessment of risk taking behavior in the questionnaire, indepth interview and psychological evaluation. Both control and RANCH HAND mortality could be corrected using these measures, with the resultant rates being less biased and, therefore, a better indicator of exposed versus control effect.

D. Analysis of Questionnaire and Physical Examination Data

The Questionnnaire and Physical Examination will produce data of three types: (1) dichotomous, (2) polytomous and (3) continuous.

Dichotomous (present-absent) rates will be evaluated using the tools described above for mortality analysis. For example, the questionnaire will provide data concerning the first occurrence of disease states by age, and standardized rates and relative risks may be calculated. The occurrence of such findings can be related to age, tour length exposure and other variables using logistic models followed by McNemar's test where appropriate. These tests will examine the presence or absence of group effect and allow assessment of the statistical significance on non-unity relative risks.

Polytomous findings will occur in both questionnaire and physical examination responses. As an example consider retinal findings categorized into four grades, and studied as a function of age and exposure group as represented in Table 9. In this table the x_{ijk}'s are counts of occurrence. In analyzing tables such as these, techniques as described by Bishop, Fienberg and Holland (1975) will be used. Specifically, if m_{ijk} is the expected value of x_{ijk}, general log-linear models of the form

$$\ell n\, m_{ijk} = u + u_1(i) + u_2(j) + u_3(k) + u_{12}(ij) + u_{13}(ik) + u_{23}(jk) + u_{123}(ijk)$$

will be used, where $u_1(i)$ is the effect of RANCH HAND membership alone on cell frequency, $u_{12}(ij)$ is the effect of an interaction on RANCH HAND membership with retinal grade, etc. This model can work with dichotomous as well as polytomous data. Under appropriate conditions on expected values of entries in Table 9, the pairing in the study design can be used with the data being organized as shown in Table 10. In Table 10, N_{ij} is the number of pairs such that the exposed person has retinal grade i, and the control person has retinal grade j. Appropriate tests for this setting are indicated by Fleiss (1973).

With regard to continuous variables, the intended method follows Carpenter (1977) who found substantial gains in analysis efficiency by matching cases, subsequently employing covariance analysis to remove non-controlled effects.

TABLE 9

FORMAT OF CATEGORICAL REPRESENTATION OF RETINAL CHANGES

	RANCH HAND PERSONNEL				CONTROLS			
Age Category / Retinal Category	1	2	3	4	1	2	3	4
1	X_{111}	X_{112}	X_{113}	X_{114}	X_{211}	X_{212}	X_{213}	X_{214}
2	X_{121}	X_{122}	X_{123}	X_{124}	X_{221}	X_{222}	X_{223}	X_{224}
3	X_{131}	X_{132}	X_{133}	X_{134}	X_{231}	X_{232}	X_{233}	X_{234}
4	X_{141}	X_{142}	X_{143}	X_{144}	X_{241}	X_{242}	X_{243}	X_{244}

TABLE 10

FORMAT OF PAIRING FOR GRADES OF RETINAL FINDINGS

	Control Grade			
RANCH HAND Grade	1	2	3	4
1	N_{11}	N_{12}	N_{13}	N_{14}
2	N_{21}	N_{22}	N_{23}	N_{24}
3	N_{31}	N_{32}	N_{33}	N_{34}
4	N_{41}	N_{42}	N_{43}	N_{44}

Analysis of Fertility/Reproduction Data. The herbicides under consideration in this study have been alleged to effect fertility and/or reproductive functioning. An attempt will be made to address these allegations by analyzing at least three primary variables: the total number of conceptions since exposure in RVN, the number of miscarriages in spouses since exposure in RVN and, the number of abnormal offspring since exposure in RVN. The study questionnaire will provide the number of miscarriages, abnormal offspring and of live births. The sum of the number of miscarriages,

still births, and live births will provide an estimate of the total number of conceptions. If differing divorce rates are found in the RANCH HAND and control groups, this may render the average number of years of marriage and the distribution of the years of marriage different in the two groups. This will be investigated and adjusted for if need be, either by analyzing total number of conceptions divided by (or normalized by) the number of years of marriage, or by using a more detailed covariance analysis. Further, the ratio of the number of miscarriages to adjusted total conceptions will be calculated and compared as will be the ratio of the number of abnormal births and adjusted total conceptions.

In summary, the following statistics relating to fertility will be calculated and analyzed at the very least:

$$\text{TOTAL CONCEPTIONS} = \#\text{Live Births} + \#\text{Still Births} + \#\text{Miscarriages}$$

$$\text{NORMALIZED FERTILITY INDEX} = \frac{\text{TOTAL CONCEPTIONS}}{\text{YEARS OF MARRIAGE}}$$

$$\text{MISCARRIAGE FRACTION} = \frac{\#\text{ MISCARRIAGES}}{\text{TOTAL CONCEPTIONS}}$$

$$\text{ABNORMALITY FRACTION} = \frac{\#\text{ ABNORMAL OFFSPRING}}{\text{TOTAL CONCEPTIONS}}$$

E. Survival Analysis

This section extends and complements sections C and D. The defining common attribute of the techniques discussed in this section is that they deal with events which (a) correspond to categorical changes in health status, and (b) occur at definite and observable times. These methods may, therefore, be applied to studies of mortality (as the name "survival analysis" implies) as well as to studies in morbidity.

Survival analysis without covariates. The first step in the statistical analysis of survival data is descriptive, i.e., the construction of summary measures which provide a basis for comparing different exposure groups without any allowance for the effects of possibly confounding variables (e.g., age) except perhaps for some limited stratification. Since one must expect many "losses to follow-up", only methods which take full cognizance of this complication will be considered. It should be pointed out that all the methods described below assume independence between censoring (e.g.,

loss to follow-up) and death or morbid event, although some techniques permit different patterns of censoring in different exposure groups.

The life table method can be adapted to obtain a step-function approximation to survival distributions in the presence of censoring (Chiang, 1968, Gross and Clark, 1975). However, the product-limit estimator of Kaplan and Meier (1958) may be preferred due to its intrinsic properties and its relationship to more refined methods.

The failure time distribution is the function $F^0(t)$ which provides the probability of death at or before time t in the study. The Kaplan-Meier estimator of $F^0(t)$ is $\hat{F}^0(t)$ where

$$\hat{F}^0(t) = 1 - \prod_{i \in D(t)} [1 - 1/R(T_i)]$$

In this equation, D(t) is the "death set" at time t, i.e., the set of all indices i of individuals who were observed to fail before time t. $R(T_i)$ is the number of individuals who were at risk just before time T_i, the time of death (or morbid event) of the i^{th} study individual in D(t). This product-limit estimator of Kaplan and Meier is maximum likelihood in the class of all possible failure time distribution functions.

Assuming that failure time distributions have been calculated for RANCH HAND individuals and controls, the next question concerns testing the null hypothesis of equality between the distributions. When only two such distributions are being compared, one may use the nonparametric procedures generalizing Wilcoxon's statistic proposed by E. Gehan (1965a, b) and discussed by N. Mantel (1967). When more than two such distributions are being compared, one may use the nonparametric procedure generalizing the Kruskal-Wallis statistic proposed by N. Breslow (1970).

Survival analysis with covariates. These methods allow adjustment of mortality rates or morbidity rates using covariates such as age, race, RVN tour length, AFSC, risk taking score etc. For the purposes of this discussion it will be assumed that the covariables are categorical, that there are only two such covariables and that the covariables do not interact in affecting the hazard of death or morbidity. These assumptions can all be relaxed using available methods.

The hazard function $h_i(t)$ for the ith individual in the study is the function which provides the conditional probability of death or morbid event in the time interval (t, t+dt) given his survival up to time t. The function $H_i(t)$ where

$$H_i(t) = \int_0^t h_i(\tau)d\tau$$

is called the cumulative hazard for the ith individual. It is readily shown that the failure time distribution $F_i^0(t)$ is given by:

$$F_i^0(t) = 1 - \exp(-H_i(t))$$

From this last equation it follows that h_i and F^0 are transforms of each other, whence the dependence of F^0 on covariables may be modeled via h_i. This may be accomplished as follows. Let $X_i(t)$ and $Y_i(t)$ denote discrete valued stochastic processes pertaining to the ith individual and describing two covariates of interest (e.g., one may be an exposure variable and the other may be covariate such as age or crew position). The basic model for hazard is:

$$h_i(t) = \exp [\xi X_i(t) + \eta Y_i(t)]$$

where ξ and η are "log-relative risks". This model (Frank, 1977) may be extended to allow for any number of possibly interacting factors. Inference about log-relative risks may be drawn using either an approach derived from D. R. Cox (1972) by E. Peritz and R. Ray (1978) or using an approach described by Frank (1977).

F. Statistical Power

The power $(1 - \beta)$ of a study design is the probability that a specified difference between populations will be detected if it in fact exists. In general, power is a direct function of sample size; that is, for a particular study design, the more subjects measured the larger the study power. It is understood that these analyses make use of the entire known RANCH HAND population (and exclude ancillary exposed groups for reasons previously cited); the exposed sample size cannot be increased. Power augmentation, therefore, can only be accomplished by the less efficient procedure of increasing the control group size which has statistical limitations as well as staggering financial and logistic considerations. Hence, considerable effort has been made to correct loss to study issues (by replacement and other techniques to induce participation) and to use the most powerful statistical design concepts. Essentially all previous animal and human studies concerning herbicide suffer from a lack of adequate consideration of study power. The following presents a preliminary analysis of study power for the case of continuous and dichotomous variables expected from the study.

(1) Power in Continuous Variable Case

Assume that blood cholesterol levels are being compared between RANCH HAND and control groups, and that the coefficient of variation for cholesterol in the control group is 0.1, where the coefficient of variation is the ratio $\sigma c/\mu c$. Assume $\sigma_{RH} = \sigma_C$. The symbol α is the probability that the study will indicate an effect where none exists, and $1-\beta$ is the power as defined before. Consider that the RANCH HAND mean cholesterol μ_{RH} is shifted from the control mean μ_C. A natural question is to inquire about the study power as a function of available pairs (n) and mean ratio $\gamma = \mu_{RH}/\mu_C$.

TABLE 11

POWER CALCULATIONS

ASSUMPTIONS: $\alpha=0.05$, $\sigma_C/\mu_C=0.1$, $\gamma=\mu_{RH}/\mu_C$

		Power = 1-β	
r	γ	n=180	n=450
.20	1.01	.20	.38
.20	1.02	.55	.88
.20	1.05	>.995	>.995
.70	1.01	.86	.995
.70	1.02	>.995	>.995
.70	1.05	>.995	>.995

Power calculations are displayed in Table 11. Study power in the case of a matched pair design is strongly dependent on the degree of positive correlation produced between the involved groups by the matching procedure. Of course, the degree of correlation can be expressed by the correlation coefficient r which can take values between -1 (negative correlation) and + 1 (positive correlation), and two values of r have been employed in Table 11. From this table it is seen that if only 450 pairs are studied a 1% shift in mean (= 1.01) will not be reliably detected, but a 2% shift will be detected with a probability of 0.88 if r = 0.2 at least. From this calculation one can infer the need to examine at least 450 pairs to obtain the 2% shift, and to strive for more if possible.

(2) Power in the Dichotomous Variable Case. There is significant discussion in the mathematical statistics literature concerning the efficacy of paired designs in the setting of dichotomous responses (Billewicz, 1974; Ury, 1975; Miettinen, 1970; and several others). Table 12 shows a set of calculations which are applicable to the present study.

TABLE 12

POWER CALCULATIONS FOR THE DICHOTOMOUS VARIABLE CASE AS A FUNCTION OF EFFICACY OF PAIRED DESIGNS

				POWER = 1 - β			
P_1	P_2	Rel. Risk	r	n= 250	n= 350	n= 450	
.05	.01	5	0	.77	.82	.92	*
.04	.01	4	0	.61	.75	.85	
.03	.01	3	0	.40	.51	.59	
.10	.05	2	0	.61	.75	.85	
.20	.10	2	0	.87	.94	.97	
.05	.01	5	.1	.89/.029	.94/.032	.98/.064	**
.04	.01	4	.1	.72/.033	.87/.038	.88/.041	
.03	.01	3	.1	.38/.020	.68/.046	.71/.077	
.10	.05	2	.1	.76/.055	.85/.048	.88/.048	
.20	.10	2	.1	.94/.043	.98/.046	.99/.057	

*α = .050**
**α as indicated

In this figure, r is again the correlation coefficient indicating the degree of correlation induced between the involved groups by the matching procedure. The probability of the disease among RANCH HAND personnel is symbolized as p_1, while p_2 is the probability of the disease among the controls. Relative risk is the ratio p_1/p_2. With r = 0.1, sign test power tables were used as an exact version of McNemar's test, and therefore different α levels are shown under each power number. Table 12 shows the positive influence of effective pairing in the higher power levels noted. For example, it appears that for p_2 = 0.01 and p_1 = 0.03, physical examination of 450 pairs (900 examinations) will disclose the three-fold relative risk with probability less than the minimum target .80. In other words, there is a greater than "20% chance" that a three-fold relative risk on a 1/100 disease state will go undetected in this study if only 350 pairs are examined and if low correlations occur. Once again the need to examine the maximum numbers of pairs in the study is seen.

To present these dichotomous power calculations more clearly, calculations in the context of actual disease states have been accomplished. The diseases considered are cardiovascular disease and cancer, corresponding to high and low rate illnesses for the age groups presently under investigation.

Cardiovascular Disease. A logistic risk function was fitted to data from 17,455 autopsies gathered in a WHO collaborative study in Czechoslovakia, Sweden and the USSR. The function fitted has the form

$$P = [1 + \exp(\alpha + \beta(x-.5) + \gamma(y-.5))]^{-1}$$

where

p = the probability of a complicated coronary lesion

x = age scaled linearly so that x = 0 is equivalent to 3 years, and x = 1 is equivalent to 58 years (the age span of the current study)

y = 1 or 0 if the subject is exposed or not

and α and β were obtained from the data. The function represents a fairly high rate disease in that at 40 years of age 7% of the group had the lesion and at 60 years of age 20% had the lesion. The coefficient γ, represents the exposure effect. Power calculations for $\gamma = \beta$ and $\gamma = .8\beta$ are shown in Table 13. This figure suggests that if, as a cell toxin, herbicide exposure accelerates cardiovascular disease, this study has a good chance of detecting that acceleration if the herbicide effect is comparable to the age effect. A slight beneficial effect of pairing is seen in this hypothetical example.

Cancer. A logistic risk function was fitted to breast cancer data presented by Breslow and Day (1975). The function fitted represents a low rate disease in that at 35 years of age only .000336 of the group had the lesion while at 70 years of age .00676 of the group will have the lesion. Using pairing to achieve a power of 0.80 in this setting, 1312 pairs would be needed, when the exposure effect is equal to the age effect. This exceeds the size of our RANCH HAND cohort, and reinforces the fact that herbicide exposure effects on rarer diseases will not have a high likelihood of being detected by this study, and again supports an attempt to examine as many pairs as possible.

TABLE 13

POWER CALCULATIONS AS A FUNCTION OF HERBICIDE EFFECT

ASSUMPTION: $\alpha = 0.05$

Number of Pairs	$\gamma = \beta$		$\gamma = .8\beta$	
	Power Neglecting Pairing	Power With Pairing	Power Neglecting Pairing	Power With Pairing
100	.93	.93	.64	.53 ($\alpha = .036$)
160	>.97	.98	.81	.82
200	>.99	>.995	.86	.87
250	>.99	>.995	.93	.95
300	>.99	>.995	.96	.97
350	>.99	>.995	.97	.98

G. Multivariate Analysis

Some questionnaire and physical examination data naturally fall into groups; for example, fertility/reproduction data, liver function tests, cardiovascular examination tests. In these cases, multivariate analysis may be in order. When the response variables are continuous, they will be analyzed by the well known multivariate extensions of the generalized linear model.

The general approach to multivariate analysis of polytomous data considers all classification factors and all variables as "factors" in a multi-way contingency table. Log-linear models as described above for polytomous data will be employed where appropriate.

H. Indices and Estimates of Exposure

Exposure estimates will be used to sharpen the statistical analysis and can be helpful in summarizing responses. In this discussion, two estimates will be considered: one related to Vietnam herbicide exposure, and another related to domestic (US) herbicide exposure independent of Vietnam experience.

Vietnam herbicide exposure. In the above discussion of statistical methodologies, exposure variables appeared. In the logistic formula on page VI-8, the variable E was shown which could

be either dichotomous, polytomous, or continuous. In the use of logistic functions to discuss study power, the exposure variable was taken as dichotomous. When a polytomous or continuous exposure variable E is constructed, significant sharpening of the study analysis will be accomplished. For example, biases in this study could lead one to suspect that differences between RANCH HAND personnel and controls were in fact due to factors other than a herbicide effect. If however, in addition to differences between RANCH HAND personnel and controls, one was able to show a regression of mortality and/or morbidity on an exposure index E, the case for a bonafide herbicide effect would be firmer. The reconstruction of exposures is discussed in Section V, G.

Domestic herbicide exposure. Individuals in both the RANCH HAND and control groups will have had varying exposure to herbicide in the United States. Particularly, individuals from specific farming and/or foresting areas, may have had and continue to have a significant background exposure. Data on place of residence and information concerning home practices (gardening etc.) could be used to build a background exposure index E_b. In lieu of constructing this index, stratification techniques will be used in the analytic phases of the study.

I. The Replacement Concept

In the mortality analysis, a randomly selected group of control individuals will be compared to the RANCH HAND group, and the data gathered will be analyzed for evidence of herbicide effect. In the questionnaire and physical examination phases of this study, one of the mortality controls will be randomly selected for each RANCH HAND individual. During the physical examination phase, we must anticipate a significant degree of unwillingness to participate, particularly on the part of control personnel. This loss to study can result in significant bias and loss in statistical power; thus the replacement concept has been developed to mitigate these consequences.

In this replacement strategy, we envision that 10 control individuals will be matched with each RANCH HAND person. This will be accomplished using computerized data files and the matching parameters of age, AFSC, RVN tour length, and race. With each RANCH HAND individual R_i there will be associated 10 controls C_{i1}, C_{i2}, C_{i3}, ..., C_{i10}. A 50% random selection from each of the control sets will be used for the mortality analysis. The first of these controls, C_{i1} will be employed in the questionnaire and physical examination phases of the study. If C_{i1} is alive, but unwilling to participate in the study, he will be replaced by another randomly selected participant with similar perception of health status. In order to avoid bias in mortality analyses, no dead control will be replaced. The randomized selection of subjects from each of the ten control cohorts for the mortality analysis will provide an assessment of the mortality pattern in each cohort. This in turn will decrease or eliminate any bias introduced by the replacement scheme.

It is important to emphasize that all replacement controls will be carefully flagged so that they may be treated separately in the statistical analysis. These replacements will be carefully compared to the lost controls to develop indicators of comparability (e.g., morbidity and mortality experience). The initial analysis will be performed on the intact exposed/control pairs. Additional analysis will be conducted on all pairs, both those intact and those with replaced controls. If we consider RANCH HAND individual R_i, with living control C_{i1}, we can calculate the probability that control C_{ik} will be available for the 1st, 2nd and 3rd physical examinations. To examine this question, a small computer Monte Carlo simulation was required. A short BASIC language computer program and glossary are included in Appendix Table A-6. This simulation examines the effect of non-participation expressed as two probabilities P_1 and P_2. Figure A-2 displays the expected participation by the RANCH HAND population, and control group participation is expected to be somewhat less. P_1 is the probability that when first asked to attend a physical examination, the control individual will not comply. P_2 is the probability that a control individual who has agreed once to a physical examination, will not comply for a subsequent examination. In general, P_1 may be greater than P_2. Note that the probabilities P_1 and P_2 must reflect all causes of non-compliance including morbidity and mortality. Table 14 displays a representative simulation run, which provides the number of controls required to find willing matches for 1000 RANCH HAND personnel.

The potential bias introduced by non-willingness in controls can be analyzed statistically. If $p_C(x)$ is the probability density function for compliant individuals and $p_{NC}(x)$ is the same function for non-compliant individuals, we have

$$p(x) = \alpha p_C(x) + \beta p_{NC}(x)$$

where $p(x)$ is the probability density function for the entire population and x is a vector of important health parameters available on each person. Since

$$\int p(x)dx = \int p_C(x)dx = \int p_{NC}(x)dx = 1$$

it follows that

$$\alpha + \beta = 1$$

and α and β may be viewed as coefficients which "mix" the two subpopulations.

TABLE 14

CONTROL DISTRIBUTIONS BY EXAMINATION
MATCHING 1000 RANCH HAND PERSONNEL

(P_1 = .70, P_2 = .25)

CONTROL COHORT	EXAMINATION NUMBER 1	2	3
C_1	318	237	177
C_2	211	188	156
C_3	131	133	136
C_4	96	101	97
C_5	74	89	90
	49	68	77
C_7	34	43	59
C_8	25	39	52
C_9	16	18	33
C_{10}	13	20	35
Number of Matching Failures	33	64	88

If M_C and M_{NC} are the means of the compliant and non-compliant subpopulations respectively, it can be shown that

$$M = \alpha M_C + \beta M_{NC}$$

where M is the mean of the entire population. From this last equation, it is clear that as noncompliant individuals are lost (i.e., β tends to zero, α tends to one), M tends to M_C. Thus the maximum bias is the quantity $M_C - M$.

In this study we propose to replace non-compliant individuals with matched RANCH HAND members, that is with individuals drawn from a population with density equal to or at least similar to $p_{NC}(x)$. The resulting new density is $P"(x)$ such that

$$p"(x) = \alpha" p_C(x) + \beta" \tilde{p}_{NC}(x)$$

where

$$\alpha" + \beta" = 1$$

$$M" = \alpha" M_C + \beta" \tilde{M}_{NC}$$

and where $\tilde{p}_{NC}(x)$ approximates $p_{NC}(x)$. If $\beta"$ is chosen to be close to or equal to β above, it appears that $M"$ can well approximate M, the true population mean. The difficulty in this approach will be to assure that the replacements are representative of the non-compliant individuals in all respects other than logistic factors impacting willingness to participate in the program.

Our proposed approach is to obtain sufficient data on the unwilling personnel so that a discrimination function of the form

$$D = f(h_1,\ldots,h_n;\ l_1,\ldots,l_1)$$

can be derived. This function is envisioned to have the following properties:

(a) larger values of D correspond to decreasing probabilities of compliance with the physical examination,

(b) the factors h_i relate to the subjects' health status, while the factors l_i relate to logistic difficulties (distance, job) which tend to preclude attendance at the physical. Factors to be considered in the formulation of this funciton are displayed in Table 15.

(c) D is an increasing function of each h_i and of each l_i,

TABLE 15

FACTORS AFFECTING COMPLIANCE

Health Status (h_i)	Logistic Difficulties (l_i)
Subjective Health Assessment (good/poor)	Time Away from Family
Current Utilization of Long-Term Health Care (Yes/No)	Time Away from Job
Absenteeism Pattern (Greater Than/Less Than Ten Lost Days in Past Six Months)	Distance to Examination Site
	Active Pilot
	Income (Greater than/Less than $17,000)

In the replacement scheme, controls substituted for non-compliant controls, should have identical health factors (h_i) as those individuals they replace. The only significant differences should be in the logistic factors (l_i). The replacement method should permit correction of non-compliance bias given that health factors h_i and logistic factors l_i are actually distinct. The determination of these two classes of factors will be made using data from the study itself. Specifically, the logistic factors l_i will be independent of health status to the degree testable by the quantity of data available in the study. This replacement strategy has two major advantages: selection bias reduction/estimation and cost reduction. Were replacements not employed, one would be compelled to start the morbidity study with a 4 to 1 or 5 to 1 design in order to insure an adequate number of participating controls on the third physical examination (see Table 14). Such a large control group for physical examination is very costly with little corresponding gain in study power and with no correction of the selection bias.

VII. Data Repository

Throughout the 6-year period of this investigation, data collection methods will be integrated by use of computer systems. A data repository will be established at the USAFSAM. Master files will be formed on each exposed member and for his matched control/controls. The individual master files will be keyed to one or more identifiers. Confidentiality of data will be maintained by the use of computer generated code numbers. Addresses and telephone numbers of all study subjects will be continually updated to insure adequacy of follow-up.

Individual data bits and their sources are as follows:

(1)	Questionnaire	a. Initial (telephone) b. Indepth interview (personal and telephone) c. Follow-up (telephone)
(2)	Psychological Battery	a. Initial b. Follow-up
(3)	Physical Examination	a. Initial b. Follow-up
(4)	Medical Records	a. Active duty b. VA c. Civilian d. Dependent
(5)	Historical Data	a. Military personnel files b. Flight records c. Military unit
(6)	Death Certificates and Autopsy Reports	a. Study members b. Dependents
(7)	Birth Certificates	a. Dependents

Mortality data will be obtained from individual medical records, VA records, the screening of personnal records, contact with family or personal physician, and other available information sources. Date of death (verified by death certificate) and cause of death (verified by death certificate and/or available autopsy reports) will be obtained. Cause of death will be expressed as an ICDA number.

The computer software for the data analysis phase will be prepared to assure proper data conversion, quality control and standardization of test measurements. Quality control areas will include verification of identification data, range checks, and identification/correction of ambiguous or conflicting data.

VIII. Recognized Study Difficulties and Corrective Measures

A. Medical Precedence

(1) Problem

A departure from the usual methodological approach characterizes this particular epidemiological investigation. Clearly there is no historical "roadmap of methodology" to conduct this study. Most occupational exposure studies use the presentation of an unusual disease to justify the initiation of a comprehensive study. A rare disease or a common disease in an uncommon site, or one with an unusual presentation appearing in space-time clusters, often in an unusual population or age group, usually generates the requirement for a new study. In the case of Herbicide Orange, the evidence for long-term human effects is tenuous and controversial. Despite the unique problems that this study possesses, such as the lack of clinically defined endpoints, there are many problems that it shares with other occupationally related exposure studies. For example, the question of a latent period in the development of symptoms/signs, the lack of accurate dose-response relationships, and the possibility of a synergistic effect with other toxins/carcinogens are all operating in this study. Since most cohort studies of occupational mortality use the general population as a standard for deriving the expected number of deaths, preemployment selection ("healthy worker" bias) affects the comparative experience. Age-standardized mortality ratios (SMR's) in general are 60-90 percent of the standard in the working population. Similar conflicting results can occur using the matched cohort method proposed in this study design. Statistical verification of the validity of utilizing such a control for a summary mortality index (e.g., SMR) has been infrequently attempted in the past. Inability to verify the validity of the more classical methods of comparing mortality will necessitate the use of multiplicative and/or logistic models to obtain a valid standardized mortality ratio.

(2) Corrective Measures

Study approaches generated by unprecedented occurrences of occupationally related medical complaints require novel approaches, and reorientation beyond standard methods. The success key to this study design is a series of effective, progressive, and helpful peer reviews (all of which have occurred to date and have been incorporated herein). Beyond even the immediacy of the current study is the growing problem of a myriad of occupationally-related exposures, both in the military and civilian sector, which will require similar epidemiological studies in the future in order to make some judgment as to whether or not an association is of causal significance.

B. Group Accountability Bias

(1) Problem

The numerous media presentations on "Herbicide Orange" issues have focused attention on the RANCH HAND group. Several attempts have been made to construct lists of former members of this group and thus the RANCH HAND population should be easier to locate and contact than the control population. This difference will be particularly evident with respect to reported mortality experience. The incentives for cooperation and study participation are likely to be greater in the exposed group than in the controls. Also, the close knit reunion association of former RANCH HAND personnel will lead to a more precise reporting of morbidity and mortality in that group. Such group identity tends to decrease the degree of unaccountability in the exposed group while its absence in the controls may lead to under ascertainment of mortality. This could then lead to the attribution of excess mortality in the exposed population.

(2) Corrective Measures

Unaccountability bias will be minimized by attempting to keep the percentages of unaccounted for study subjects below 1% in both exposed and control groups. The morbidity and mortality status of all individuals selected for the study will be strongly pursued utilizing a variety of techniques previously described.

C. "Risk Taking" Behavior Bias

(1) Problem

The early RANCH HAND aircrew population was an exclusively volunteer group; the C-130 control population, while volunteers in the Air Force, were not volunteers for special hazardous missions. RANCH HAND mission conditions were considered to be more dangerous than those encountered in the normal combat environment. This suggests that some differences may exist in the psychological profiles of the two groups. A sensation seeking or risk taking psychological orientation may have altered the accident mortality or morbidity patterns of the exposed group. In addition, an accident rate affected by peripheral neuropathy could be masked by undetected risk taking behavior bias.

(2) Corrective Measures

In an attempt to correct for the unique psychological factors that affect the choice of an aeronautical career, and to adjust for the effects of combat stress, transport aircrew members were matched with crewmembers of similar transport aircraft. However, the volunteer nature of the early RANCH HAND

operation suggests that this basic matching (as an attempt to control for the psychological effects of combat stress) is not totally ideal. The factors of volunteerism and risk-taking behavior must be considered from both the individual and group perspectives. The assessment of individual risk-taking behavior has been quantified by psychological instruments such as the Sensation Seeking Scale (SSS) of Zuckerman, et al. and the Life Experience Inventory (Torrance). The SSS has been demonstrated to have considerable validity in measuring a variety of phenomena including volunteerism and participation in risky activities and has been applied to naval aviation trainees (Waters). This study was unable to demonstrate an increased accident-related mortality in this group of individuals. These models will be adapted for use throughout all phases of the study.

D. Response Bias

(1) Problem

False positive response is anticipated as the primary bias operating in this study. Compensation issues arising from individual claims to the VA or from class action suits, heightened health concern generated by extensive publicity, disenchantment with military service, and the simple desire to please the interviewer may introduce positive responses that exceed the study's ability to correct or adjust. False negative response will also operate, and such bias is even more difficult to assess than the spurious response in a positive direction. Significant factors in this direction include: issues of patriotism and loyalty, personal conviction as to the propriety of the defoliation program and their participation in it, the strong virility orientation of the pilot/aircrew population (particularly with reference to questions of libido and fertility), personal inconvenience caused by study participation, errors of memory, and fear of the adverse effects on career goals that abnormal physical examination results could produce (a significant problem for active civilian and military pilots).

(2) Pending Retirement Bias

The military retirement system also creates a potential source of bias. A "pending-retirement phenomenon" occurs when personnel who are approaching the end of their careers exaggerate their symptoms so that they may become eligible for disability benefits.

(3) Corrective Measures

The primary correction technique for questionnaire response bias will be a carefully constructed and standardized physical examination. Multiple verification and bias indicator questions will be designed and included in the initial questionnaire. Memory verification will be conducted by cross-referencing responses

to medical and personnel records. Detailed statistical correlations between the questionnaire responses and the physical examination results will be conducted. All telephone interviews and physical examinations will be conducted on a "blind" basis to the maximum extent possible. Self-administered and group-administered questionnaires, which would allow for uncontrolled response changes, will not be conducted. Models of anticipated biases and their estimated impact on the study will be attempted prior to the final analysis of any phase in order to justify the analytic methods used. Conclusions drawn from this study will be predicated and coupled to a bias estimate.

E. Interview Bias

(1) Problem

Voice inflection, speed of interview, intonation and ethnicity are recognized factors which can affect positive or negative interview response. These factors will definitely operate in this study.

(2) Corrective Measures

An extensive interviewer training program will be conducted in order to limit the effects of interviewer bias. The Survey Research Center of the University of Illinois and the Center for Disease Control, Venereal Disease Training Branch, Atlanta, Georgia, will assist in this effort. The training will concentrate on techniques to elicit sensitive personal and medical information in an accurate manner, while minimizing discomfort to the subject and the interviewer. Quality assurance methodology and information verification techniques will also be included in the training. Interviewers will be randomly monitored by the supervisor in an unannounced manner. For particularly sensitive questions (e.g. illicit drug usage), randomized response techniques (coin flip method) will be used, recognizing that responses will be valid on a group basis only.

F. Political Implications

(1) Problem

The question of adverse health effects due to Herbicide Orange exposure in Vietnam has evoked many strong emotions. The actions of consumer groups, environmentalists, and other special interest groups have generated defensive responses on the part of some governmental agencies, and reactive decisions by others. Frequently, these responses have been based on unsubstantiated claims and/or scientific evidence of questionable validity. As a result of these governmental actions, the political impact on the planning of this study has been substantial. Suggestions to increase the scope

of the effort to include other "exposed" individuals or poorly defined ancillary groups continue to surface. However, monumental problems of group ascertainment, exposure validation, control group selection, and control of additional bias make the inclusion of such individuals undesirable from a sound scientific perspective. If such decisions are made without regard for their scientific impact, compromise of study validity is assured.

(2) Corrective Measures

The dilution of the scientific credibility of this effort by politically motivated decisions will be diplomatically resisted. While all suggested improvements will be considered, any alterations or corrections to the study protocol will be based on sound scientific assessments of the proposed changes. Such issues will be clearly presented to appropriate peer review agencies for comment. If studied, ancillary groups will be analyzed separately from the main study group and reported anecdotally.

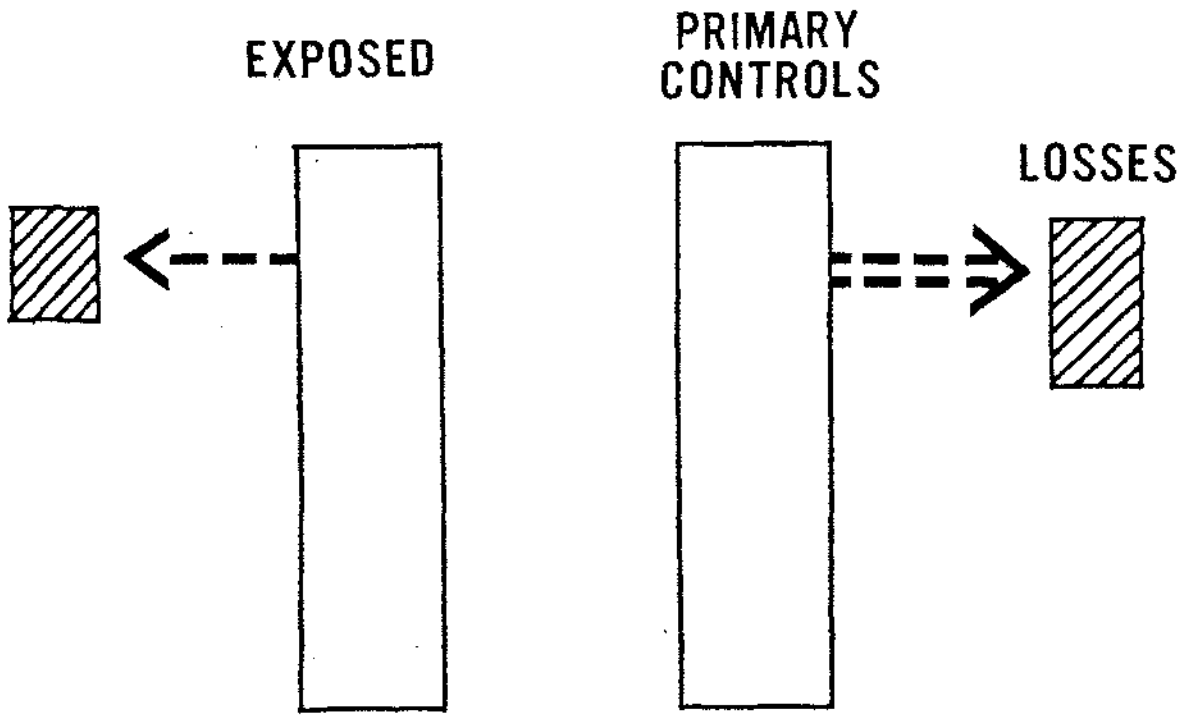
G. Loss to Study/Statistical and Bias Considerations

(1) Problem

Loss to study in the RANCH HAND group poses a major problem to the validity of the inferences that can be made from any subsequent comparisons between or within groups. The avenues of loss will conceivably arise from individual apathy (volunteer bias), lack of appropriate financial reimbursement for loss of salary, the presence or absence of illness (perception of health), and the lack of a desire for "treatment". Losses of matched controls during the questionnaire and physical examination phases of the study, though predictably greater than in the exposed group may be managed by replacement from the predetermined set of controls. The estimated participation of individuals is shown in Section XII, Figure A-3. It is estimated that the overall response rate of the exposed group will be 65% in the initial questionnaire and 40% in the physical examination phase of the study. These high non-compliance estimates are expected to occur despite great efforts to keep the questionnaire at an acceptable length, and to coordinate questionnaire administration and physical examination with the subject's personal schedule. Losses to study in either the exposed or control groups will obviously lead to decrements in statistical power, while losses in the control group could cause severe bias in the analytic phases of the study. Such losses would skew the distribution of controls, (Figures 3 and 4) and thus alter the characteristics of the population available for study. If differential losses in the control group occur (i.e., "well" controls dropout more frequently than "ill" controls), a "true" herbicide effect would be diluted (Figure 10). Conversely, if "ill" controls are differentially lost, a spurious effect would be attributed to herbicide exposure. To a lesser extent, losses in the exposed group could create similar effects; however, loss to study in the RANCH HAND population should be much less of a problem then in controls, due to their vested interest.

RATIONALE OF REPLACEMENT

DILUTIONAL BIAS



$P(L/W) > P(L/I)$ $P(L/W) >>> P(L/I)$

CONDITIONAL PROBABILITIES:

L = LOSS

W = WELL

I = ILL

(2) Corrective Measures

The USAF is committed to expending maximal effort to encourage participation. Loss to study problems in the study participants will be avoided as much as possible by detailed and exhaustive efforts to contact and followup each identified participant. NON-PARTICIPANTS WILL BE STRONGLY ENCOURAGED TO RECONSIDER THEIR INITIAL DECISIONS. Design considerations have been made to minimize loss to study in both the exposed and control populations. Although the USAF can not compensate study subjects for lost wages during the physical examination, transportation costs, per diem, and lodging costs will be reimbursed.

The replacement concept will help to counteract the decrement in statistical power, and offset the bias created by differential patterns of loss. The exposed group is already of maximum size and cannot be increased, but non-compliant controls can be replaced. This will maximize the degree of pairing between the two study groups. If a non-compliant control is replaced by a control with a similar perception of his own state of health, the alteration of the control group distribution is offset; (i.e., an "ill" control is replaced with an "ill" individual, and a "well" control with another "well" individual.) This concept of replacement, coupled with extensive efforts to encourage compliance will minimize losses to study and offset the adverse effects of those losses that do occur.

H. Statistical Power Limitations

(1) Problem

As discussed above, statistical power considerations are heavily dependent on loss to study rates. Since the design of the study is also limited by the small exposed population, statistical power for identifying the relative risk of an uncommon disease or symptom-complex (<1/100) is very low (<.50), (See Section VI F). This study will, to a greater extent, be able to detect increased risks only in common diseases or symptom-complexes (>1/100).

(2) Discussion

The "herald sign" of TCDD exposure, chloracne, is expected to have the greatest likelihood of achieving adequate statistical power in this study. Recent findings from Seveso, Italy, support the importance of chloracne as the primary marker symptom. The incidence of chloracne has been reported by Reggiani (personal communication) and Homberger, et al., to be 14.9 cases per 1000 residents in the region of highest contamination of Seveso (Zone A) and 6 to 12 cases per 1000 in the Seveso community as a whole. These rates vary by age group, with children being at highest risk. Only 1 to 5 cases per 1000 were seen in other regions of Northern Italy. (Milan, Como, and Lecco). The incidence of adolescent acne in all

of these populations varies between 21% and 30%. These incidence rates probably place chloracne at the lower limit of adequate statistical power within the constraints imposed on this study. In the Nitro, West Virginia studies, residuals of chloracne, as well as exacerbations of previously active disease, continue to be seen 10 years after the most recent exposures, and 30 years after the industrial accident. Thus, it is likely that any chloracne in the exposed population may be detected, despite the intervening years since RANCH HAND exposures.

In addition to chloracne, other recently reported human effects of TCDD exposure at Seveso, Italy, appear to fall within the capabilities of this study design (e.g., peripheral neuropathy, neuropsychiatric effects, and liver dysfunction).

In general, with respect to statistical power, continuous data even from relatively small samples fair much better than either categorical or dichotomous data. Consequently, a concerted effort will be made to obtain physical examination data in a scored and/or continuous manner.

I. Variability of Procedures

(1) Problem

The variation of physical examination findings from differences in technique and the random errors inherent in laboratory testing are items of concern, particularly if attributable health effects are subtle or of low magnitude. Nonstandardized procedures and techniques are major contributors to this variance.

(2) Corrective Measures

Variability in examination procedures will be minimized by the use of standardized procedures, examination protocols, similar equipment, and training. Most laboratory procedures will be conducted centrally at the USAFSAM, and quality control will be stressed at all times. Where at all possible, the same physician/technician teams will be used for each study phase.

J. Confounding Exposure Factors

(1) Problem

While virtually all of the media attention has been directed toward the 2,4,5-T-containing herbicide formulations, other herbicides were applied concurrently by the C-123 aircrews in Vietnam. Herbicide Blue (Cacodylic acid with 15.4% pentavalent arsenic) and Herbicide White (2,4-D and Picloram) were used throughout the 1962-1970 time period. Any long-term health effects from these

additional compounds may confound the results of the study. Peripheral neuritis, tremors, skin and lung cancer, loss of hair and nails, skin rashes, and gastric symptoms have been alleged after exposure to arsenical pesticides. The organophosphate insecticide, Malathion, was also sprayed by many of these same aircrewmembers when RANCH HAND duties permitted their temporary assignment to mosquito/malaria control units. Many of these individuals were involved in the aerial spray application of these and other pesticides both before, during, and after their Vietnam service. Long-term effects from these chemicals would confound the study results.

The small size of the RANCH HAND population will allow very little opportunity for analytic stratification for these confounding variables. Differing patterns of exposure to aircraft fuels in the study populations have been suggested as confounding factors. The C-130 aircraft were powered by turbo-prop engines which used jet fuel (JP-4), while the C-123 and C-7 aircraft were powered by standard reciprocating engines which used leaded aviation fuel (AV-GAS). After June 1968, many C-123s were modified by the addition of auxiliary jet engine boosters for added power on takeoffs and emergencies.

(2) Discussion and Corrective Measures

While the extent of confounding caused by exposure to these other pesticides is undetermined at this time, assessment of its magnitude must rely on responses of the subjects to that portion of the questionnaire dealing with other occupational exposures. For this reason, information concerning exposures to other herbicides/insecticides used in Vietnam will be collected. Whenever possible, stratification techniques will be used to adjust for these confounding variables during data analysis. Variations in fuel between C-130 and C-123 aircraft would be significant factors if individuals in the study were heavily and repetitively exposed. However, the normal duties of the study participants did not involve aircraft refueling or other fuel handling activities. Thus, fuel exposures can be minimized as a significant confounding factors.

IX. Reporting Procedures

Interim synoptic progress reports will be provided to the Surgeon General through Quarterly Management Reviews conducted each January, April, July and October. Key data analyses will be displayed, but inferences and conclusions will await full data analysis at the conclusion of each phase. A formal report for each of the three phases will be completed with forecasted submission dates of: Mortality Study, September 1981; Morbidity Study, December 1981; and Follow-up Study, July 1986. Findings and conclusions of each phase will be published in a journal of stature. Total study design, findings, and conclusions will be published in the USAFSAM Aeromedical Reviews or Technical Reports.

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XII. APPENDIX

TABLE A-1

SUMMARY OF 2,4-D, 2,4,5-T, AND TCDD ANIMAL STUDIES

	2,4-D	2,4,5-T	TCDD
LD_{50} RANGE (ACUTE)	100-1000 mg/kg	100-1000 mg/kg	1-1000 μg/kg
CHRONIC TOXIC DOSE	APPROACHES ACUTE LEVEL RAPID CLEARANCE	1/2 ACUTE LEVEL; VARIABLE CLEARANCE	MARKEDLY LOWER LEVEL BIOACCUMULATION
SIGNS OF ACUTE/ CHRONIC TOXICITY	ANOREXIA	ANOREXIA	WEIGHT LOSS
	WEIGHT LOSS	ATAXIA	INVOLUTION OF THYMUS
	MUSCULAR WEAKNESS	G.I. INJURY	ALOPECIA
	IRRITATED G.I. TRACT	LIVER CONGESTION	EPITHELIAL CHANGES
	MINOR LIVER INJURY	KIDNEY CONGESTION	LIVER LESIONS (VARIABLE)
	MINOR KIDNEY INJURY		HYPOTHYROIDISM
	MINOR LUNG CONGESTION		
EMBRYO TOXIC DOSE	APPROACHES TOXIC LEVEL	APPROACHES TOXIC LEVEL	MARKEDLY BELOW TOXIC MATERNAL LEVELS
TERATOGENICITY	QUESTIONABLE; WEAK AT BEST	*LOW INCIDENCE ONLY IN MICE (CLEFT PALATES DILITATED RENAL PELVIS)	SPECIES VARIATIONS: YES MICE NO RATS
CARCINOGENICITY	QUESTIONABLE; WEAK AT BEST	ONE STUDY: YES NUMEROUS STUDIES: NO	EPITHELIAL CHANGES IN PRIMATES: YES IN RATS

*TCDD CONTAMINATION OF 2,4,5-T HAS BEEN SHOWN TO BE A CONTRIBUTOR TO TERATOGENIC EFFECT IN MICE

TABLE A-2 "SYMPTOM COMPLEX" DERIVED FROM LITERATURE REVIEW OF CASE STUDIES

EXPOSED TO 2,4-D; 2,4,5-T AND/OR TCDD

2,4-D	2,4,5-T (+ TCDD)	TCDD
	CHLORACNE	CHLORACNE
	PORPHYRIA	PORPHYRIA
	HYPERPIGMENTATION	HYPERPIGMENTATION
ASTHENIA	ASTHENIA	ASTHENIA
PERIPHERAL NEUROPATHY	PERIPHERAL NEUROPATHY	PERIPHERAL NEUROPATHY
SWEATING/FEVER		
CARDIAC DISTURBANCE	CARDIAC DISTURBANCE	CARDIAC DISTURBANCE
RENAL DYSFUNCTION		RENAL DYSFUNCTION
LIVER DYSFUNCTION	LIVER DYSFUNCTION	LIVER DYSFUNCTION
GI DISTURBANCE	GI DISTURBANCE	GI DISTURBANCE
HEADACHE		
PNEUMONITIS		
CSF PROTEIN ALTERATIONS		HYPOTHYROIDISM
CONVULSIONS		HEARING/SMELL DISTURBANCES

TABLE A-3 DETAILED LISTING OF SYMPTOMS/SIGNS BY MAJOR CATEGORY

FROM LITERATURE REVIEW OF CASE STUDIES EXPOSED TO 2,4-D; 2,4,5-T AND/OR TCDD

NEURO-PSYCHIATRIC ABNORMALITIES

AESTHENIA	PERIPHERAL NEUROPATHY
ANXIETY	HYPOREFLEXIA
DEPRESSION	WEAKNESS
FATIGUE	PARESTHESIAS
APATHY	EXTREMITY NUMBNESS
LOSS OF DRIVE	MYALGIA
LIBIDO	GAIT DISTURBANCE
IMPOTENCY	"MILD" PARESIS
SLEEPLESSNESS	
EMOTIONAL INSTABILITY	
ANOREXIA	
DIZZINESS	
DECREASED LEARNING ABILITY	

TABLE A-3 (CONTINUED) DETAILED LISTING OF SYMPTOMS/SIGNS BY MAJOR CATEGORY FROM LITERATURE REVIEW OF CASE STUDIES EXPOSED TO 2,4-D,2,4,5-T AND/OR TCDD

DERMATOLOGIC DISEASE

CHLORACNE

PORPHYRIA CUTANEA TARDA

HYPERPIGMENTATION

HIRSUTISM (BODY)

ALOPECIA OF THE SCALP

OTHER DISORDERS

HEPATIC DYSFUNCTION	RENAL DYSFUNCTION
INCREASED CHOLESTEROL AND TRIGLYCERIDE	PROTEINURIA
	DECREASED OUTPUT
INCREASED LIVER FUNCTIONAL TESTS	TUBULAR DEGENERATION
	GLOMERULAR DEGENERATION
	RENAL GLUCOSURIA

GI DISTURBANCE	CARDIAC DISTURBANCE
NAUSEA	BRADYCARDIA
VOMITING	TACHYCARDIA
DIARRHEA	ATRIAL FIBRILLATION
GASTRITIS	
ABDOMINAL PAIN	

TABLE A-4

HERBICIDE RELATED CLAIMS SUBMITTED TO THE VETERANS ADMINISTRATION BY SYMPTOM CATEGORY AS OF 30 JUNE 1979 (N=417)

SYMPTOM CATEGORY	PERCENT
Dermatologic	49.9
Nervousness/Headache/Fatigue	23.0
Neuritis	14.6
Gastrointestinal/Genitourinary	13.4
Malignancy	11.0
Decreased Libido	5.8
Respiratory	5.3
Ear/Nose/Throat	5.0
Cardiovascular/Hypertension	4.1

NOTES:

625 total claims

190 claimed exposure without symptoms

18 claims paid for non-herbicide related conditions

1 claim paid for documented chloracne

TABLE 5

SCHEDULE AND MODE OF CONTACTS WITH

STUDY SUBJECTS

STUDY PHASE	CONTACT MADE	TIME
Morbidity Study	Introductory Letters	Dec 79 - Jul 80
Morbidity Study	Comprehensive Telephone Questionnaire	Jan 80 - Dec 80
	Baseline Physical Exam	Mar 80 - Mar 81
	Confirmation Face-to-Face Interview during Baseline PE	Mar 80 - Mar 81
Follow-up Study	Adaptive Phone Questionnaire	Jan 83 - Dec 83
	Adaptive Physical Examination	Mar 83 - Mar 84
Follow-up Study	Adaptive Phone Questionnaire	Jan 85 - Dec 85
	Adaptive Physical Examination	Mar 85 - Mar 86

TABLE A-6

MONTE CARLO SIMULATION

PROGRAM

```
10 DIM C(10,3)
20 DIM A(10,3)
30 P2=.25
40 D1=.45
50 M=0
60 N=0
70 FOR I=1 TO 10
80 FOR J=1 TO 3
90 A(I,J)=0
100 C(I,J)=0
110 NEXT J
120 NEXT I
130 M=M+1
140 PRINT M
150 IF M=1001 THEN 330
160 F=1
170 I=1
180 J=1
190 C(I,J)=RND(1)
200 X=P2+F*D1
210 IF C(I,J) > X THEN 270
220 I=I+1: F=1
230 IF I > 10 THEN 250
240 GOTO 190
250 N=N+1
260 GOTO 130
270 A(I,J)=A(I,J)+1
280 J=J+1
290 IF J>3 THEN 320
300 F=0
310 GOTO 190
320 GOTO 130
330 STOP
340 SELECT PRINT 215
350 FOR I=1 TO 10
360 PRINT A(I,1), A(I,2),
370 NEXT I
380 PRINT
390 PRINT "N(1)", N(1)
400 PRINT "N(2)", N(2)
410 PRINT "N(3)", N(3)
420 END
```

GLOSSARY

I = Control individual index

J = Examination number index

A(I,J) = Attendance array = number of times the ith control was used for the jth examination

C(I,J) = Testing variable array

N = number of times no control was available

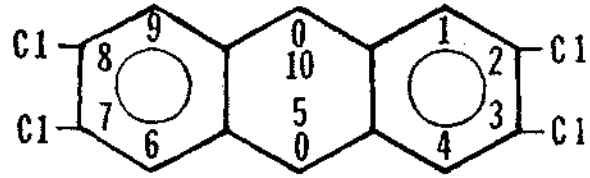
M = number of matches attempted

D1
P2 = preselected probabilities. P_1 = D1 + P2 and P_2 = P2

RND = Random
DIM = Dimension
F = Flag

Figure A-1

2, 3, 7, 8-TETRACHLORODIBENZO-p-DIOXIN (TCDD)

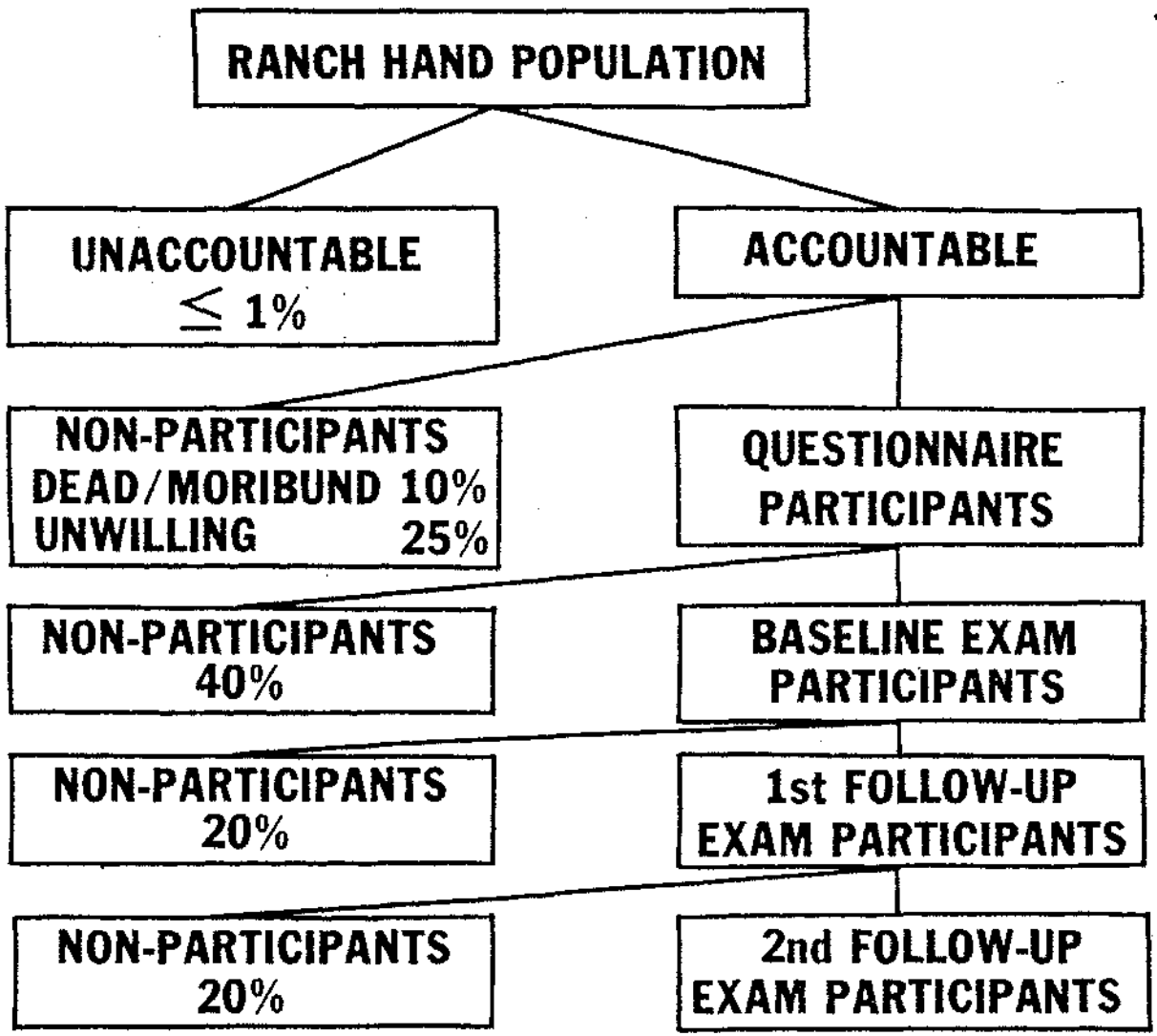


- MOLECULAR WEIGHT 321.8935
- MELTING POINT 303-305°C
- DECOMPOSITION POINT 980-1,000°C
- SOLUBILITY, GRAMS/LITER

ORTHO-DICHLOROBENZENE	1.40
CHLOROBENZENE	0.72
ORANGE HERBICIDE	0.58
BENZENE	0.57
CHLOROFORM	0.37
ACETONE	0.11
METHANOL	0.01
WATER	2×10^{-7}

Figure A-2

ESTIMATED IDENTIFICATION/PARTICIPATION OF THE RANCH HAND POPULATION



	RESPONSE ESTIMATE	ESTIMATED NUMBER OF PARTICIPANTS
RANCH HAND POPULATION	—	1200
ACCOUNTABLE	99%	1188
QUESTIONNAIRE PARTICIPANTS	65%	772
BASELINE EXAM PARTICIPANTS	60%	463
1st FOLLOW-UP EXAM PARTICIPANTS	80%	371
2nd FOLLOW-UP EXAM PARTICIPANTS	80%	297

XIII. QUESTIONNAIRE

The release of the actual questions within the questionnaire could possibly result in irreparable damage to the study from an avoidable source of responder bias. Consequently, the following is a summary of the general subjects to be covered as well as those specific areas that will receive particular emphasis.

Preliminary information will be obtained from a short mail-back questionnaire during the first contact with the study subjects. Personal identification data, RVN tour information, willingness to participate, acknowledgement of the Privacy Act Statement and an Informed Consent for release of records and medical evaluation/emergency treatment will be obtained at that time. This information will be verified by review of the individual's military personnel records. Addresses and telephone numbers will be verified and updated by use of several sources (i.e., Worldwide Locator Service, Social Security Service, Credit Bureaus, Rosters of RANCH HAND Association, Veterans Administration Benefits Records, the Internal Revenue Service, and other available sources). In addition, at this time a request will be made to have the wife present for the telephone interview in order to obtain an accurate obstetrical history.

The telephone questionnaire will, of necessity, be lengthy, but it will be convenient to the subject, and given in two interviews if necessary. It will verify personal identification data such as name, SSAN/AFSN, date of birth, address, telephone numbers, race, military status, effective date of status, location of military medical records and marital history information. RVN tour information will be rechecked and expanded to include data such as date of tour, tour end date, AFSC, organization of assignment, PCS and TDY status, combat missions, medal(s) awarded (i.e., Air Medal, Distinguished Flying Cross, Purple Heart), and whether or not the tour was a RANCH HAND affiliated tour.

Pre and Post-RVN exposure information, both occupational and avocational, to asbestos, radiation, herbicides, pesticides, will be elicited, including the frequency and time of the exposure. RVN exposure to these chemical and physical agents will also be collected.

Medical information obtained during this telephone interview will include of a statement of general health, smoking history, alcohol consumption history and long-term medication/drug use. In addition, questions dealing with infertility, birth defects of offspring, as well as the wife's obstetrical history (i.e., total conceptions, live births, miscarriages, stillbirths and premature pregnancies) will be asked. A family history specifying cancer, heart disease, liver disease and inherited disorders in both the subject's and wive's families will be collected.

A review of systems will be attempted, specifically emphasizing the neurologic, dermatologic, reproductive, and hepatic systems as well as specific symptoms associated with a neoplastic condition.

Specific questions will address the personality makeup of the individual in order to assess his potential risk-taking behavior (see Section VIII.C). Information verification and bias indicator questions will also be integrated into the questionnaire.

During this initial telephone questionnaire, those participants agreeing to undergo a physical examination will be instructed to insure that the medical examiners are "blind" as to whether or not they were former RANCH HAND personnel.

At the time of physical examination(s), the subject will receive a comprehensive face-to-face questionnaire which will expand and verify the information that was obtained in the telephone questionnaire and records review. An extensive review of systems will be covered at that time, including a more extensive occupational and avocational exposure history.

Just prior to the time of follow-up physical examinations (FY 1982 and FY 1984), a preliminary telephone contact will establish the subject's current health status and his willingness to continue participation in the study. An appointment for the followup examination will also be arranged. An adaptive telephone questionnaire will be given emphasizing those symptoms and systems that were found to be significantly associated with the exposed population on statistical analysis of the first year's results. If the subject expresses a desire to cease participation at this time, he will be encouraged to reconsider his decision and reasons for dropping out of the study will be sought.

XIV. Physical Examination Design

A. General Comments

This phase of Project RANCH HAND II is a cross sectional study of the subject's health at the time of examination. It is important that examiners remain unaware of the subject's status as a RANCH HAND participant or as a control subject. The physician examiner is tasked to examine and objectively record his findings. The examining physician is not, and cannot be expected to arrive at any definitive diagnosis as the full history and laboratory results are not available to him. The compilation and analysis of data will be performed by the study investigators at Brooks Air Force Base, Texas. They will notify the subject and the physician of his choice of the results of the examination.

These examinations will define the health status of the subjects at a point in time, and will establish the presence of physical findings, if any exist. After statistical review of the study groups, these findings may permit definition of a chronic effect due to exposure. An inaccurate examination may lead to falacious study results in two ways: a presumed syndrome may be defined which does not in fact exist, or a syndrome which in fact exists may not be defined with enough validity to warrant further actions.

The examining physician is responsible for recording a complete and detailed report of the physical examination. In this role, the examining physician is tasked to collect evidence of the presence or absence of physical signs of abnormality only. Formulation of impressions is not requested nor desired. If, during the examination, the physician discovers evidence of acute serious illness requiring immediate treatment, the normal emergency or urgent care procedures of the medical facility would apply. If during the examination, the examining physician finds evidence of present illness requiring further medical attention, he should so state to the subject and offer to forward or have forwarded pertinent information to the subject's physician. A clear record of any such advice and treatment should be recorded. The ultimate value of the RANCH HAND II Study will lie in complete accurate and, whenever possible, quantitative data permitting the most stringent and powerful statistical analysis. For that reason, the physical examination protocol requires exact measurements in many instances, and the use of defined meanings of semiquantitative indicators in other places.

C. Conduct of the Examination

SECTION	PHYSICAL EXAMINATION	SUBJECT NUMBER

1. GENERAL APPEARANCE

a. ___ Appearance/Stated Age ☐ Younger Than ☐ Older Than ☐ Same As
b. ☐ Well-nourished ☐ Obese
☐ Under-nourished ☐ Older Than
c. Appearance of illness or distress ☐ Yes ☐ No
d. Hair Distribution ☐ Normal ☐ Abnormal
SPECIFY:

2. HEIGHT cm	WEIGHT (Undressed) kg	SITTING BLOOD PRESSURE RIGHT ARM AT HEART LEVEL
		SYSTOLIC _____ DIASTOLIC _____

3. PULSE RATE _____ REGULAR: ☐ YES ☐ NO Describe any irregularities.
a. Irregular ☐
b. Irregularly irregular ☐
c. VPBs per minute _____

4. EYE GROUNDS ☐ NORMAL ☐ ABNORMAL Describe any vascular lesions, hemorrhages, exudates, papilledema.
☐ A-V nicking ☐ Hemorrhages ☐ Papilledema
☐ ↑ light reflex ☐ Exudates ☐ ↑ Cupping
☐ Arteriolar spasm ☐ Disk Pallor

5. ARCUS SENILIS ☐ PRESENT ☐ ABSENT 5a. Abnormal Ocular Pigmentation ☐ Yes ☐ No

6. ENT ☐ NORMAL ☐ ABNORMAL Describe any abnormality.
Tympanic membranes intact ☐ Yes ☐ No R ☐ L ☐
Nasal ulcerations ☐ No ☐ Yes

7. NECK (Especially thyroid gland) ☐ NORMAL ☐ ABNORMAL Describe any abnormality.
Thyroid gland palpable ☐ ☐ Parotid gland enlargement
Enlarged ☐ ☐ R ☐ L
Nodules ☐
Tenderness ☐

8. THORAX AND LUNGS ☐ NORMAL ☐ ABNORMAL Describe any abnormality, especially basilar rales.
☐ Asymmetrical expansion ☐ Wheezes Circumference at nipple level
☐ Hyperresonance ☐ Rales Expiration _____ cm
☐ Dullness Inspiration _____ cm

9. HEART ☐ NORMAL ☐ ABNORMAL Describe any enlargement, irregularity of rate, murmurs, or thrills.
Displacement of apical impulse ☐ No ☐ Yes Precordial thrust ☐ No ☐ Yes
Heart sounds normal ☐ Yes ☐ No ☐ S_1 ☐ S_2 ☐ S_3 ☐ S_4

(Continued in Item 18 on Reverse)

10. ABDOMEN ☐ NORMAL ☐ ABNORMAL Describe any abnormality with special attention to the spleen and liver.
☐ Heptomegaly ☐ Other mass - Specify: Record waist measurement on attached form.
_____ cm Liver Span
☐ Splenomegaly ☐ Tenderness
☐ Liver ☐ Spleen ☐ Other, specify:

11. EXTREMITIES ☐ NORMAL ☐ ABNORMAL Describe any edema or signs of vascular insufficiency.
☐ Absence, specify:
☐ Edema ☐ Clubbing of nails
☐ Pitting ☐ Non-pitting ☐ Varicosities
☐ Loss of hair on toes
☐ R ☐ L

SECTION — PHYSICAL EXAMINATION *(Continued)*

12. PERIPHERAL PULSES	NORMAL	DIMIN.	ABSENT	COMMENTS
RADIAL				
FEMORAL				
POPLITEAL				
DORSALIS PEDIS				
POSTERIOR TIBIAL				

13. SKIN ☐ NORMAL ☐ ABNORMAL — Indicate type and location of lesions on the attached anatomical figure

- ☐ Comedones
- ☐ Acneiform lesions
- ☐ Acneiform scars
- ☐ Depigmentation
- ☐ Inclusion cysts
- ☐ Cutis Rhomboidalis
- ☐ Hyperpigmentation
- ☐ Jaundice
- ☐ Spider angiomata
- ☐ Palmar erythema
- ☐ Petechiae
- ☐ Ecchymoses
- ☐ Positive lesions with UV light

Full-Face and Bilateral profile photos taken ☐ Yes ☐ No

14. MUSCULOSKELETAL ☐ NORMAL ☐ ABNORMAL

- ☐ Muscle - Specify:
  - ☐ Weakness
  - ☐ Tenderness
  - ☐ Abnormal Consistency
  - ☐ Atrophy
- ☐ Spine
  - ☐ Scoliosis
  - ☐ Kyphosis
  - ☐ Tenderness, Level ____
  - ☐ Decreased range of motion
  - ☐ Pelvic tilt
  - ☐ Straight Leg Raising: Right/Left

15. GENITOURINARY - RECTAL - HERNIA ☐ NORMAL ☐ ABNORMAL

- ☐ Inguinal hernia ☐ R ☐ L
- ☐ Testes

	Absent	Enlarged	Atrophic
☐ R	☐	☐	☐
☐ L	☐	☐	☐

- ☐ Varicocele
- ☐ Epididymis
- ☐ Scotal mass - Specify ____ cm dia
- ☐ Hemorrhoids
- ☐ Prostatic Enlargement
- ☐ Rectal mass

Stool Hemacult: ☐ Positive ☐ Negative

16. LYMPH NODES - CHECK ALL AREAS. ☐ NORMAL ☐ ABNORMAL - SPECIFY CERVICAL, OCCIPITAL, SUPRACLAVICULAR, AXILLARY, EPITRACHLEAR, INGUINAL, FEMORAL

☐ Enlarged ____ ☐ Tender ____ ☐ Hard ____ ☐ Fixed ____ ☐ Confluent

17. NERVOUS SYSTEM - SEE ATTACHED FORMS

18. HEART AND OTHER OBSERVATIONS

(Continued from Item 9)

Murmur ☐ No ☐ Yes Area ☐ Ao ☐ Pu ☐ Apex ☐ S4

Sys ☐ ☐ ☐ ☐

Dia ☐ ☐ ☐ ☐

DATE OF EXAMINATION (MONTH / DAY / YEAR)	TYPED OR PRINTED NAME OF EXAMINING PHYSICIAN	RETURN FORM TO:
EXAMINING FACILITY	SIGNATURE	USAFSAM/ ES BROOKS AFB TX 78235

CLINICAL RECORD NEUROLOGICAL EXAMINATION

HEAD AND NECK - Normal to Palpations/Inspection ☐Y ☐N Specify Scar ☐
Asymmetry ☐ Depression ☐
Carotid Bruit ☐No ☐R ☐L
Neck Range of Motion ☐ Normal or Decreased to ☐ Left ☐ Right
☐ Forward ☐ Backward

TRUNK

MOTOR SYSTEM - Handedness Right ☐ Left ☐

Gait ☐ Normal or ☐ Broad Based ☐ Ataxic ☐Small Stepped ☐Other-Specify

Associated Movements ☐Arm Swing ☐Normal or Abnormal ☐R ☐L

Muscle Status (strength, tone, volume, tenderness, fibrillations)
Bulk ☐ Normal ☐Abnormal

Tone Upper Extremities ☐Normal or ☐Increased ☐Decreased
☐Right ☐Left

Lower Extremities ☐Normal or ☐Increased ☐Decreased
☐Right ☐Left

Strength - Distal wrist extensors ☐Normal ☐Decreased
Ankle/Toe Dors/Flexors ☐Normal ☐Decreased ☐R ☐L
Proximal Deltoids ☐Normal ☐Decreased ☐R ☐L
Hip Flexors ☐Normal ☐Decreased ☐R ☐L

Abnormal Movements (tremors, tics, choreas, etc.) Fasiculations ☐No ☐Yes (1-4+)
Tenderness ☐No ☐Yes (1-4+)

Tremor ☐No ☐Yes - Specify
Upper Extremity ☐R ☐L ☐Resting ☐Essential ☐Intention
Lower Extremity ☐R ☐L ☐Other

Coordination (a) Equilibratory - Eyes Open
Eyes Closed - Romberg ☐Positive (Abnormal) ☐Negative (Normal)

Right Foot Left Foot

(b) Nonequilibratory (F to N; F to F; H to K) Finger-to-nose-to-finger
☐Normal ☐Abnormal ☐Right ☐Left ☐Both
Heel-Knee-Shin ☐Normal ☐Abnormal ☐Right ☐Left ☐Both
(c) Succession Movements (including check, rebound, posture-holding)
If indicated, check ☐Normal ☐Abnormal ☐R ☐R

Rapidly alternative movements ☐Normal ☐Abnormal ☐R ☐L ☐Both

Skilled Acts (a) Praxis
(b) Handwriting. If indicated, ☐Normal ☐Abnormal

(c) Speech (articulation, aphasia, agnosia) Grossly ☐Normal
☐Abnormal - Specify Dysarthria ☐

Aphasia ☐

Reflexes (0-absent; 1-sluggish; 2-active; 3-very active; 4-transient clonus; 5-sustained clonus)

Deep	R	L	Deep	R	L	Other	R	L	Abnormal	R	L
									Babinski		
Biceps			Patellar								
Triceps											
			Achilles								
Remarks											

MENINGEAL IRRITATION Spurling Maneuver of Neck ☐Normal ☐Abnormal
☐R ☐L ☐Both
Straight Leg Raising ☐Normal ☐Abnormal ☐R ☐L ☐Both

NERVE STATUS (tenderness, tumors, etc.)

SENSORY SYSTEM (tactile, pain, vibration, position. If positive sensory signs are present, summarize below and indicate details on Anatomical Figure, Std. Form 531)

Light Touch ☐Normal ☐Abnormal
Pin Prick ☐Normal ☐Abnormal (Map on Anatomical Figure)
Vibration (at ankle, 128 hz tuning fork): ☐Normal ☐Abnormal ☐R ☐L ☐Both
Position (Great toe): ☐Normal ☐Abnormal ☐R ☐L ☐Both

CRANIAL NERVES

I R Smell ☐Present ☐Absent
L Smell ☐Present ☐Absent

II Fundus R Normal ☐ Abnormal ☐ Disk Pallor/atrophy
☐Exudate ☐Papilledema ☐Hemorrhage

Fundus L Normal ☐ Abnormal ☐ Disk pallor/atrophy
☐Exudate ☐ Papilledema ☐Hemorrhage

Fields (to confrontation)
Right ☐Normal ☐Abnormal Left ☐Normal ☐Abnormal

III Normal ☐ ☐Abnormal - Specify
IV
VI Pupils-Size (mm) Equal ☐ Unequal ☐ Difference mm____
Shape, position Round ☐ Other ☐ ☐R ☐L
Light, Reaction Normal ☐ Abnormal ☐ ☐R ☐L
Position of Eyeballs

Movements R L

Nystagmus Rotary ☐ Horizontal ☐ Vertical ☐
(Draw position)

XI

Ptosis R☐ L☐

V Motor R Clench Jaw - Symmetric ☐ Deviated ☐ R☐ L☐
L

Sensory R Normal ☐ Abnormal ☐ V_1☐ V_2☐ V_3☐
L Normal ☐ Abnormal ☐ V_1☐ V_2☐ V_3☐

Corneal Reflex R L

VII Motor R Normal smile ☐Yes ☐No Palpebral Fissure ☐Yes ☐No
L Normal smile ☐Yes ☐No Palpebral Fissure ☐Yes ☐No

IX Palate and Uvula
X Movement Normal ☐ Deviation to ☐R ☐L

Palatal Reflex R ☐Normal ☐Abnormal
L ☐Normal ☐Abnormal

XII Tongue-Protruded-Central ☐ R ☐ L ☐
Atrophy ☐No ☐Yes

MENTAL STATUS (alert, clear, cooperative, etc.) Gross abnormalities: ☐No ☐Yes - Specify

SUMMARY OF POSITIVE FINDINGS

Subjective Objective

Diagnostic Impression

Date Signature

C. Special Procedures

(1) Nerve Conduction Velocities (NCV)

(a) These studies have been determined to be an important parameter in long-term follow-up studies of persons thought to have been exposed to Herbicide Orange components.

(b) The Nerve Conduction Velocities should be performed by a physician or by a specialty qualified technician under the supervision of a physician trained in neurophysiological methods.

(c) Specific NCVs

(1) Ulnar Nerve (one side only)

(a) motor (above elbow, below elbow)

(b) values recorded

(i) distal latency

(ii) NCV

(2) Peroneal Nerve (one side only)

(a) motor

(b) values recorded

(i) distal latency

(ii) NCV

(3) Sural Nerve (one side only)

(a) sensory: orthodromic

(b) values recorded: NCV

(d) Methods

(1) Standardized, published methods will be used (e.g., Smorto, Marcio P., and John V. Besmajian; Electrodiagnosis; Harper and Row; NY, 1977).

(2) Psychological Test Battery

(a) General

(1) This battery yields objective numerical data, and is well-standardized and clinically validated. The individual tests were chosen to insure an adequate analysis of the major alleged manifestations of Herbicide Orange toxicity. Each test either validates the other tests or is considered to be a "definitive" test for analysis of a suspected psycho-neuropathic effect under study.

(2) Compared to the general civilian population, characteristic response tendencies are observed on the MMPI and Cornell Index among active duty aircrewmen being evaluated in an aeromedical setting. It is also important to consider the effect that pending retirement has exerted on the reporting of medical history and symptomatology. This may also alter responses to psychological testing.

(b) Specific Tests

(1) Wechsler Adult Intelligence Scale (WAIS): Individually-administered collection of verbal and nonverbal intellectual measures; also useful for clinical inferences when combined with the neuropsychological battery below.

(2) Reading subtest of the Wide Range Achievement Test (WRAT): Individually-administered measure of word recognition ability. Important so as to rule-out reading inefficiency should response to the personality instruments below be of questionable validity (e.g., high F Scale on MMPI).

(3) Halstead-Reitan Neuropsychological Test Battery: Individually-administered collection of brain behavior relationship measures for establishing the functional integrity of the cerebral hemispheres. The battery must include the following subtests: Category, Tactual performance, Speech-Sounds, Seashore Rhythm, Finger Tapping, Trail Making, and Grip Strengths. The Aphasia Screening and Sensory-Perceptual Exams are considered optional in view of their redundancy with the clinical neurologic exam included in this project. Individualized test debriefing is conducted to clarify test performances in the WAIS and Neuropsychological Battery.

(4) Three subtests of the Wechsler Memory Scale I (WMS I): Individually-administered measures of immediate and delayed recall of verbal and visual materials. The Local Memory, Associate Learning and Visual Reproduction subtests are to be administered in the standard, immediate-recall fashion initially. After 30 minutes has elapsed, the examinee is asked, without prior alerting, to recall as much as he can about the Logical Memory and

Visual Reproduction subtest stimuli. Standard scoring is used for both test-retest administrations.

(5) Cornell Index (CI): Self-administered and standardized neuropsychiatric symptom and complaint inventory, including items involving asthenia, depression, anxiety, fatigue, and GI symptoms in lay language. Endorsement of items are to be explored and clarified in test-debriefing.

(6) Minnesota Multiphasic Personality Inventory (MMPI): Self-administered clinical psychiatric screening instrument; also capable of estimating response biases (e.g., "fake good," or "fake bad"). The shortened version of Form R (i.e., items 1 to 399) may be substituted for the 566-item Long Form. Standard scoring and Minnesota norms are to be used, with the possible exception of active duty examinees where USAFSAM aircrew norms may be applied. Clarification of profiles showing response biases, questionable validity, and/or unusual item endorsements will be conducted in individual test debriefing.

(3) 12-Lead Electrocardiogram

(a) A standard 12-lead scalar electrogram is required. If an arrhythmia is observed, a one minute rhythm strip is requested, in addition.

(b) Interpretation:

The electrocardiograms will be interpreted by physicians in the USAF Central ECG Library and compared to previous individual ECG records in the case of rated (pilot or navigator) subjects.

(c) Disposition (USAF Central ECG Library):

(1) Pilots and Navigators - The original tracings will be microfisched and a permanent record established for each individual.

(2) Enlisted Subjects - The original tracings will be microfisched and a permanent record established for each individual.

(4) Radiographic Examination

(a) A standard 14x17 in., standing, teleroentgenogram in the PA position using small nipple markers.

(5) Laboratory

(a) Specific Tests

(1) Performed at the Local Examining Facility

(a) Hematocrit

(b) Hemoglobin

(c) RBC Indices

(d) While Blood Cell Count

(e) Platelet Count

(f) Erythrocyte Sedimentation Rate

(g) Urinalysis

(h) Semen Analysis (Number, Motility, % Abnormal, Volume)

(2) Performed by USAFSAM Clinical Pathology Laboratory

(a) Blood Urea Nitrogen

(b) Fasting Plasma Glucose

(c) Creatinine

(d) 2-hour Post Prandial Plasma Glucose

(e) Differential Cortisol

(f) Cholesterol & HDL cholesterol

(g) Triglycerides

(h) SGOT

(i) SGPT

(j) GGTP

(k) Alkaline Phosphatase

(l) LDH

(m) Serum Protein Electrophoresis

(n) CPK

(o) VDRL

(3) Performed by USAFSAM Epidemiology Division Reference Laboratory

(a) LH

(b) FSH

(c) Testosterone

(d) Thyroid Profile (RIA)

(e) Delta-aminolevulinic Acid

(f) Urine Porphyrins

(4) Performed at USAFSAM if liver function studies are abnormal

(a) Anti-nuclear Antibody

(b) Hepatitis Antigens/Antibodies (A and B)

(5) Performed if medical history indicates an increase in infectious diseases:

(a) Immuno electrophoresis

(b) Monilia Skin Test

(c) Quantitative Immunoglobulin Determinations

(b) Rationale for Laboratory Procedures

(1) Studies on the toxicity of TCDD in animals have shown that the following organ systems are damaged:

(a) Liver: Hepatic necrosis, liver enzyme changes, hypoproteinemia, hypercholesterolemia, hypertriglyceridemia.

(b) Reticuloendothelial System: Thymic atrophy, altered cellular immunity, decreased lymphocyte counts.

(c) Hemopoietic System: Anemia, thrombocytopenia, leukopenia, pancytopenia.

(d) Endocrine System: Hemorrhage and atrophy of adrenal cortex, hypothyroidism.

(e) Renal: Increase in blood urea nitrogen.

(f) In addition, statistically significant increases in hepatocellular carcinomas (liver) and squamocellular carcinomas of the lung were found.

(2) Studies on the toxic effects of TCDD in man have shown that the following organ systems are damaged:

(a) Skin: Chloracne, hirsutism.

(b) Liver: Porphyria cutanea tarda. Increased levels of transaminase and of GGTP. Enlarged, tender liver, hyperlipidemia.

(c) Renal: Hemorrhagic cystitis, focal Pyelonephritis.

(d) Neuromuscular System: Asthenia, i.e., headache, apathy, fatigue, anorexia, weitht loss, sleep disturbances, decreased learning ability, decreased memory, dyspepsia, sweating, muscle pain, joint pain and sexual dysfunction.

(e) Endocrine System: Hypothyroidism.

(3) Based upon the reports of toxic effects in animal and human exposures, the following organ panels are recommended:

(a) Hemopoietic

(b) Reticuloendothelial

(c) Renal

(d) Endocrine

(e) Neuromuscular

(4) Hemopoietic screening should include:

(a) Hematocrit

(b) Hemoglobin

(c) RBC indices

(d) Erythrocyte sedimentation rate

(e) Platelet count

(5) Reticuloendothelial system:

(a) White blood cell count

(b) Differential

(c) Serum protein electrophoresis

(d) Selective use of skin testing, immunoelectrophoresis, and quantitative immunoglobulin determinations

(6) Hepatic screen:

(a) SGOT

(b) SGPT

(c) GGTP

(d) Alkaline phosphatase

(e) LDH

(f) Cholesterol

(g) HDL cholesterol

(h) Triglyceride

(i) Urine prophyrins

(j) Urine porphobilinogen

(7) Renal screen:

(a) Urinalysis

(b) BUN

(c) Creatinine

(8) Endocrine screen

(a) Differential cortisol (0730 and 0930 hours)

(b) Thyroid profile (RIA)

(c) Fasting plasma glucose

(9) Neuromuscular system: CPK

(10) The following tests should be peformed only as follow-up for abnormalities in the liver panel:

(a) HAVAB (IgG and IgM)

(b) HB_sA_g

(c) Anti HB_cA_g

(d) Anti HB_sA_g

(e) ANA

(11) Elucidation of sympoms of asthenia:

(a) Testosterone

(b) LH

(c) FSH