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USAMRMC ltr, 1 Jun 2001.

AD_____

GRANT NUMBER DAMD17-94-J-4245

TITLE: Multigenerational Breast Cancer Risk Factors in African-American Women

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REPORT DATE: October 1997

TYPE OF REPORT: Annual

PREPARED FOR: Commander
U.S. Army Medical Research and Materiel Command
Fort Detrick, Maryland 21702-5012

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. AGENCY USE ONLY *(Leave blank)*	2. REPORT DATE	3. REPORT TYPE AND DATES COVERED
	October 1997	Annual (15 Sep 96 - 14 Sep 97)

4. TITLE AND SUBTITLE
Multigenerational Breast Cancer Risk Factors in African-American Women

5. FUNDING NUMBERS
DAMD17-94-J-4245

6. AUTHOR(S)
Selina A. Smith, Ph.D.

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)
University of Miami School of Medicine
Miami, Florida 33101

8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING/MONITORING AGENCY NAME(S) AND ADDRESS(ES)
Commander
U.S. Army Medical Research and Materiel Command
Fort Detrick, Frederick, Maryland 21702-5012

10. SPONSORING/MONITORING AGENCY REPORT NUMBER

11. SUPPLEMENTARY NOTES

12a. DISTRIBUTION / AVAILABILITY STATEMENT
Distribution authorized to U.S. Government agencies only (proprietary information, Oct 97). Other requests for this document shall be referred to U.S. Army Medical Research and Materiel Command, 504 Scott Street, Fort Detrick, Maryland. 21702-5012.

12b. DISTRIBUTION CODE

13. ABSTRACT *(Maximum 200*

An increased rate of breast cancer has been observed in African-American women under 50 years of age. Few epidemiologic studies of breast cancer focus explicitly on African-American women, and it is not established whether standard risk factors apply. This study seeks to determine relationships between risk factors and genetics in breast cancer etiology among pre-menopausal African-American women. Breast cancer cases and their primary female relatives (PFRs), were administered several questionnaires (dietary, psychosocial, reproductive, genetic and lifestyles) related to disease risk. Cases were matched by ethnicity and age to two cancer-free women participating in a screening mammography program . DNA samples were extracted from the blood of all cases and PFRs. Data on selected reproductive risk factors indicate that cases and controls are similar except with a difference in age at first birth. The diets of cases and their PFRs were similar. More cases than controls reported taking Vitamins A, C & E. Ninety four (94) percent of cases are *pleased with emotional support provided by family/friends.* Further work is in progress to identify BRCA1 gene mutations in women with affected relatives with breast/ovarian cancer.

14. SUBJECT TERMS	15. NUMBER OF PAGES
Breast Cancer; African American, Lifestyles, Psychosocial	24
	16. PRICE CODE

17. SECURITY CLASSIFICATION OF REPORT	18. SECURITY CLASSIFICATION OF THIS PAGE	19. SECURITY CLASSIFICATION OF ABSTRACT	20. LIMITATION OF ABSTRACT
Unclassified	Unclassified	Unclassified	Limited

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Silvia A. Smith 10/21/97

PI - Signature Date

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INTRODUCTION

Breast cancer is the most commonly diagnosed nondermatologic cancer and is the second leading cause of cancer-related deaths among women in the United States (1-3). The overall incidence rate increased from 82.5 to 110.6 during 1973-1992 (4), with varying race-specific rates. For white women, the age-adjusted rate increased 34% (from 84.3 to 113.1) and for black women, increased 47% (from 68.7 to 101.1) during that same time period (2).

Reasons cited for this increased incidence among African Americans have been numerous. Increases in screening mammography rates, earlier detection of cancer and increased access to care for a group disproportionately represented at the low socioeconomic status level are among reasons cited. Although these explanations may represent partial answers for the growing trend of breast cancer diagnosis for black women, none account fully for ethnic differences in breast cancer incidence.

Epidemiologic studies have shown risk factors such as age; socio-economic class; race/ethnicity; lifestyle; and reproductive factors increase a woman's chance of developing breast cancer. There have been few epidemiologic studies of breast cancer focusing explicitly on African-American women, and it is not established whether the standard risk factors apply. Evidence relative to younger versus older African American women is also scarce. In work by Robert Mayberry (5), risk of breast cancer among black women younger than 40 years of age was nearly three times greater for those who used oral contraceptives for more than 10 years relative to never-users (odds ratio, 2.8; 95% confidence interval, 1.2 to 6.8) and more than four times greater for severely obese women (body mass index $\geq$ 32.30 kg/m^2) relative to women whose relative weights were less than 24.90 kg/m^2. The report also indicated similarities among younger and older black women with regards to additional breast cancer risk factors (i.e., a surgical menopause, age at first full term pregnancy and multiple births).

Women with a family history of breast cancer are also at increased risk for developing breast cancer. Studies indicate women with a familial pattern of this disease are thought to have a lifetime risk of breast cancer as high as 50% (6). This risk varies from woman to woman, depending on the presence of other risk factors. Research suggests the risk is two to three times higher in women who have first-degree relatives with breast cancer (7). There is also evidence that, for women with first-degree relatives with bilateral, premenopausal breast cancer, the risk is even higher (7,8).

In 1990, linkage studies localized an inherited susceptibility gene for breast cancer aggregates in certain families to chromosome 17q (9). Generally, women who inherit the BRCA1 gene are more likely to develop cancer than those who do not.

Female carriers of BRCA1 mutations are estimated to have an 85% lifetime risk of developing breast cancer, with more than 50% of the breast cancers occurring before the age of 50 years (10). It is, however, very likely that other genes may be responsible for inherited breast cancer. Since the cloning of the BRCA1 gene (10,11), it is possible to detect the presence of mutations in this gene in about 45% of the families with a clearly inherited pattern of this disease.

The purpose of this research is to determine the relationship between risk factor and genetics in the etiology of breast cancer. The objectives include: 1) analyze multigenerational reproductive and lifestyle risk factors; 2) dietary assessment; 3) psychological assessment; and 4) complete pedigree data and storing DNA samples for the study of mutations on the BRCA1 gene.

Several questionnaires, including a Breast Cancer Risk Appraisal (BCRA), Food Frequency Questionnaires (HHHQ), and Psychosocial Questionnaires were administered to study risk factors for breast cancer in premenopausal African American women by identifying genetic and environmental influences in the etiology of breast cancer in cases and their family groups.

BODY

For Study Year 3, the project is off target in terms of the recruitment timeline with only 107 breast cancer cases enrolled. An explanation of the low case accrual rate and our plans to meet our final target of 200 cases will follow below. Currently, there are 436 women, including breast cancer cases and their primary female relatives (PFR) (mother, sister(s), and/or female offspring) and two disease-free controls, enrolled. Each family group member has completed the BCRA, HHHQ, 24 Hour Dietary Recall and Psychosocial Questionnaires and had their blood drawn for DNA extraction. Family history forms were administered to capture information related to illnesses and causes of deaths within family groups.

Cases were identified from several sources including: 1) the Jackson Memorial Medical Center Tumor Registry (JMMC/TR); 2) Cedars Hospital Tumor Registry; and 3) South Miami Hospital Tumor Registry, to enlist the required cases. Permission was obtained from the Florida Department of Health and Rehabilitative Services (HRS) to obtain the names and addresses of African American women age 50 years or less diagnosed with breast cancer in the past five years entered into the Florida Cancer Data System (FCDS). This FCDS identified a total of 3,174 age-eligible breast cancer cases in the State, with a large percentage of them residing in South Florida. Letters were mailed to eligible women in the South Florida area.

Because recruitment sources varied, participants were contacted in-person, by telephone or by letter. Participants were given the option of completing the assessment interview by phone or by home visit. Informed consent and blood for

DNA testing were obtained prior to the arranged interview, which lasted approximately 90 minutes. Allowing several options for interview location increased participant enrollment.

Controls were randomly selected from the total number of participants undergoing screening mammography in the Early Detection Program (EDP) mobile screening project. A control was defined as the first woman enrolling in the EDP immediately following the identification of the case matched by age (within 10 years) and ethnicity. Following study protocol, two cancer-free women were matched to one case.

A Risk Appraisal Project or RAP (an acronym for the Multigenerational Breast Cancer Risk Factors in African American women) database was developed in Study Year 1 to ensure data are continuously accumulated to meet study objectives. Data were entered on a routine basis, following editing and checks for outliers by the data manager. Data analysis for cases and controls was completed by linking RAP and EDP databases.

Objective 1: Analyze multigenerational reproductive and lifestyle risk factors in African American family groups. Aim 1: Conduct a breast cancer risk factor appraisal. A breast cancer risk appraisal (BCRA), designed to capture all known and suspected risk factors for breast cancer, developed and pilot tested in Study Year 1, continues to be administered to all participants. Risk factors such as diet, reproductive factors, activity level, body build, alcohol use and fat distribution for cases, primary female relatives and controls, were analyzed. In our last annual report, we presented preliminary data on these factors among family groups. These findings were presented at the 124th Annual Meeting of the American Public Health Association, November 17-21, 1996 New York, NY. Multigenerational reproductive and lifestyle risk factors will continue to be examined in Study Year 4. Table 1 represents selected demographics and health characteristics analyses completed to-date on the total project sample. Approximately 76% of cases received a high school diploma compared to 63.6% of mothers, 80.6% of siblings and 87.7% of offspring. Cases were more likely to have a yearly income less than $10,000 (47.3%) compared to siblings (28.6%), mothers (45.5%) and offspring (55.4%). In addition, analyses showed more cases (83.8%) rarely/never exercise.

Table 1. Sociodemographic and Health Characteristics of Breast Cancer Cases, and their Siblings, Mothers, & Offspring*

	Cases	Siblings	Mothers	Offspring
Sociodemographic				
Age (years) (SD)	44 (7.83)	45 (11.51)	62 (9.13)	27 (8.87)
Education Level				
< high school	24.2%	19.4%	36.4%	12.3%
GED/high school grad/some college	62.6%	38.9%	54.5%	81.5%
college grad	13.1%	41.7%	9.1%	6.2%
Income				
< $10,000	47.3%	28.6%	45.5%	55.4%
$10,000-19,999	26.9%	28.6%	27.3%	32.1%
$20,000-29,999	11.8%	20.1%	18.2%	5.4%
$30,000+	14.0%	22.9%	9.1%	7.1%
Exercise				
> 1/wk.	13.13%	25%	-0-	24.6%
About 1/wk.	1.01%	8.3%	-0-	1.54%
Few times/month	2.02%	2.78%	18.2%	3.07%
Rarely/never	83.8%	63.8%	81.8%	70.8%

*Does not include missing responses.

Of the captured risk factors, those associated with reproduction have the strongest scientific support in breast cancer etiology. However, the salient absence of African American women in previous large scale case-control epidemiological studies examining breast cancer risk factors has left a void in our understanding of which factors are applicable to black women and which factors or not.

Estrogen is of special interest in breast cancer etiology. Its role in breast cell multiplication, the total number of menstrual cycles and possible genetic alterations in cells during this replication is thought to be important in disease development. The typical woman is thought to have approximately 300-400 menstrual cycles in her lifetime, exposing her breasts to several estrogen-progesterone cycles (12). Factors affecting this number, as well as cycle duration, are implicated in breast cancer. Repeated cellular multiplication is believed to increase the likelihood of genetic accidents. Women with fewer menstrual periods, i.e., those beginning their menstrual cycles later, having their first child young, repeated pregnancies, longest breast-feeding durations and earlier menopause, are at lowest breast cancer risk.

To address the issue of estrogen and breast cancer risk, we examined reproductive factors among breast cancer cases and their age-ethnicity matched controls. Using cited values as the goal standard and comparing breast cancer cases to controls, Table 2 summarizes results of analyzed reproductive factors.

Table 2. Probable Reproductive Risk Factors Among African American Breast Cancer Cases and Age-Ethnicity Matched Controls

Reproductive Factor	Established Risk Levels*	Breast Cancer Cases** (N = 94)	Controls** (N = 182)
Age @ Menarche	≥12 Years	13 Years	13 Years
Age @ First Full Pregnancy	>30 Years	21.4 Years	20 Years
Number of Full Term Births	+	4	5
Breast Feeding Duration	≥9 months	2	3
Age @ Menopause	≤55 Years	41	42
Exogenous Hormones	_	17	9
Oral Contraceptives	_	76	52

*Values associated with reduced breast cancer risk.
**Does not include missing responses

Age at menarche: women starting their menstrual cycles later in life (i.e., $\geq$ 12 years of age) are thought to be at lower risk for breast cancer (13). Cases and controls reported starting their menstrual cycle at age 13. *Age at full pregnancy*: having the first child at or before age 30 years lowers breast cancer risk (14). Cases reported having their first birth at an average age of 21.4 years, compared to controls, who reported first birth at age 20 ($p=0.04$). *Number of full term births:* Parity is thought to modify disease risk (15). Controls reported having an average of 5 full term births compared to cases reporting an average of 4 full term births. *Breast feeding:* Because lactation interrupts the estrogenic cycle (i.e., nursing women menstruate less), breast-feeding duration is important in examining breast cancer risk. Breast-feeding nine months or more is associated with lower risk (16). Both cases and controls reported breast-feeding their children less than 9 months, but controls breast-fed their children slightly longer than cases. Within families, sisters most often reported having ever breast-fed ($p=0.02$). *Age at menopause:* women undergoing menopause later as opposed to earlier (i.e., age 55 years or less) are at greater breast cancer risk (17). *Exogenous hormone use:* current use of birth control pills and/or estrogen replacement therapy, because of their influence on estrogen in the body, may also elevate risk (18).

Aim 2: Evaluate the role of food choice and food practices and breast cancer. The Health, Habits, History Questionnaire (HHHQ) has been administered to all participants except the community controls. The HHHQ estimates total calories, dietary fat intake including total Vitamin A, beta carotene and retinol from both dietary sources and vitamin supplementation, vitamin C, Vitamin E, fruit, vegetables and meat consumption, and fiber intake for the year prior to the interview. This instrument also captures food practices, i.e., frying versus baking meats, which may be related to breast cancer risk. Responses to each HHHQ are recorded on scannable forms which are forwarded in bulk to a commercial analyzer, Survey and Ballot Systems, Inc., Minnesota. Results are sent back to the project office on a diskette, which is then down loaded to the RAP database.

In addition to the HHHQ, a 24 Hour Dietary Recall (24 HDR) was also administered. The 24 HDR asks participants to recall everything they had to eat /or drink in the 24 hours preceding the interview. Survey results are being used to augment the HHHQ food database with target population culturally specific foods. The Minnesota Nutrition Data System (NDS), a microcomputer-based system for collection and analysis of dietary data, was used to analyze 24 HDR data. The NDS has been updated to provide a more accurate analysis during the past Study year.

A third measure, the Nutrition Survey, included in the BCRA, is also administered. Developed by Alan Kristal of the Fred Hutchinson Cancer Research Center, Seattle, this instrument provides a fat and fiber score which allows researchers to estimate

total dietary content of these items (19). Additionally, recent changes made in fruit and vegetable consumption can also be determined. A manuscript, Family Resemblance in Modifiable Breast Cancer: A study among three generations of African American women, was submitted to the Journal of the American Dietetics Association. Salient findings from this paper were presented in the last report.

Results showed participants had an average daily Vitamin A intake of >5,000 IU, which is above the Recommended Dietary Allowance (RDA) of 4,000 IU. Cases and siblings consumed the highest levels of Vitamin A (6,770 IU); however, results from the nutrient estimates from supplements show none of the participants' average intake exceed the RDA for Vitamin A. All participants exceeded the RDA for Vitamin C (60 mg). Participants did not exceed the RDA for Vitamin E (8 mg). Mothers had the lowest average of Vitamin E (4 mg) as opposed to an average of 7 mg for both cases, siblings and offspring. In addition, the percent of calories from fat, excluding alcohol, was tabulated for participants from the HHHQ. Results demonstrated all members, excluding mothers, had diets higher in fat than is recommended. Cases, Siblings and offspring had an estimated fat intake $\geq$ 34%. Mothers' fat intake was at the recommended level ($\leq$ 30%).

The Food Frequency Questionnaires (HHHQ) were not done on the controls due to the impracticality of administering a lengthy questionnaire in a mobile screening environment. However, for a dietary intake of vitamins, the risk appraisal questionnaire is adequate to assess the controls' self-reported intakes as a categorical response. Higher Vitamin C intakes were reported by cases than by controls with an OR of 4.04 (95% CI 3.40, 4.68). A significantly higher percentage of cases reported taking Vitamins A, C and E than did controls ($p=0.001$).

Our health attitudes survey (a part of the Psychosocial Questionnaire) has allowed the examination of dietary beliefs related to breast cancer risk. These findings were reported during the recent American Psychological Association's Psychological and Behavioral Factors in Women's Health, September 17-20, 1996, Washington, D.C.. Table 3 outlines dietary health attitudes results from the Psychosocial questionnaire. Results indicate 88% of mothers believed *eating a low-fat diet reduces one's risk of getting breast cancer* as opposed to 79% of cases, 85% of siblings and 80% of offspring. No mothers or siblings disagreed that *It is important to eat foods low in fat.* Of those that disagreed with this statement, fifty-eight (58) percent were cases.

Table 3. Dietary Health Attitudes of Breast Cancer Cases and their Siblings, Mothers and Offspring

STATEMENT	CASES %	SIBLINGS %	MOTHERS %	OFFSPRING %
EATING A LOW-FAT DIET REDUCES ONE'S RISK OF GETTING BREAST CANCER	79	85	88	80
EATING A DIET HIGH IN VITAMIN A REDUCES ONE'S RISK OF GETTING BREAST CANCER	78	76	100	73
IT IS IMPORTANT TO EAT FOODS LOW IN FAT	93	100	100	92

AIM 3: Examine the role of obesity and adipose distribution in breast cancer risk. There is some evidence that regional distribution of adipose tissue is related to breast cancer risk (20). Women carrying excess weight on their upper bodies are at increased risk of getting breast cancer. For this aim, the role of obesity and adipose distribution in breast cancer risk were examined. Analyses showed the majority of participants had an even distribution of fat. There were no significant differences in fat distribution across family groups, although of those reporting fat distribution above the waist, cases more often reported this physical characteristic (45.5%).

Objective 2: Determine psychosocial profiles of breast cancer family groups.

Aim 1: Examine the impact of psychosocial factors on breast cancer. A psychosocial assessment was completed in three components: 1) Social Networks section, assessing participants' ties with friends, family and the community; 2) Health Attitudes section, examining cognitive/affective dimensions of personality which have been associated with health behaviors; and 3) General functioning determining the association between general emotional functioning, mood and health behavior. Preliminary findings related to this aim were presented in our last report as well as during the American Psychological Association's Psychological and Behavioral Factors in Women's Health, September 17-20, 1996, Washington, D.C.. The Principal Investigator also presented findings on the psychosocial

impact of breast cancer on African American family groups during the American Association of Cancer Research Inc. 88th Annual Meeting, April 12-16, 1997, Washington, D.C..

TABLE 4. PSYCHOSOCIAL HEALTH ATTITUDES OF BREAST CANCER CASES AND THEIR SIBLINGS, MOTHERS AND OFFSPRING

STATEMENT	CASES* %	SIBLINGS* %	MOTHER* %	OFFSPRING* %
THERE IS A LOT I CAN DO TO KEEP FROM GETTING SICK	80	100	100	91
THERE IS A LOT I CAN DO TO KEEP FROM GETTING CANCER	57	96	100	65
I WORRY A LOT ABOUT DEVELOPING CANCER	64	79	46	90
I AM PLEASED WITH THE EMOTIONAL SUPPORT PROVIDED BY MY FAMILY	94	97	81	92

***PERCENTAGES ARE BASED ON NON-MISSING VALUES**

Data in Table 4 represent responses to psychosocial health attitude statements. More than 93% of cases and siblings were *pleased with the emotional support provided by family and friends, respectively.* More offspring (90%) and siblings (77%) *worry a lot about developing cancer,* compared to 45% of mothers and 64% of cases. Fewer cases believed *there is a lot they can do to keep from getting sick.*

Objective 3: Construct pedigree data and bank DNA samples for conducting linkage studies to map breast cancer genes by analyzing the co-transmission of candidate markers, breast cancer and risk factors in multiple-case families. Cases and PFRs have completed family histories and had blood drawn. DNA extractions have been completed on the total sample. Family pedigrees were drawn using the Canvas computer-based program. Results are maintained in the project office in participant files (in a locked file cabinet), RAP database and the CANVAS computer program/Macintosh computer. **Aim 1: Identification of families carrying the BRCA1 gene. a) identify common risk factors in high risk families** Family history data were collected. Inherited mutations of BRCA1 gene are associated with a high risk of breast (82%), ovarian (44%), prostate (8%) and colon (6%) cancers in some families (21). Family history data, including self-reported family history of cancer, was collected on 51

breast cancer cases and their PFRs (total n = 151). Breast cancer cases reported 34 of their female relatives had breast cancer, 3 ovarian, 5 prostate and 3 colon cancer. Other cancers reported included cervical, brain, bone, stomach, lung, throat, uterine, spinal, rectal, sarcoma, gland, head and neck. Preliminary findings related to family history were presented in the Epidemiology Section of the American Public Health Association's 124th Annual Meeting, New York, NY, November 17-21, 1996 in a poster entitled: Genetic Investigation of African American Families with Breast Cancer.

b) Determine if breast cancer is familial due to specific genes or because risk factors themselves are familial DNA was extracted for each of the family groups and stored for later testing for BRCA1 mutations. Our participation in an adjunct study funded by the Health Foundation of South Florida (Fernando Arena, MD, Ph.D., Principal Investigator), allowed testing of selected DNA samples for BRCA1 mutations. A select sample of three (3) patients with highly positive family history was tested. Mutations were found in three cases and all were previously unreported mutations in exon 11 (943 ins 10bp, 3888 del GA and 4160 del AG). Because of these findings, 42 additional study participants with early-onset breast cancer were investigated for the presence of mutations in exon 11 of BRCA1. Eight (8) participants, or 19% had a positive family history for breast/ovarian cancer with at least one more affected relative. Our search, using four sets of PCR primers, was directed to the regions in exon 11 surrounding these mutations. Only one (1) mutation was found, a second 943 ins 10bp mutation, in an unrelated family. However, we found a previously undescribed polymorphism in exon 11 (A3557/g), which was present in 4 of 100 chromosomes from African American controls and in none of 46 chromosomes of white controls. These results were submitted in an abstract form to the 46th Annual Meeting of the American Society of Human Genetics, held in San Francisco, CA from October 29 - November 2, 1996 and was accepted for platform presentation at the section Cancer Genetics II: Breast Cancer.

CONCLUSIONS

Multigenerational Breast Cancer Risk Factors in African American Women or the Risk Appraisal Study (RAP), has gained momentum among breast cancer survivors and their primary female relatives. This is a descriptive study, and although the recruitment rate has not approached 100% the retention rate continues to be successful. The use of tumor registry data in participant recruitment has added considerably to our confidence to obtain an appropriate sample size. Controls, recruited from the ten year old EDP, are on target in terms of the expected numbers. Continued success in recruitment and retention for duration of the project is anticipated.

During Study Year 3, there were setbacks, some of which affected the case accrual rate.

The drawing of blood for the BRCA1 study was interrupted from October 1996 through April 1997, because of the Principal Investigator's (PI's) concerns related to issues of genetic testing. During this time, the massive letter writing campaign continued for the purposes of recruiting eligible participants to the study. On May 14, 1997, issues related to the genetic testing were resolved. Permission to begin scheduling the blood drawings and interviews was initiated.

On May 21, 1997, the University of Miami, Medical Sciences Subcommittee for the Protection of Human Subjects in Research (MSSPHSR) informed the PI that no further recruiting of subjects be done until there was compliance with modifying the informed consent. A copy of the revised Informed Consent is found in Appendix A. On September 15, 1997, the revised consent was approved and recruitment and follow-up on subjects resumed. This project studies family groups and female offspring of the breast cancer case were invited to participate. Breast cancer cases are women under the age of 50 years which suggest their children could be minors. In our two previous annual reports, data on offspring included minors, but due to the compliance with MSSPHSR requests, at this time, the data collected for minors cannot be used. A family now consists of the case and at least one primary female relative.

As stated in the last report, the intent was to complete all interviews in the homes of breast cancer cases and their primary female relatives. However, interviews were restructured to take place in the most convenient location for the participant. Additionally, many local women were interested in participating in the project but didn't have PFRs residing in Dade County. In order to meet recruitment goals, relatives living in other parts of the country were included. Strict quality control issues were considered in blood handling and informed consent is obtained from all study participants, prior to data collection.

We are committed to continuing our increased effort to meet the recruitment and retention goals. We plan to meet the set goal of 200 cases at the end of Study Year 4.

Family history and age are accepted risk factors for breast cancer. Pedigree data collected on study participants indicates a strong relationship between family history and our breast cancer cases. Age, however, has not been as strong an indicator. Cases in the present study are below 50 years of age. For the most part, cases in the study reporting a family history of breast cancer, had older female relatives (i.e., mothers, aunts, grandmothers) with personal histories of breast cancer at ages within 10 years of their diagnoses.

Data on selected reproductive risk factors indicate that cases and controls are similar except with a difference in age at first birth.

Primary prevention of breast cancer through lifestyle intervention has become increasingly important in epidemiologic research. Antioxidant vitamins, in particular, are in the forefront of this research. Data on vitamins A, C, E and beta carotene from the present study are valuable in that they will assist in determining diet-genetic links in breast cancer risk and these data will also assist in designing breast cancer interventions. Based on the participant enrollment at this time, it is difficult to draw conclusions related to dietary intakes.

Psychosocial results from the study show that 96% of siblings feel they *can keep from getting breast cancer*. This is an important finding in that most female relatives of breast cancer survivors often feel helpless in terms of preventing this disease. Ninety-four (94) percent of cases are *pleased with the emotional support provided by family and friends, respectively.* Additional psychosocial data analysis will continue in Study Year 4.

At the time the original grant application was submitted, methods included the extraction of DNA for later testing for BRCA1 mutations. Since the award, the location of the gene presented unique opportunities for actual testing for mutations of the gene. Through collaboration with Fernando Arena, MD, Ph.D., the project geneticist, we were able to present previously unreported BRCA1 mutations in a sub-sample of high risk cases. Further work is in progress to identify additional and confirmed mutations in the BRCA1 gene in patients with one or more affected relatives with breast/ovarian cancer.

Study Year 4 will include a continuation of the descriptive study. The development of the RAP System database has equipped project leadership with ability to track project participants. Risk factors will be assessed on an additional 100 breast cancer cases and their primary female relatives. Cases will continue to be matched to age-ethnic-specific controls. Through linkage studies, likely cases for BRCA1 mutations will complete genetic assessments. As genetic and epidemiological links to breast cancer continue to be uncovered (as is anticipated by the end of this research), counseling programs will emerge. Environmental and genetic risk factors will continue to be analyzed. A better understanding of the interaction between environmental factors and genes will prepare the scientific and

medical community to cope with breast cancer and issues of prevention.

REFERENCES

1. CDC. Breast and cervical cancer surveillance, United States, 1973-1987. In: CDC surveillance summaries (April). *MMWR* 1992'41(no. SS-2):1-15.

2. Kosary CL, Ries LAG, Miller BA, et al, eds. SEER cancer statistics review, 1973-1992: tables and graphs. Bethesda, Maryland: US Department of Health and Human Services, Public Health Service, *National Institutes of Health, National Cancer Institute*, 1995; publication no. (NIH)96-2789.

3. American Cancer Society. Cancer facts and figures, 1996. Atlanta, Georgia: *American Cancer Society*, 1996; publication no. 5008.96.

4. NCHS. Vital statistics mortality data, multiple cause of death, 1973-1992 [Machine-readable public-use data tapes]. Hyattsville, Maryland: US Department of Health and Human Services, Public Health Service, CDC, 1996.

5. Mayberry, Robert M. (1994) Age-specific patterns of association between breast cancer risk factors in black women, ages 20 to 39 and 40 to 54. *Ann Epidemiol* 4:205-213.

6. Lerman C, Lustbder E, Rimer B, et al., (1995) Effects of individualized breast cancer risk counseling: a randomized trial. *Journal of the National Cancer Institute*, February 15, 87(4):286-292.

7. Kash KM, Holland JC, Marilyn SH, Miller DG (1992). Psychosocial distress and surveillance behaviors of women with a family history of breast cancer. *Journal of the National Cancer Institute* 84:24-30.

8. Sattin RW, Rubin GL, Webster LA, et al., (1985). Family history and the risk of breast cancer. *JAMA* 253:1908-1913.7.

9. Bowcock AM, Anderson LA, Friedman LS, Black DM, Osborne-Lawrence S, Rowell SE, Hall JM, Solomon E, King MC (1993) THRA1 and D17S183 flanked an interval of <4 Cm for the breast-ovarian cancer gene (BRCA1) on chromosome 17q21. *Am J Hum Genet* 52:718-722.

10. Easton DF, Bishop DT, Ford D, Crockford GP and Breast Cancer Consortium (1993). Genetic Linkage analysis in familial breast and ovarian cancer: Results from 214 families. *Am. J. Hum. Genet* 52(4):678-7-1.

11. Miki Y, Swensen J, Shattuck-Eidens D, Futreal PA, Harshman K, Tavtigian S, Liu Q, et al (1994). A strong candidate for the breast and ovarian cancer susceptibility Gene BRCA1. *Science* 266:66-71.

12. Henderson BE, Ross RK, Judd HL et al., (1985). Do regular ovulatory cycles increase breast cancer risk. *Cancer* 56:1206-1208.

13. MacMahon B (1973) Age at menarche. *Vital Health Stat* 11:11-27.

14. MacMahon B, Worcester J (1966) Age at menopause: United States-1960-1962. *Vital Health Stat* 11:1-20.

15. Gray GE, Henderson BE, Pike MC (1980) Changing ratio of breast cancer incidence rates with age of black females compared with white females in the United States. *J Natl Cancer Inst* 54:461-463.

16. Kelsey JL, Gammon MD, John EM (1993) Reproductive factors and breast cancer. *Epidemiol Rev* 15:36-47.

17. McPherson K, Steel CM, Dixon JM (1994) Breast cancer--epidemiology, risk factors and genetics. *BJM* 309:1003-1006.

18. Moormeier J (1996) Breast cancer in black women. *Ann Intern Med* 124:897-905.

19. Kristal AR, Beresford SAA, Lazovich D (1994) Assessing change in diet-intervention research. *Am J Clin Nutr* (Suppl):1858-189S.

20. Folsom AR, Kaye SA, Prineas RJ (1990). Increased incidence of carcinoma of the breast associated with abdominal adiposity in postmenopausal women. *Am J Epidemiology* 131:794.

21. Simard J, Tonin P, Durocher F, Morgan K, et al., (1994). Common origins of BRCA1 mutations in Canadian breast and ovarian cancer families. *Nature Genetics* 8:392-398.

APPENDIX A

INFORMED CONSENT
Multigenerational Breast Cancer Risk Factors
in African American Women

PURPOSE:
You are being asked to participate in a project to study risk factors in breast cancer patients and their families. This is a screening program and will not include a complete examination. If you agree to participate in this study, you will be asked questions related to your medical history, your diet, and your family history.

PROCEDURES:
The interview will be conducted in your home or over the phone and will take approximately 1 hour. Some participants will be asked to take part in a research study to see if known and suspected risk factors for breast cancer are present in patients with breast cancer and their family groups. A family group will be defined as mother, sister(s), and/or female offspring. In order to participate in the study, the breast cancer patients and at least two members of her family group must agree to participate in the study. If agreed, you will be asked to complete a breast risk appraisal form. This form will be administered to you by a health researcher and will take 20 to 30 minutes to complete. Information on your usual and present diet will also be obtained. A laboratory technician may draw 20 ml (about 1 tablespoon) of blood from you in order to examine the DNA content of your blood. The DNA content of the blood sample will be frozen and stored for future testing.

Genetics research is an increasingly important way to try to understand the role of genes in human disease. You have been given this consent form because University of Miami investigators want to include your tissue, cell or blood sample in a research project, or because they want to save such biological samples for future research. There are several things you should know before allowing your tissues, cells or blood to be studied or to be stored for future study.

1. Your tissue, cell or blood sample will be stored under your name or other unique identifier. If there is a medical reason to seek specific information from you in the future, your doctor will tell you about this. A process called "genetic counseling" is often appropriate in such cases; you should ask your doctor or nurse about this if you have any questions.

2. Your confidentiality will be protected to the extent permitted by law. Your records might be reviewed by government officials or by corporate research sponsors. The University of Miami collaborates with many other organizations, and data are generally shared among them. No data shared with other investigators will include your name or other public identifier, however.

I.R.B.
APPROVAL DATE: 9/15/97
INITIALS: A.A.

3. Genetic research may affect your insurability. For instance, information about your DNA might result in discrimination that would make it difficult for you to obtain certain health insurance coverage in the future. You will still be responsible for paying for health care, however. The University of Miami will not be responsible for such costs, even if care is needed for a condition revealed during research or clinical testing.

4. You have the right to refuse to allow your tissue or blood to be studied or saved for future study. You may withdraw from a study at any time, and remove from research use any samples that contain identifiers after the date of your withdrawal. This means that while the university might retain the identified samples--the law often requires this-- they would not be used for research. Samples without identifiers might still be retained for research; a different consent form is usually used in such cases.

5. Genetic information about you will often apply (in one degree or another) to family members. It is not generally the University's policy to provide genetic information about you to your family members. However, certain studies, called "pedigree studies," share such information among family members. For this and related research, you will be asked if you are willing to share your genetic information with your family members.

6. Your tissue samples might be used to develop commercially valuable medical products. By signing this form you agree not to seek a share of any proceeds that might result; that is, you waive any claim to share in the commercialization of products developed from your tissue or blood samples.

7. In addition to your name, other information about you might be connected to your blood or tissue sample. For instance, information about race, ethnicity, sex, your medical history, and so forth might be available to investigators studying your tissue or blood. Such information is important for scientific context and sometimes for public health. It is possible that genetic information might come to be associated with your racial or ethnic group.

8. It is possible that more tissue or blood samples will be obtained than are necessary for your treatment. That is, investigators might take samples purely for study purposes.

9. Genetic research raises difficult questions about informing you and other subjects of any results, or of future results. Some people want to know what is found out about them; others do not. The risks of knowing include anxiety and other psychological distress, and the possibility of insurance discrimination. The risks of not knowing what is found include not being aware of the need for treatment. These risks can change depending on whether there is a treatment or cure for a particular disease.

10. **In this study, investigators will *not* tell you what they find out about you, nor will they contact you if a test becomes available to diagnose a condition you might have or later develop.**

    For instance, suppose the investigators discover that your tissue sample carries a gene for a disease. Neither the university nor your doctor will try to contact you or find you to tell you about this gene. While we might not know how to test for a particular disease gene today, we might be able to test for it in the future. The number of genes for which this will be possible in the future is quite large--and a policy that required contact at a later date would be overwhelming.

11. The presence of a genetic marker does not necessarily mean that a patient will develop a disease. Informing people of all such markers independently of medical need can cause unnecessary anxiety. Conversely, the absence of a marker does not mean that someone will not get the disease. "Genetic diseases" appear as a result of a complex mixture of hereditary, environmental, behavioral and other factors.

12. There are alternatives to notification by investigators. If you are concerned about a potential genetic malady, you and your doctor might choose to test specifically for it; this would require additional blood or tissue samples. You should discuss this option with your doctor or genetic counselor.

13. With respect to the future use of tissue samples, you agree that: 1) you waive the right to consent to or be notified of any future research, test or analysis which might be performed using these tissue samples, and 2) you waive any rights to any results or information generated by any future research, tests or analysis using those tissue samples and assign any such rights to the University of Miami.

These are the best-known risks and challenges of genetic research. There might be other risks we do not know about yet. No direct benefits can be promised from your participation, though some people find satisfaction in contributing to scientific knowledge about genetic problems and their medical consequences. It is very important that you talk to your doctor, nurse or genetic counselor if you have questions or concerns about the research study or any of the information in this document.

RISKS:
There are some risks involved in having your blood drawn. There is the risk of temporary discomfort and/or bruising at the site of puncture and fainting. On rare occasions infection or the formation of a small clot or swelling to the vein and surrounding area may occur.

BENEFITS:
Although no benefit is promised to you from your participation in this study, the knowledge gained could prove beneficial to you and your family members specifically, and to the African American community in general. There will be no costs associated with your participation.

COMPENSATION:
In any screening program diseases or medical conditions may be uncovered that are not related to the medical condition under study, it is agreed that any conditions that are uncovered will be the ultimate responsibility of you to follow-up. If any abnormal findings are uncovered, you will be referred to a physician for further diagnosis and follow-up. The responsibility of seeking this follow-up is up to you. You are authorized all necessary medical care for injury or disease which is the proximate result of your participation in this research. Other than medical care that may be provided, you will not receive any compensation for your participation in this research study.

CONFIDENTIALITY:
Your consent to participate in the study includes consent for the investigator and her assistants to review all your medical records as may be necessary for the purpose of the study. The investigator and her assistants will consider your records confidential to the extent permitted by law. Your records and results will not be identified as pertaining to you in any publication without your expressed permission. Your stored blood samples will not be used in any other studies without your expressed permission. In rare circumstances, the U.S. Food and Drug Administration (FDA) or the U.S. Department of Health and Human Services (DHHS) may request copies of your records. If this happens, the FDA or DHHS request will be honored. Representatives from the U.S. Army Medical Research, Development, Acquisition and Logistics Command are also eligible to inspect the records of this research project as a part of their responsibilities to protect human subjects in research. Your records may also be reviewed for audit purposes by authorized University of Miami employees or other agents who will be bound by the same provisions of confidentiality.

ALTERNATIVE:
You have the right not to participate in this study.

RIGHT TO WITHDRAW:
Participation in this research project is voluntary and you have the right to withdraw from or refuse to participate in this study at any time with no prejudice to you seeking future medical care at the UM/JMMC. You may ask and will receive answers to any questions concerning your rights as a research subject by contacting Maria Arnold, Institutional Review Board Director, University of Miami, at (305)243-3327. A copy of the signed consent form will be provided to you. If you agree to participate in this study, please initial the front page of the consent form and sign below.

I have read and understand the Informed Consent and agree to participate in the project and I agree to have blood drawn for the DNA testing.

CONSENT FOR SUBJECTS:

_____ _____
Signature of patient Date

_____ _____
Signature of witness Date

For specific questions about the research study or to report any research related problems you may contact Selina A. Smith, Ph.D., Principal Investigator (305)243-6599. For specific medical related questions about this study you may contact Dr. Stephen Richman, M.D. (305) 243-4909.

Revised 9/15/97

DEPARTMENT OF THE ARMY
US ARMY MEDICAL RESEARCH AND MATERIEL COMMAND
504 SCOTT STREET
FORT DETRICK, MARYLAND 21702-5012

REPLY TO
ATTENTION OF:

MCMR-RMI-S (70-1y)

1 JUN 2001

MEMORANDUM FOR Administrator, Defense Technical Information Center (DTIC-OCA), 8725 John J. Kingman Road, Fort Belvoir, VA 22060-6218

SUBJECT: Request Change in Distribution Statement

1. The U.S. Army Medical Research and Materiel Command has reexamined the need for the limitation assigned to technical reports. Request the limited distribution statement for reports on the enclosed list be changed to "Approved for public release; distribution unlimited." These reports should be released to the National Technical Information Service.

2. Point of contact for this request is Ms. Judy Pawlus at DSN 343-7322 or by e-mail at judy.pawlus@det.amedd.army.mil.

FOR THE COMMANDER:

Encl

PHYLIS M. RINEHART
Deputy Chief of Staff for
Information Management

Reports to be changed to "Approved for public release; distribution unlimited"

Grant Number	Accession Document Number
DAMD17-94-J-4147	ADB221256
DAMD17-93-C-3098	ADB231640
DAMD17-94-J-4203	ADB221482
DAMD17-94-J-4245	ADB219584
DAMD17-94-J-4245	ADB233368
DAMD17-94-J-4191	ADB259074
DAMD17-94-J-4191	ADB248915
DAMD17-94-J-4191	ADB235877
DAMD17-94-J-4191	ADB222463
DAMD17-94-J-4271	ADB219183
DAMD17-94-J-4271	ADB233330
DAMD17-94-J-4271	ADB246547
DAMD17-94-J-4271	ADB258564
DAMD17-94-J-4251	ADB225344
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DAMD17-94-J-4251	ADB259028
DAMD17-94-J-4499	ADB221883
DAMD17-94-J-4499	ADB233109
DAMD17-94-J-4499	ADB247447
DAMD17-94-J-4499	ADB258779
DAMD17-94-J-4437	ADB258772
DAMD17-94-J-4437	ADB249591
DAMD17-94-J-4437	ADB233377
DAMD17-94-J-4437	ADB221789
DAMD17-96-1-6092	ADB231798
DAMD17-96-1-6092	ADB239339
DAMD17-96-1-6092	ADB253632
DAMD17-96-1-6092	ADB261420
DAMD17-95-C-5078	ADB232058
DAMD17-95-C-5078	ADB232057
DAMD17-95-C-5078	ADB242387
DAMD17-95-C-5078	ADB253038
DAMD17-95-C-5078	ADB261561
DAMD17-94-J-4433	ADB221274
DAMD17-94-J-4433	ADB236087
DAMD17-94-J-4433	ADB254499
DAMD17-94-J-4413	ADB232293
DAMD17-94-J-4413	ADB240900

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