Award Number: DAMD17-98-1-8083

TITLE: Breast Cancer Biomarkers Based on Nipple and Fine Needle Aspirates

PRINCIPAL INVESTIGATOR: Michael Torosian, M.D.

CONTRACTING ORGANIZATION: Fox Chase Cancer Center
Philadelphia, Pennsylvania 19111

REPORT DATE: May 2000

TYPE OF REPORT: Annual

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

DISTRIBUTION STATEMENT: Approved for Public Release;
Distribution Unlimited

The views, opinions and/or findings contained in this report are those of the author(s) and should not be construed as an official Department of the Army position, policy or decision unless so designated by other documentation.
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Breast cancer is the most common cancer and a leading cause of cancer deaths in women in the United States. The purpose of this project is to identify one or more biomarkers which can be used to identify women at high-risk of developing breast cancer before cancer develops. Prior to undergoing breast surgery, women at increased and normal risk of developing breast cancer will undergo fine needle aspiration (FNA) for determining a series of cytologic, cell cycle and molecular biologic markers of clinical significance for cancer development. These biomarkers include: cytology, DNA index, cell cycle parameters, proliferation index, epidermal growth factor receptor overexpression, p53 and RAS hotspot mutations and hypermethylation of specific gene products (p16, GSTP and E-cadherin). Michael Torosian, M.D. has recently assumed the role of principal investigator of this project and has updated the clinical and scientific aspects of this project. Submission of this project to the IRB at Fox Chase Cancer Center is anticipated within the next 3 months and subject accrual should begin immediately after IRB approval.
FOREWORD

Opinions, interpretations, conclusions and recommendations are those of the author and are not necessarily endorsed by the U.S. Army.

( ) Where copyrighted material is quoted, permission has been obtained to use such material.

( ) Where material from documents designated for limited distribution is quoted, permission has been obtained to use the material.

( ) Citations of commercial organizations and trade names in this report do not constitute an official Department of the Army endorsement or approval of the products or services of these organizations.

( ) In conducting research using animals, the investigator(s) adhered to the "Guide for the Care and Use of Laboratory Animals," prepared by the Committee on Care and Use of Laboratory Animals of the Institute of Laboratory Animal Resources, National Research Council (NIH Publication No. 86-23, Revised 1985).

( ) For the protection of human subjects, the investigator(s) have adhered to policies of applicable Federal Law 32 CFR 219 and 45 CFR 46.

( ) In conducting research utilizing recombinant DNA technology, the investigator(s) adhered to current guidelines promulgated by the National Institutes of Health.

Michael X. Forouzan, MD
Principal Investigator's Signature

Date

5-26-02

Attachment J.5.
Table of Contents

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Breast cancer is the most common cancer and a leading cause of cancer deaths in women in the United States. The purpose of this project is to identify one or more biomarkers which can be used to identify women at highest risk of developing breast cancer before cancer develops. Prior to undergoing breast surgery, women at increased and normal risk of developing breast cancer will undergo fine needle aspiration (FNA) for determining a series of cytologic, cell cycle and molecular biologic markers of clinical significance for cancer development.

The specific aims of this study are to:

1. Compare the findings of biomarkers in FNA specimens in women at increased versus normal risk of breast cancer.
2. To determine if FNA specimens are adequate to perform biomarker assays and if this technique could be used in the general outpatient setting.
3. To determine if biomarker levels between high risk and control patients are concordant or discordant and if one or more of the specific biomarkers can distinguish high-risk from control patients. Specifically, the biomarkers will include cellular morphology (cytology), cell cycle analysis (DNA index, cell cycle parameters and proliferation index) and molecular biologic markers (epidermal growth factor receptor overexpression, p53 and RAS hotspot mutations and hypermethylation of specific gene products including p16, GSTP 1 and E-cadherin) of clinical significance for the development of cancer.

At Fox Chase Cancer Center, a combined research team of clinicians and researchers are able to perform these analyses. Samples will be collected on patients who consent to the study and who are undergoing elective breast surgery. Our goal is to study approximately 200 patients over a 3 year period with this technique.

This is an import research project with tremendous clinical implications. All of the biomarkers chosen for testing and analysis have known importance for the development of cancer. Our goal is to identify one or more of these biomarkers which will provide important information regarding breast cancer risk and diagnosis not obtainable with currently available breast cancer screening methods. Michael H. Torosian, M.D. has recently assumed the role of principal investigator of the project and has updated the clinical and scientific aspects of this project. Over the next several months, several important meetings of the co-investigators will occur prior to formal submission of this research application to the IRB at Fox Chase Cancer Center. Once approval is received from the IRB, subject accrual to this important project is expected to occur immediately.
Statement of Work

Months 1-2: Meet with all Co-Investigators to finalize project
Perfor final statistical projections and make final project revisions

Months 3-4: Submit project proposal to Research Review Committee and IRB at Fox Chase Cancer Center

Months 5-12 Patients to be accrued to study
FNA specimens to be obtained
Biomarker assays to be performed on an ongoing basis
a. Cellular morphology (cytology)
b. DNA index (ploidy) and cell cycle parameters (% cells in S phase, % cells in G2M, % hypertetraploid cells)
c. Proliferation index
d. Epidermal growth factor receptor (EGFR) overexpression
e. p53 hot spot mutations
f. RAS hot spot mutations
g. Hypermethylation of specific gene products
   1. p16
   2. GSTP 1
   3. E-Cadherin

Months 13-36: Continue to accrue patients to study, obtain FNA specimens and perform biomarker assays.
Perform statistical analysis and prepare results for publication
**BREAKDOWN OF PROPOSED COST AND HOURS**

15-Jun-00  
**Period:** July 1, 2000 - June 30, 2001

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## Breakdown of Proposed Cost and Hours

**Period:** July 1, 2001 - June 30, 2002

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<td>Principal Investigator</td>
<td>$72.46</td>
<td>195.00</td>
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<td>P. Cairns, Ph.D.</td>
<td>Co-Investigator</td>
<td>43.51</td>
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<td>L. Giles</td>
<td>Clinical Research Coordinator</td>
<td>24.97</td>
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### Health Benefits at 32%

- 11,136

### Major Equipment

- 0

### Materials

- Clinical Supplies: 1,030
- Laboratory Supplies: 6,286

### Subcontracts

- 0

### Travel

- PI to annual DOD Breast Cancer Meeting: 500

### Publication and Report Costs

- 0

### Consultant

- 0

### Other

- Data Management: 1,030
- Image Analysis: 515
- Cytology Evaluations: 4,635
- Statistical Support: 515

### Sub-total direct costs

- 60,446

### Indirect Costs at 72.0% MTDC per HHS agreement date 12/31/97

- 43,521

**Total Costs**

- $103,968
### Breakdown of Proposed Cost and Hours

**15-Jun-00**  
*Period: July 1, 2002 - February 28, 2003*

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<th>Name</th>
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<th>Hourly Rate</th>
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<td>M. Torosian, M.D.</td>
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<td>P. Cairns, Ph.D.</td>
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<td>44.81</td>
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<td>L. Giles</td>
<td>Clinical Research Coordinator</td>
<td>25.72</td>
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(B) Health Benefits at 32%  

(C) Major Equipment  

(D) Materials  
- Clinical Supplies: $707  
- Laboratory Supplies: $5,300  

(E) Subcontracts  

(F) Travel  
- PI to annual DOD Breast Cancer Meeting: $0  

(G) Publication and Report Costs  

(H) Consultant  

(I) Other  
- Data Management: $696  
- Image Analysis: $350  
- Cytology Evaluations: $3,180  
- Statistical Support: $1,400  

Sub-total direct costs: $39,310  

(J) Indirect Costs at 68.0% MTDC per HHS agreement date 12/31/97  

TOTAL COSTS: $66,041
Breast Cancer Biomarkers Based on Fine Needle Aspirates

Appendix

A. Curriculum Vitae
MICHAEL H. TOROSIAN, M.D.

CURRICULUM VITAE

Home Address: 

Office Address: Fox Chase Cancer Center
Department of Surgery – C308
7701 Burholme Avenue
Philadelphia, Pennsylvania 19111
Telephone: 215-728-2596
Fax: 215-728-2773

Social Security Number: 

Birthdate: 

Birthplace: 

Child: 

Undergraduate Education: Princeton University
Princeton, New Jersey
(1970 - 1974)

Degree: Bachelor of Arts
Major: Biochemical Sciences
Honors: Summa Cum Laude
Phi Beta Kappa
Sigma Xi

Medical Education: University of Pennsylvania School of Medicine
Philadelphia, Pennsylvania
(1974-1978)

Degree: Doctor of Medicine
Medical Licensure: Pennsylvania - MD-024131-E

Postgraduate Training:

1978 - 1979 Intern in Surgery, Hospital of the University of Pennsylvania Philadelphia, Pennsylvania

1979 - 1984 Resident in Surgery, Hospital of the University of Pennsylvania Philadelphia, Pennsylvania

1984 - 1985 Chief Resident in Surgery, Hospital of the University of Pennsylvania Philadelphia, Pennsylvania

1985 - 1986 Advanced Fellow in Surgical Oncology, Memorial Sloan-Kettering Cancer Center, New York, New York

1985 - 1986 Fellow in Surgery, Cornell University School of Medicine New York, New York

Surgical Oncology Fellowships:

1980 - 1981 Clinical Fellowship, American Cancer Society, Department of Surgery, Hospital of the University of Pennsylvania

1985 - 1986 Clinical Fellowship, American Cancer Society, Department of Surgery, Memorial Sloan-Kettering Cancer Center

1985 - 1986 Advanced Surgical Oncology Fellowship, Department of Surgery Memorial Sloan-Kettering Cancer Center

1985 - 1986 Fellow in Surgery, Cornell University School of Medicine

Research Fellowships:

1972- Research Fellowship, National Science Foundation, Department of Biochemical Sciences, Princeton University

1973 - Research Fellowship, Department of Biochemical Sciences Princeton University

1981 - 1983 Research Fellowship, Harrison Department of Surgical Research, Department of Surgery, Hospital of the University of Pennsylvania
1981 - 1983  Research Fellowship, Measey Foundation, Department of Surgery, Hospital of the University of Pennsylvania

Faculty Appointments:

1979 - 1984  Assistant Instructor of Surgery, University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania

1984 - 1985  Instructor of Surgery, University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania

1986 - 1992  Assistant Professor of Surgery, University of Pennsylvania School of Medicine, Philadelphia, Pennsylvania

1993 - 1998  Associate Professor of Surgery, University of Pennsylvania, School of Medicine, Philadelphia, Pennsylvania

1998-        Associate Professor of Surgery, Temple University School of Medicine, Philadelphia, Pennsylvania

1998-        Clinical Director of Breast Surgery Research, Fox Chase Cancer Center, Philadelphia, Pennsylvania

1999-        Program Director, Surgical Oncology Fellowship, Fox Chase Cancer Center, Philadelphia, Pennsylvania

Hospital Appointments:

1986 - 1998  Attending Surgeon, Division of Surgical Oncology, Department of Surgery, Hospital of the University of Pennsylvania, Philadelphia, Pennsylvania

1986 – 1998  Attending Surgeon, Department of Surgery, Veterans Administration Medical Center, Philadelphia, Pennsylvania

1998-        Attending Surgeon, Department of Surgery, American Oncologic Hospital, Philadelphia, Pennsylvania

Specialty Certification:

1986        American Board of Surgery

1996        American Board of Surgery -- Recertification

Fellowship:

1989        American College of Surgeons
Research Awards:

1983 Harry M. Vars Research Award, American Society for Parenteral and Enteral Nutrition, Seventh Clinical Congress, Washington, D.C.

1984 Jonathan E. Rhoads Research Award
First Annual Award, Department of Surgery, University of Pennsylvania School of Medicine

1987 - 1990 Career Development Award, American Cancer Society, National Division

1988 - 1992 Pew National Nutrition Program, Faculty Nutrition Fellowship, National Award

Research Award Mentor: Scott L. Charland - American Society for Parenteral and Enteral Nutrition, 1992, National Award

Teaching Awards:

1985 - Residents' Honor Roll, Excellence in Teaching
University of Pennsylvania School of Medicine

1992 - Outstanding Teaching Award, Medical School Class of 1992, University of Pennsylvania School of Medicine

Clinical Distinctions:

1994-1996 Top Docs in Philadelphia
Philadelphia Magazine

1996-1998 The Best Doctors in America
1st Edition -- Northeast Region
S.W. Naifeh, Editor
Woodward/White, Inc.

1997 Guide to Good Health
Top Doctors
Philadelphia Magazine

Professional Advisory Boards:

1992-present The Wellness Community
Cancer Patients & Recovery
Philadelphia, PA

Disease-Management System
Fort Washington, PA
Professional Society Memberships:

National and International Societies:

1974 - Sigma Xi - Scientific Research Society
1980 - American College of Surgeons - Candidate Group
1982 - American Society for Parenteral and Enteral Nutrition
1983 - American Medical Association
1985 - Association for Academic Surgery
1985 - American Association for the Advancement of Science
1986 - New York Academy of Sciences
1988 - American Society of Clinical Oncology
1988 - American Association for Cancer Research
1988 - Radiation Therapy Oncology Group
1988 - Eastern Cooperative Oncology Group
1988 - American College of Surgeons -- Fellowship
1989 - Society of University Surgeons
1989 - American Institute of Nutrition
1989 - American Federation for Clinical Research
1989 - American Society for Clinical Nutrition
1990 - Society of Surgical Oncology
1991 - Collegium Internationale Chirurgiae Digestivae
1991 - Society for Surgery of the Alimentary Tract
1991 - Physiological Society of Philadelphia
1993 - Surgical Biology Club III/American College of Surgeons
1994 - International Society for Nutrition and Cancer
1994 - International Association for Surgical Metabolism and Nutrition
1994 - Southeastern Surgical Congress
1995 - Societe Internationale de Chirurgie
1996 - Society for Nutritional Oncology Adjuvant Therapy
1997 - Association of American Medical Colleges
1999 - American Society of Breast Surgeons

Local Societies:

1986 - Ravdin-Rhoads Surgical Society
1988 - Pennsylvania Oncologic Society
1988 - John Morgan Society
1989 - College of Physicians of Philadelphia
1995 - Philadelphia Cancer Research Association
Editorial Advisory Board:

1995 - Cancer

Editorial Reviewer:  
Cancer Research  
Cancer  
Surgery  
Journal of Parenteral and Enteral Nutrition  
Critical Reviews in Clinical Laboratory Sciences  
Life Sciences  
Southern Medical Journal

Committees of National and International Organizations:

1988 - 1990  Education Committee, Association for Academic Surgery  
1988 - 1989  Standards Committee, American Society for Parenteral and Enteral Nutrition  
1988  - GI Surgery Subcommittee, Radiation Therapy Oncology Group  
1989 - 1990  14th Clinical Congress Planning Committee, Co-Director American Society for Parenteral and Enteral Nutrition  
1990 - 1992  Publications Committee, American Society for Parenteral and Enteral Nutrition  
1991 -  Committee on Issues and Government Affairs, Society of Surgical Oncology  
1991 -  Pennsylvania District #5 Committee on Applicants, American College of Surgeons  
1994 -  Research and Data Committee, American Society for Parenteral and Enteral Nutrition  
1995 -  Scientific Program Committee, International Association for Surgical Metabolism and Nutrition  
1995 -  North American Delegate, Regional Council of Continental Delegates, International Association for Surgical Metabolism and Nutrition  
1995 -  Bone and Soft-Tissue Sarcoma Subcommittee, Radiation Therapy Oncology Group  
1997 -  Group on Educational Affairs, Association of American Medical Colleges

Committees of the University of Pennsylvania Medical Center:

1984 - 1991  Medical School Annual Giving, Executive Committee  
Co-Chairman Morgan Shippen Society, University of Pennsylvania School of Medicine  
1987 - 1993  Medical Board, Hospital of the University of Pennsylvania
1987 - 1988  Tumor Registry Committee - Chairman, Hospital of the University of Pennsylvania

1989 - 1991  Senate Executive Committee and University Council, Elected Member, University of Pennsylvania

1989 -  1991  Medical-Legal Committee, Hospital of the University of Pennsylvania

1990 -  Committee to Review the Department of Radiation Oncology, Hospital of the University of Pennsylvania

1993 -  Admissions Committee, University of Pennsylvania School of Medicine

1993 -  Principles of Practice, Executive Committee, University of Pennsylvania Medical Center

1993 -  Clinical Trials Scientific Review and Monitoring Committee, University of Pennsylvania Cancer Center

1993 -  Oncology Strategic Planning Committee, University of Pennsylvania Medical Center

1994 -  Medical Quality Assessment & Improvement Committee Hospital of the University of Pennsylvania

1994 -  Gastrointestinal Disease Strategic Planning Committee, Gastrointestinal Oncology Subcommittee

1996 -  National Alumni Council University of Pennsylvania Medical Center

1997 - 1998  Reunion Chairman
             20th Year Reunion - Medical School Class of 1978
             University of Pennsylvania Medical Center
Research/Academic Grants:

1986 - 1987
American Cancer Society
Institutional Research Grant IN-135
Philadelphia, PA
"Effect of Parenteral Nutrition on Host/Tumor Metabolism"
M. H. Torosian (P.I.)
$7,500.

1987 - 1990
Career Development Award
American Cancer Society, National Division
M. H. Torosian (Recipient)
$90,000

1987 - 1988
Thomas B. and Jeannette E. Laws McCabe Fund
Research Grant
Philadelphia, PA
"Nutritional and Chemotherapy Related Toxicity"
M.H. Torosian (P.I)
$7,500

1987 - 1989
Clintec Nutrition Company
Research Grant #9088E
Deerfield, IL
"Parenteral Nutrition Can Improve Tumor Response to Chemotherapy"
M.H. Torosian (P.I.)
$31,790

1987 - 1991
Merit Review Grant
Veterans Administration Medical Center
Washington, D.C.
"Effect of Nutrition on Tumor Response and Host Toxicity to Chemotherapy"
M.H. Torosian (P.I.)
$113,100

1988 - 1992
Pew National Nutrition Program
Faculty Nutrition Fellowship, T86-00279-011
Ithaca, NY
"Abnormal Carbohydrate and Lipid Substrate Cycling in the Tumor - Bearing Host"
M.H. Torosian (Recipient)
$50,000
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<td>Bethesda, MD</td>
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<td>1994-1997</td>
<td>John Wayne Cancer Institute</td>
<td>JWCI Melanoma Multicenter Selective Lymphadenectomy Trial</td>
<td>M.H. Torosian (P.I.)</td>
<td>$10,000</td>
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<tr>
<td>Years</td>
<td>Sponsor</td>
<td>Description</td>
<td>Principal Investigator(s)</td>
<td>Funding Amount</td>
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<td>1996-1998</td>
<td>American Cancer Society, National Division</td>
<td>&quot;Breast Cancer Staging with High Resolution MR Imaging&quot;</td>
<td>Michael H. Torosian (Co-Investigator)</td>
<td>$200,000</td>
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<td>1997-2000</td>
<td>National Institutes of Health, Bethesda, MD</td>
<td>&quot;Breast Cancer Patterns of Care and Morbidity&quot;</td>
<td>M.H. Torosian (Co-Investigator)</td>
<td>$2,077,782</td>
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<tr>
<td>1998-2003</td>
<td>National Cancer Institute, Bethesda, MD</td>
<td>Surgical Oncology Research Training Grant #T32-CA09619</td>
<td>M.H. Torosian (Faculty Preceptor)</td>
<td>$788,919</td>
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<td>1998-2002</td>
<td>National Institutes of Health, Bethesda, MD</td>
<td>&quot;Breast Cancer Patterns of Care and Morbidity&quot;</td>
<td>M.H. Torosian (Co-Investigator)</td>
<td>$2,077,782</td>
</tr>
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</table>
Major Teaching and Clinical Responsibilities at the University of Pennsylvania:

1986 - Clinical Practice of Surgical Oncology -- 12 Months/Year.  
       Surgical Residents & Medical Students

1986 - 1991 Basic Science Lecture Series - Moderator  
       Surgical Residents & Medical Students

1986 - 1993 Lecture, Surgery Core Course (Surgery 200) - Bi-Monthly  
       2nd - 3rd Year Medical Students

1988 - Basic Science Research Mentor, Elective Work - Study Program  
       3rd - 4th Year Undergraduate Students

1991   Surgical Oncology: Advances in Breast & Gastrointestinal Cancers,  
       Course Co-Director

1991 - Surgical Oncology Preceptor, Elective Course (Surgery 321) - 11 Month/Year  
       3rd - 4th Year Medical Students

1993 - Problem-Based Learning Session, Core Course (Surgery 200) Bi-Monthly  
       2nd - 3rd Year Medical Students

1996 - Surgical Oncology Research, Elective Course (Surgery 800),  
       4th Year Medical Students
ORIGINAL PAPERS:


62. **Torosian MH:** Feeding the tumor; fact or fiction, good or bad. Cancer Prevention International (In Press).


BOOKS - EDITOR


BOOK AWARD

1. Doody's Book Award -- 1 of 250 Best Health Sciences Books Published in 1996: Nutrition for the Hospitalized Patient: Basic Science and Principles of Practice. Editor -- Torosian, MH.

VIDEO/CINE CLINICS:

1. Vascular Access Techniques, By: J.M. Daly and M.H. Torosian
   American College of Surgeons, 1993
BOOK CHAPTERS:


