GRANT NUMBER DAMD17-94-J-4177

TITLE: Breast Cancer Resource for Research and Banking, with Emphasis on Early Tumors and Precursor Lesions

PRINCIPAL INVESTIGATOR: Helen Feiner, M.D.

CONTRACTING ORGANIZATION: New York University Medical Center
New York, New York 10016

REPORT DATE: October 1998

TYPE OF REPORT: Final

PREPARED FOR: Commander
U.S. Army Medical Research and Material Command
Fort Detrick, Frederick, 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 1998	Final (1 Dec 94 - 1 Sep 98)

4. TITLE AND SUBTITLE
Breast Cancer Resource for Research and Banking, with Emphasis on Early Tumors and Precursor Lesions

5. FUNDING NUMBERS
DAMD17-94-J-4177

6. AUTHOR(S)
Helen Feiner, M.D.

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)
New York University Medical Center
New York, New York 10016

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
Approved for public release; distribution unlimited

12b. DISTRIBUTION CODE

13. ABSTRACT *(Maximum 200*

The Breast Cancer Resource for Research and Banking has accrued breast cells or tissues from 1,467 patients during the grant period (3 year grant with 9 month extension without additional funding). The emphasis of this project has been on the collection of microscopic at risk and precursor lesions as imprints/scrapes. Additionally, throughout the grant period, all invasive carcinomas with available tissue have been accrued, since most investigators who have requested samples have requested frozen pieces of tumor tissue paired with normal tissue from the same patient, i.e. their interest has been in established carcinomas and not in precursor lesions. During the last year we also filled requests for specimens for microdissection. Over the entire grant period 26 investigator requests for tissue have been filled. At termination of the grant the Breast Cancer Resource was transferred to the auspices of the Resource for Tumor Tissue and Data of the NYU Kaplan Comprehensive Cancer Center.

14. SUBJECT TERMS	15. NUMBER OF PAGES
Breast Cancer	34
	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	Unlimited

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.

NA Citations of commercial organizations and trade names in this report do not constitute an official Department of 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 Resources, national Research Council (NIH Publication No. 86-23, Revised 1985).

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

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

____ In the conduct of research utilizing recombinant DNA, the investigator(s) adhered to the NIH Guidelines for Research Involving Recombinant DNA Molecules.

NA In the conduct of research involving hazardous organisms, the investigator(s) adhered to the CDC-NIH Guide for Biosafety in Microbiological and Biomedical Laboratories.

Helen Fenus MD 9/28/98

PI - Signature Date

TABLE OF CONTENTS

INTRODUCTION

Basic, clinical, and translational research on breast cancer in the United States has been stimulated in recent years by increased government and private funding. Translational research, in particular, requires the availability of human breast cancer tissue, as well as breast tissue with "precursor" and "at risk" lesions. At risk lesions are the proliferative and atypical proliferative components of fibrocystic change, and the precursor lesion is carcinoma in situ. These lesions have been defined histologically and their roles in breast carcinogenesis have been validated epidemiologically. This grant was funded in the category of "Infrastructure Enhancement" specifically to make breast cancer tissue, precursor, and at risk lesions available to investigators in the field of human breast carcinogenesis.

METHODS

Clinical cancers, that is invasive carcinomas that have resulted in a palpable mass lesion, were banked in standard fashion as snap-frozen pieces of tissue, together with pieces of non-neoplastic breast tissue from the same patient, and a portion of lymph node when available.

Collecting at risk and precursor lesions of breast cancer, which are almost invariably microscopic, is difficult, firstly because the lesions are so small, and secondly because good medical practice requires that all the tissue excised be subjected to routine histopathologic examination in order to properly classify the lesion.

Accordingly, in years 1 to 3 of the grant we collected at risk and precursor lesions of breast cancer as slide imprints/scrapes prepared from excised breast tissue prior to histopathologic examination. By the end of year two 782 imprint samples had been collected from mammographically detected (non-palpable) lesions. These covered the spectrum of fibrocystic change (non-proliferative, proliferative, and proliferative with atypia), as well as ductal carcinoma in situ and lobular carcinoma in situ. It was disappointing that despite considerable effort to publicize this collection of material, investigators did not request it. The reasons were twofold. Firstly, most basic scientists are unfamiliar with the histopathologically defined at risk and precursor lesions of breast cancer; they are more interested in established cancers (mass lesions). Secondly, the samples are small and comprised of mixtures of cells (of necessity stromal cells and lymphohistiocytic cells are admixed with lesional cells in imprint/aspirate specimens). At about the same time the technique of microdissection was evolving, and provided an alternate method for acquiring such lesions for research purposes. Accordingly in year 3 and in the extension period we have been supplying investigators with material prepared for microdissection. This technique allows one to obtain pure specimens of microscopic precursor and at risk lesions, from either fixed paraffin embedded tissue (for DNA - PCR studies) or from frozen sections (for RNA based studies). Specimens obtained by microdissection are superior to imprints and aspirates inasmuch as the histologic context from which samples are obtained can be documented, and the samples are pure. Because of the aforesaid, in year 3 we turned our efforts away from imprints and toward providing samples for microdissection.

Use of the Resource has been stimulated by the award of 16 pilot projects from developmental funds from the Kaplan Comprehensive Cancer Center's NCI Breast Cancer Program Grant during the 1995-1998 period.

Outside NYU, the Resource has been included in the Breast Cancer Specimen and Data Information System, a collaborative project sponsored by the National Action Plan for Breast Cancer Biologic Resources Banks Working Group and the NCI. The DOD Breast Cancer Research Program "Era of Hope" in Washington D.C. in October/November 1997 provided another forum for publicizing the Resource.

To obtain feedback on the satisfaction of investigators with the material sent to them, two contacts are made with each recipient. The first is to determine the state of material given or shipped and occurs within a day or two of shipping. The second contact is made 6 - 18 months later to determine the level of satisfaction in terms of results obtained. User files are maintained for each recipient of samples. User records are initiated with "Investigator Request" forms (Appendix 1).

To obtain information on the quality and durability of banked tissues and cells, specimens obtained in 1995, 1996 and 1997 have been subjected to a variety of analyses. These analyses were immunohistochemistry, immunofluorescence microscopy, fluorescence in situ hybridization, and RT-PCR. Analyses were done in various laboratories at NYU that have expertise in these assays. Records are maintained on "Evaluation of Banked Material" forms (Appendix 2).

The Resource technician has also culled the NYU departmental records retrospectively so that all patients with mammographically detected lesions from 1991-1994 have been entered into the database. Even though no fresh samples (imprints/aspirates) are available in these cases, the ability to microdissect the archival samples has made them a valuable addition to the Resource.

RESULTS

The numbers of the various types of breast tissue samples that have been banked and entered into our database during each grant year, as well as the cumulative numbers of samples for the entire collection period are shown in Tables 1 to 4. Tables 1 and 2 use the format of previous annual reports. Tables 3 and 4 show data for all four years.

In Table 1 the breakdown is by type of samples available. In Table 2 the breakdown is by type of lesion as defined histopathologically. Total number of samples in Table 1 exceeds total number of cases in Table 2 because some cases (patients) generated more than one sample type.

TABLE 1

BANKED CASES OF BREAST CANCER AND PRECURSOR LESIONS AT NYU MEDICAL CENTER BY SAMPLE TYPES

	Grant Year #4 12/97 - 8/98	4 Yr. Cumulative 12/94 - 8/98
Imprints/scrapes	0	633
Aspirated cells	115	642
Snap frozen tissue fragments*	69	757
TOTAL	184	2,032

*includes 308 paired samples of breast cancers with normal tissue.

TABLE 2

BANKED CASES OF BREAST CANCER AND PRECURSOR LESIONS AT NYU MEDICAL CENTER BY HISTOPATHOLOGIC DIAGNOSIS

	Grant Year #4 12/97 - 8/98	4 Yr.Cumulative 12/94 - 8/98
Invasive ductal carcinoma	67	383
Invasive lobular carcinoma	12	59
Ductal carcinoma in situ*	7	151
Lobular carcinoma in situ*	0	46
Secondary carcinoma, lymph node	27	110
Lymph node without tumor	0	81
Fibrocystic change, non-proliferative	1	190
Fibrocystic change, proliferative**	1	215
Fibrocystic change, proliferative with atypia**	0	80
Other (mostly fibroadenoma)	13	153
TOTAL	128	1,468

*precursor lesion **at risk lesion

Table 5 indicates the number of patients from whom samples were obtained during years 1 and 3 and the extension period, and during the entire grant period. Table 6 indicates the numbers of requests for specimens that have been filled over similar time periods.

TABLE 3

SPECIMENS BY SAMPLE TYPE

	1995	1996	1997	1998	Total
Imprints/TP	367	415	102	0	633
Aspirates	149	219	159	115	642
Tissue	233	199	256	69	757
TOTAL	749	833	517	184	2,032

TABLE 4

SPECIMENS BY HISTOPATHOLOGIC DIAGNOSIS

	1995	1996	1997	1998	Total
Invasive ductal carcinoma	118	112	86	67	383
Invasive lobular carcinoma	16	20	11	12	59
In situ ductal	55	50	39	7	151
In situ lobular	11	25	10	0	46
Secondary carcinoma	25	23	35	27	110
FCD - proliferative	86	92	90	1	269
FCD - non-proliferative	71	107	11	1	190
Other	48	104	97	13	262
TOTAL	430	533	379	128	1,470

TABLE 5

NUMBER OF PATIENTS WITH BANKED SAMPLES

Grant Year #1	Grant Year #2	Grant Year #3	Grant Year #4	4 Yr. Cumulative
12/94 - 11/95	12/95 - 11/96	12/96 - 11/97	12/97 - 8/98	12/94 - 8/98
430	537	365	135	1,467

TABLE 6

REQUESTS FOR SPECIMENS FILLED

	Grant Yr. #1 12/94 - 11/95	Grant Yr. #2 12/95 - 11/96	Grant Yr. #3 12/96 - 11/97	Grant Yr. #4 12/97 - 8/98	4 Yr. Cumulative 12/94 - 8/98
Imprints/scrapes	1	1	0	0	2
Frozen tissue	2	4	9	4	19
Tissue for microdissection	0	0	2	3	5
TOTAL	3	5	11	7	26

As shown in Table 3, we reduced the numbers of imprint/scrape samples collected in years 3 and 4 of the grant. These represent samples of microscopic lesions, mainly in situ carcinoma and proliferative fibrocystic changes. The reason for this reduced collection is twofold. Firstly, we now have a large collection of these lesions and requests for such samples have been very low. Secondly, the technique of microdissection has been gaining increasing favor as an alternative method for obtaining samples of microscopic lesions. Current amplification techniques allow the analysis of cells from a single microdissected duct or lobule of breast tissue. Microdissection can be done on frozen or on fixed, paraffin embedded tissue. Furthermore, the purity of specimens can be monitored by examination of sections before and after the microdissection is done. The success of this technique may be the reason for the underutilization of our imprint/scrape samples. Prior to the use of microdissection, scrapes/imprints represented the only means for obtaining precancerous and microscopic breast lesions for research purposes. The disadvantages of imprint/scrapes as compared to microdissection relates to the fact that imprint/scrape samples represent mixtures of cells, albeit the lesional cells predominate. In both instances the samples are small, but investigators prefer to use samples of known and verifiable purity.

There have been several opportunities for publicizing the Resource at NYU. It has been written up three times in the Kaplan Comprehensive Cancer Center newsletter, "LAB NOTES". The principal investigator has lectured on the Resource to the Kaplan Comprehensive Cancer Center Core Grant Working Group and at the NYU Breast Cancer Research Program (BCRP). She is also a major participant at the NYU monthly clinical multidisciplinary breast cancer conferences and a member of the Executive Committee of the NYU Breast Center, both of which provide forums for continually updating colleagues on the size of the Resource and the spectrum of available material. Additionally, the Kaplan Comprehensive Cancer Center Breast Cancer Research Program Grant has funded pilot projects for translational research from 1995-1998 generating intramural users (Appendix 3).

Our Internet listing through the National Action Plan has generated 6 outside users of the Resource, one in 1996, three in 1997, and two in 1998.

Based on investigator feedback, our efforts in filling requests for specimens and determining investigator satisfaction with specimens has produced results ranging from good to

excellent. All investigators have been very satisfied with the state in which they have received specimens shipped or delivered to them. Feedback from 1995, 1996, and early 1997 recipients indicates that the material was suitable for the research techniques that they used. An example of such feedback and publications referring to the Resource and its funding source are shown in Appendix 1.

Several slide-based techniques performed in the principal investigator's department and elsewhere in the Medical Center produced good results of immunohistochemistry (Appendix 2) and immunofluorescence microscopy on archived samples. Fluorescent in situ hybridization (Appendix 2) results have been excellent on 1997 samples, and good on 1995 and 1996 samples.

At termination of the grant the Breast Tissue Resource is being transferred to the auspices of the Resource for Tumor Tissue and Data of the Kaplan Comprehensive Cancer Center. Thus, the Resource technician, materials, and database will remain available. Collection of samples will continue and the materials collected will remain available to investigators.

CONCLUSIONS

The Resource has acquired 2,032 specimens from 1,467 patients.

Requests for snap frozen samples of established breast cancers, matched with normal tissue from the same patient are the most frequent requests received.

Sample preservation is good.

We have met investigator's needs in all instances.

The Resource has provided the principal investigator with outstanding opportunities for ongoing collaboration in various aspects of breast cancer research (1-13).

REFERENCES

1. Moezzi, M., Melamed, J., Vamvakas, E., Inghirami, G., Mitnick, J., Quish, A., Bose, S., Roses, D., Harris, M., and Feiner, H. Morphologic and biologic characteristics of mammogram-detected invasive breast cancer. Human Pathology, 27:944-948, 1996.

2. Mitnick, J.S., Vazquez, M.F., Feiner, H.D., Pressman, P., and Roses, D.F. Mammographically detected breast lesions: Clinical importance of cytologic atypia in stereotaxic fine-needle aspiration biopsy samples. Radiology, 198:319-322, 1996.

3. Illei, P., Feiner, H.,Yin, Z., Chan, W., and Perle, M. Interphase cytogenetics of chromosomes 7,18, and X in proliferative fibrocystic change and ductal carcinoma in situ of breast: A study of 30 cases. American Association for Cancer Research, 1996. AACR Proceedings, 37:111, 1996.

4. Liu, J., Yin, Z., Liu, W., Yee, H., Wieczorek, R., Inghirami, G., and Feiner, H. Evaluation of BRCA1 in human breast cancer. Modern Pathology, 10, 1997.

5. Feinder, H.D. and Delgado, Y. Breast Cancer Resource for Research and Banking with Emphasis on Early Lesions. U.S. Department of Defense-sponsored meeting "An Era of Hope", Washington, D.C., 1997.

6. Fasano, M., Vamvakas, E., Hwang, D., Inghirami, G., and Feiner, H. Tubular carcinoma of the breast: Morphologic and molecular characteristics. Modern Pathology, 11:19A, 1998.

7. Liu, J., Prolla, G., Yee, H., Feiner, H., and Inghirami, G. A potential human BRCA1 knockout. Modern Pathology, 11:23A, 1998.

8. Illei, P., Feiner, H.D., Symmans, W.F., Mitnick, J., Roses, D.F., and Perle, M.A. Numerical abnormalities of chromosomes 7, 18, and X in precancerous breast disease defined by fluorescent in situ hybridization. The Breast Journal, 4:252, 257, 1998.

9. Mitnick, J.S., Gianutsos, R., Pollack, A.H., Susman, M., Pressman, P.I., Feiner, H.D., and Roses, D.F. Comparative value of mammography, fine needle aspiration biopsy, and core biopsy in the diagnosis of invasive lobular carcinoma. The Breast Journal, 4:75-83, 1998.

10. Lopez, C., Rao, T.D., Feiner, H., Shapiro, R., Marks, J., and Frey, A.B. Repression of Interleukin-2 mRNA translation in primary human breast carcinoma tumor infiltrating lymphocytes. (Submitted)

11. Oddoux, C., Robson, M., Clayton, C.M., Delgado, Y., Jhanwar, S., Tan, L., Offit, K., Feiner, H., and Ostrer, H. Differences in heritable susceptibility to invasive and non-invasive breast cancer among Ashkenazi Jews. (Submitted)

12. Cui, X., Li, H., Liu, J., and Feiner, H. Genetic analysis of tubular carcinoma using microdissected samples. (Submitted)

13. Cui, X., Feiner, H., and Li, H. Development of tubular carcinoma of the breast may involve a distinct gene set. (Submitted)

LIST OF PERSONNEL RECEIVING PAY FROM THIS EFFORT

Helen Feiner, M.D.
Jaishree Jagirdar, M.D.
Ms. Yara Delgado

Feiner, Helen, D.
DAMDM17-94-J-4177

APPENDIX 1

NYU Medical Center

Request for Tissue/Cells
NYU Breast Cancer Resource
Director: Helen D. Feiner, M.D.
560 First Ave. NY, NY 10016
Tel (212) 263-8826
Fax (212) 263-7916

Name:	Dr. CHARLES CARMECI/ D. THOMPSON Ph.D
Title:	MD/Ph.D
Address:	STANFORD UNIVERSITY. DEPTO OF SURGERY MSLS P229. 1201 WELCH RD STANFORD, CA 94305
Phone:	(415) 725-1671 (415) 498-5510
Fax:	(415) 725-8762
e-mail:	--
Grant Support:	YES

Material Requested: BREAST TISSUE IN VIAL
CONFIRMED ER+ AND ER-
Date Shipped: 8-5-96 4-21-97
Date Received: NEXT DAY
State of Specimen on receipt: GOOD

Brief Summary of intended use:
(Use additional page if necessary)
TO IDENTIFY AND CHARACTERIZE GENES THAT ARE COORDINATELY EXPRESSED WITH ER AND DETERMINE THEIR INFLUENCE ON BREAST CANCER PROTOTYPE.

Charles Carmeci, MD
Stanford University
Dept of Surgical Oncology
MSLS P229
1201 Welch Rd
Stanford, CA 94305

Ph (415) 725-1671

8/16/96

Helen Feiner, MD
Dept of Surgical Pathology
NYU Medical Center
560 First Avenue
New York, NY 10016

Dear Dr. Feiner,

Thank you for the primary breast cancer specimens. They arrived in excellent condition.

We have recently isolated and partially characterized several genes from breast cancer cell lines which are coordinately expressed with the gene for estrogen receptor. We feel that this set of genes plays a critical role in determining the differing phenotypes between ER positive and ER negative carcinomas. Using Northern blots from the samples which you have provided, we aim to determine the expression of these genes in primary tumors. The NIH has provided funding for this project (Grant #: NIH/NRSA#1F32CA69715-01A1 PI: Ronald Weigel, MD, PhD).

Thank you for providing such a valuable resource.

Charles Carmeci, MD

By phone from Dr. Carmeci 5/5/97
- yield of 20-30 ng total RNA / spec.
- Not great for Northerns
- RT PCR yield good of all ER+ samples and most ER- samples. Low yield from normal breast tissue
HF

NEW YORK UNIVERSITY MEDICAL CENTER
Anatomic Pathology, Room 461
560 First Avenue
New York, N.Y. 10016

FAX

Date: 1/31/97

Number of pages including cover sheet: 1

TO: DR CHARLES CARMECI FROM: DR HELEN FEINER

Phone: Phone: (212) 263-5470

Fax: 415 723-8762. Fax: (212) 263-7916

As discussed, information to add to your publication(s):

REMARKS: ☐ Urgent ☐ For your review ☐ Reply ASAP ☐ Comment

Acknowledgement: Breast cancer tissue was obtained from the Breast Cancer Resource of the Department of Pathology, N.Y.U. Medical Center, Dr Helen Feiner, Director. The Resource is funded by The Department of the Army, a Grant DAMD 17-94-J-4177

NYU Medical Center

560 First Avenue, New York, N.Y. 10016
Cable Address: NYUMEDIC

Department of Pathology

(212) 263-

8/12/96

Dr. Charles Carmeci
Dept of Surgery
Standford University
1201 Welch Rd
Stanford, CA 94305

Dear Dr. Caremeci:

This is to confirm that on August 5, 1996 we shipped you 18 frozens breast tissue specimens, as follows: 8 estrogen receptor positive carcinomas
8 estrogen receptor negative carcinomas
2 non tumor breast tissue

Please let us know the state in which the specimens were received, a brief statement of the intended use, and how well the material served your purposes.

Many thanks in advance for this important feedback.

Yours sincerely,

Helen Feiner

Helen Feiner, M.D.
Director, Anatomic Pathology
Director, Breast Cancer Resource
PH (212) 263-8826 FAX (212) 263-7916

cc: Rita Demopoulos, M.D.

STANFORD UNIVERSITY SCHOOL OF MEDICINE

DEPARTMENT OF SURGERY
MEDICAL SCHOOL OFFICE BLDG. (MSOB),
SUITE X300
STANFORD, CALIFORNIA

Ph: ~~415/725-7280~~ FAX No: 415/~~725-3918~~ 725-8762

TO: Helen Feiner

PH. NO: (212)263-5470

FAX NO: (212)263-7916

NO. OF PGS: 3 (Including this page)

FROM: Devon Thompson

PH. NO: (415)498-5510

COMMENTS:

If you have an email address I could forward you the list of tumours so you would have it on your computer

STANFORD UNIVERSITY
Department of Surgery

1201 Welch Road
MSLS Building, Room P228
Stanford, CA 94305-5486
phone: (415) 498-5510 or (415) 725-1671
fax: (415) 725-8762

Devon A. Thompson, Ph.D.
Postoral Doctoral Fellow
devont@leland.stanford.edu

Helen Feiner, M.D.
Department of Surgical Pathology
N.Y.U. Medical Center
560 First Avenue,
New York, NY 10016

July 9th 1997

Dear Dr. Feiner,

I have been working in collaboration with Charles Carmeci, M.D., with whom you have had previous discussions. We have received 31 breast tumour and 4 normal breast samples from your Breast Cancer Tissue Bank. This source has been invaluable to us. We have used these samples to extract RNA and then perform RT-PCR to detect several different genes. At this juncture it would be extremely useful if we could obtain any information you have in your files pertaining to the specific tumours that you have provided to us. Information such as, histological grade, the method(s) used to establish the estrogen receptor phenotype and quantitative values for the ER levels determined. Following I have listed our ID number and your ID number for each of the tumours that we have received.

I will be away on vacation from July 12th until July 26th. You can contact me by e-mail devont@leland.stanford.edu, by phone (415) 498-5510, or fax (415) 725-8762 after this date. Thank you for your help with this matter.
Sincerely,

Devon A. Thompson, Ph.D.
devont@leland.stanford.edu

Stanford ID	NYU Tumour ID	Current Information
1	s95 10787	ER + tumour
2	s95 11102	ER + tumour
3	s95 11319	ER + tumour
4	s95 12182	ER + tumour
5	s95 17621	ER + tumour
6	s96 2526	ER + tumour
7	s95 8934	normal breast
8	s95 12034	normal breast
9	s95 14255	ER - tumour
10	s95 14788	ER - tumour
11	s95 21162	ER - tumour
12	s96 1236	ER - tumour
13	s96 2129	ER - tumour
14	s96 2423	ER - tumour
15	s95 10239	ER + tumour
16	s95 12254	ER - tumour
17	s96 12828	ER - tumour
19	s97 4250	ER + tumour
20	s97 4288	ER + tumour
21	s96 20823	ER + tumour
22	s97 2396	ER + tumour
23	s97 4379	ER + tumour
24	s97 3778	ER + tumour
25	s96 20371	ER + tumour
26	s97 4926	ER + tumour
27	s97 596	ER - tumour
28	s96 19122	ER - tumour
29	s97 528	ER - tumour
30	s96 12363	ER - tumour
31	s96 14116	ER - tumour
32	s96 20358	ER - tumour
33	s96 15075	ER - tumour
34	s97 1647	normal breast
35	s97 1792	normal breast

Subject: NYU breast cancer samples.
From: Helen Feiner 7/24/97 12:50 PM
To: devont@leland.stanford.edu at PMDF

Dear Dr Thpmpson,

I have sent you, by mail, two reports from each of the patients listed in your communication of July 9th. One is the surgical pathology report from which you can derive a histologic grade. The most common grading system utilizes architectural grade + nuclear grade + mitotic rate. The second report is from our molecular pathology lab from which you can obtain the estrogen receptor quantitative values. Let me know if you need help with any of these data.

Method used to obtain ER phenotype: Indirect immunoperoxidase technique. Estrogen receptor antibody is obtained from AMAC (clone ER1D5, Westbrook, ME) and Novo Castra (clone 6F11, distributed by Vector, Burlingame, CA). Secondary antibody is horse anti mouse IgG. A standard avidin-biotin-peroxidase technique is used on formalin fixed, paraffin embedded tissue sections. Antibody expression is evaluated in 10 40x fields in a CAS Image Analyzer. Result is expressed as percent positive nuclear area.

Helen Feiner M.D.

STANFORD UNIVERSITY SCHOOL OF MEDICINE
STANFORD, CALIFORNIA 94305-5486

Devon A. Thompson, Ph.D.
Postdoctoral Fellow
Department of Surgery
Division of Surgical Oncology

MSLS Building, Room P228
1201 Welch Road
(650) 498-5510
(650) 723-8762 (fax)
email: devont@leland.stanford.edu

Helen Feiner, M.D.
Department of Surgical Pathology
N.Y.U. Medical Center
560 First Avenue,
New York, NY 10016

August 25 1998

Dear Dr. Feiner,

Thank you for the information pertaining to the breakdown of race status, with regard to patients from whom the breast tumour specimens are obtained. Enclosed are re-prints from some papers in which we have used the tumour specimens that you provided. These frozen tumours have been invaluable to us in extrapolating our findings in breast cancer cell lines to breast tumour biology. We hope to continue using the Breast Cancer Resource of the Department of Pathology, New York University Medical Center, to obtain breast cancer samples.

Sincerely,

Devon A. Thompson, Ph.D.

PUBLICATIONS THAT ACKNOWLEDGE THE RESOURCE :

Eur. J. Biochem. 252, 169–177 (1998)
© FEBS 1998

Characterization of a gene that is inversely correlated with estrogen receptor expression (ICERE-1) in breast carcinomas

Devon A. THOMPSON and Ronald J. WEIGEL
Department of Surgery, Stanford University, Stanford CA, USA

(Received 22 September/10 December 1997) – EJB 97 1350/1

1116–1123 *Nucleic Acids Research, 1998, Vol. 26, No. 4*

Differential screening and suppression subtractive hybridization identified genes differentially expressed in an estrogen receptor-positive breast carcinoma cell line

Wayne W. Kuang, Devon A. Thompson, Renee V. Hoch and Ronald J. Weigel*

Department of Surgery, Stanford University, Stanford, CA 94305, USA

Received June 10, 1997; Revised and Accepted December 18, 1997

DDBJ/EMBL/GenBank accession no. AF007170

GENOMICS **45**, 607–617 (1997)
ARTICLE NO. GE974972

Identification of a Gene (GPR30) with Homology to the G-Protein-Coupled Receptor Superfamily Associated with Estrogen Receptor Expression in Breast Cancer

Charles Carmeci,* Devon A. Thompson,* Huijun Z. Ring,† Uta Francke,†‡ and Ronald J. Weigel*,[1]

*Department of Surgery, †Department of Genetics, and ‡Howard Hughes Medical Institute, Stanford University, Stanford, California 94305

Received April 4, 1997; revised August 11, 1997

NYU Medical Center

Request for Tissue/Cells
NYU Breast Cancer Resource
Director: Helen D. Feiner, M.D.
560 First Ave. NY, NY 10016
Tel (212) 263-8826
Fax (212) 263-7916

Name:	Dr. KEN TAKASHITA
Title:	ASSISTANT PROFESSOR
Address:	NYU MEDICAL CENTER DEPT. OF HEMATOLOGY
Phone:	(212) 263-5465
Fax:	(212) 263-8444
e-mail:	--
Grant Support:	YES
Material Requested:	FROZEN SECTIONS OF METASTATIC BREAST CANCER. Date Shipped: 8-15-97 Date Received: SAME DAY State of Specimen on receipt: GOOD
Brief Summary of intended use:	(Use additional page if necessary) TO PERFORM IN SITY HYBRIDIZATION AND IMMUNOHISTOCHEMISTRY, IN ORDER TO DETERMINE WHETHER THE DECREASED EXPRESSION OF RAR-ALPHA RETINOIC ACID RECEPTOR EXPRESSION SEEN IN METASTATIC BREAST CA. IS DUE TO A TRANSCRIPTIONAL DEFECT OR A TRANSLATIONAL DEFECT.

NYU Medical Center

12/9/97
Per Dr Takashita & confirmed by H.F. on slide review.
Good signal in IHC
x 1 slide

Hematology Division, Department of Medicine
New York University Medical Center
550 First Avenue, New York, N.Y. 10016 U.S.A.

e-mail takeshtk@is.nyu.edu
Tel 1-212-263-5465, Fax 1-212-263-8444

August 15, 1997

Dr. Helen Feiner
Department of Pathology
Breast Cancer Archives

Dear Dr. Feiner:

I am writing to notify you that we have requested and received from Yara Delgado of the breast tumor registry frozen sections of lymph nodes containing known breast cancer metastasis from 7 different patients. We received 6 slides for each patient.

These sections will be used to perform in situ hybridization and immunohistochemistry. The objective of this experiment is to determine whether the decreased expression of RAR-alpha retinoic acid receptor expression seen in metastatic breast cancer is due to a transcriptional defect or a translational defect.

We are grateful for your assistance in our studies. Please contact me if you have any questions.

Sincerely yours,

Ken Takeshita, M.D.
Assistant Professor of Medicine

Feiner, Helen, D.
DAMDM17-94-J-4177

APPENDIX 2

NYU Medical Center

NYU BREAST CANCER RESOURCE FOR RESEARCH AND BANKING

Phone: (212) 263 8826-8079
Fax #: (212) 263 7916

RECORD OF EVALUATION OF BANKED MATERIAL:

Type of Specimen: Imprint ✓ Frozen Tissue _____

Date of evaluation: 9/4/97

Duration in freezer: 2 years

Type of evaluation: IMMUNOHISTOCHEMISTRY

Results: Excellent

Entered by: HELEN FEINER MD

Signature and date: [signature] 9/13/97

IHC#

IMMUNOHISTOCHEMISTRY

* Place completed form in Tisch-379 (ext 8922)

RESIDENT/ ATTENDING Dr. Fenoglio

PATIENT Anonymous SURG PATH#: ~~98-~~ 98-16437 Block __ *
(*only single block/case will be stained - except by special request)

DATE 5/12/98 SITE Breast SPECIMEN: ~~biopsy/ major~~ Implant

DIAGNOSTIC ISSUE None / QC material

ANTIBODIES : CIRCLE (if limited tissue, number antibodies according to priority, and request "numbered PLL" slides under special requests)

LEU- M1	Calretinin	CK19	GFAP	B72.3	Adenovirus
Muscle Specific ACTIN	(CAM 5.2 (CK))	NSE	*HCG	PSA	*HBsAg
DESMIN	AE1/AE3	CHRO	PLAP	PAP	*HBcAg
SMA	EMA	*SYN	*AFP	FVIII	CMV
VIMENTIN	34BE12	*CALCITON	pCEA	CD34	*HSV
*S-100	CK7	THYRO	mCEA	CD 68	ER/PR
HMB45	CK20	*MYOGLOBIN	(LCA)	BerEP4	Brst-2

PLL SLIDES REQUESTED _____ (circle # if ordered on gross sheet)

SPECIAL REQUESTS:(CIRCLE): RUSH / USE H&E SECTION / _____

RCVD_____ STAINED _____ SIGNED OUT_____ TURNAROUND _____

SPECIAL PROCESSING _____

IHC INTERPRETATION: NEGATIVE CONTROL- ⊖ POSITIVE CONTROLS - ⊕

CAM 5.2 - 3+

LCA - 2-3+

remedial step(s): result- Conclusion-

ABBREVIATIONS AND CLONE # * = POLYCLONAL ANTIBODY

SMA= SMOOTH MUSCLE ACTIN (*1A4*), MUSCLE SPECIFIC ACTIN (*HHF-35*), AE1/AE3 & CAM 5.2= LOW MOL WT KERATINS, 34BE12= HIGH MOL WT KERATIN, CALC= CALCITONIN, THYRO= THYROGLOBULIN (*Tg6*), PSAP= PR ACID PHOS (*PASE/4LT*), PSA= PROSATE SPECIFIC ANTIGEN (*EA-PR8*), FVIII=FACTOR VIII (*F8/86*), SYN= SYNA CHRO= CHROMOGRANIN (*tk2H10*), CMV= CYTOMEGALOVIRUS (*DDG9, CCH2*), DESMIN (*D33*), VIMENTIN (*V9*), LCA= LYMPHOCYTE COMMON ANTIGEN *2B11*), CEA= (*A5B7*), EMA= (*E29*), AFP= ALPHAFETOPROTEIN (*M1A1301*), HSV= HERPES. PLAP= PLACENTAL A PHOSPHATASE (*8B6*)

HCG= HUMAN CHORIONIC GONADOTROPHIN, HBsAg =HEPATITIS B SURFACE Ag, HBcAg=HEPATITIS B CORE Ag CD34=(*QBEND10*.), CD68=(*KP1*). CK7= *OV-TL 12/30*, CK20= *IT-KS20.8*

Histology: Date/time submitted date/time cut

NYU Medical Center

NYU BREAST CANCER RESOURCE FOR RESEARCH AND BANKING

Phone: (212) 263 8826-8079
Fax #: (212) 263 7916

RECORD OF EVALUATION OF BANKED MATERIAL:

Type of Specimen: Aspirated Cells _____ Frozen Tissue ✓
Imprint _____

Date of evaluation: September/ 97

Duration in freezer: 2 YEARS

Type of evaluation: IMMUMOHISTOCHESMISTRY: MIB-1
p21

Results: SATISFACTORY TO GOOD

Entered by: Dr. H. FEINER

Signature and date: 9/12/97

IHC#

IMMUNOHISTOCHEMISTRY

* *Place completed form in Tisch-379 (ext 8922)*

RESIDENT/ ATTENDING HF Eme

PATIENT _____ SURG PATH#: ~~98-~~ 95-11102 Block

(*only single block/case will be stained - except by special request)

DATE 9/5/97 SITE Breast SPECIMEN: biopsy/ major

DIAGNOSTIC ISSUE QA material - (FS)

ANTIBODIES : CIRCLE (if limited tissue, number antibodies according to priority, and request "numbered PLL" slides under special requests)

LEU- M1	Calretinin	CK19	GFAP	B72.3	Adenovirus
Muscle Specific ACTIN	CAM 5.2 (CK)	NSE	*HCG	PSA	*HBsAg
DESMIN	AE1/AE3	CHRO	PLAP	PAP	*HBcAg
SMA	EMA	*SYN	*AFP	FVIII	CMV
VIMENTIN	34BE12	*CALCITON	pCEA	CD34	*HSV
*S-100	(CK7)	THYRO	mCEA	CD 68	ER/PR
HMB45	CK20	*MYOGLOBIN	LCA	BerEP4	Brst-2

PLL SLIDES REQUESTED _____ (circle # if ordered on gross sheet)

SPECIAL REQUESTS:(CIRCLE): RUSH / USE H&E SECTION / _____

RCVD_____STAINED _____SIGNED OUT_____TURNAROUND _____

SPECIAL PROCESSING _____

IHC INTERPRETATION: *NEGATIVE CONTROL-* (-) *POSITIVE CONTROLS -* (+)

p21 - 2+

CK7 - 2+ HF

remedial step(s): *result-* *Conclusion-*

ABBREVIATIONS AND *CLONE #* * = POLYCLONAL ANTIBODY

SMA= SMOOTH MUSCLE ACTIN *(1A4)*, MUSCLE SPECIFIC ACTIN *(HHF-35)*, AE1/AE3 & CAM 5.2= LOW MOL WT KERATINS, 34BE12= HIGH MOL WT KERATIN, CALC= CALCITONIN, THYRO= THYROGLOBULIN *(Tg6)*, PSAP= PROSTATE ACID PHOS *(PASE/4LT)*, PSA= PROSATE SPECIFIC ANTIGEN *(EA-PR8)*, FVIII=FACTOR VIII *(F8/86)*, SYN= SYNAPTOPHYSIN *(SY38)*, CHRO= CHROMOGRANIN *(tk2H10)*, CMV= CYTOMEGALOVIRUS *(DDG9, CCH2)*, DESMIN *(D33)*, VIMENTIN *(V9)*, LCA= LYMPHOCYTE COMMON ANTIGEN *(2B11)*, CEA= *(A5B7)*, EMA= *(E29)*, AFP= ALPHAFETOPROTEIN *(M1A1301)*, HSV= HERPES, PLAP= PLACENTAL ALKALINE PHOSPHATASE *(8B6)*

HCG= HUMAN CHORIONIC GONADOTROPHIN, HBsAg =HEPATITIS B SURFACE Ag, HBcAg=HEPATITIS B CORE Ag

CD34=*(QBEND10.)*, CD68=*(KP1)*, CK7= *OV-TL 12/30*, CK20= *IT-KS20.8*

Histology: Date/time submitted date/time cut

NYU Medical Center

NYU BREAST CANCER RESOURCE FOR RESEARCH AND BANKING

Phone: (212) 263 8826-8079
Fax #: (212) 263 7916

RECORD OF EVALUATION OF BANKED MATERIAL:

Type of Specimen: Imprint ✓ Frozen Tissue _____

Date of evaluation: 9/9/98

Duration in freezer: 2 years

Type of evaluation: FISH

Results: attached

Entered by: AFEINER

Signature and date: [signature] 9/18/98

NYU Medical Center

NEW YORK UNIVERSITY MEDICAL CENTER
CYTOGENETICS LABORATORY

New Bellevue Hospital
Dept. of Pathology/Cytogenetics Lab
Room 4 North 20
27th Street & First Avenue, New York, NY 10016

(212) 263-6454
(212) 562-3496
Fax: (212) 263-7930

PATIENT S96-20371 CASE # Research-S9620371

REFERRAL Dr. H. Feiner DATE COMPLETED 9/09/98

HOSPITAL NYU-Research DATE COLLECTED unknown
DATE RECEIVED 9/03/98

MOLECULAR CYTOGENETIC ANALYSIS

SPECIMEN TYPE Tissue Imprints
QUALITY OF PREPARATION adequate
NO. OF CELLS EXAMINED 50+

CHROMATID BREAKS
CHROMOSOME BREAKS
ANEUPLOID CELLS

INTERPRETATION:

Slides were received from air-dried material described as imprints/scrapes from breast tissue.

Interphase molecular cytogenetic analysis was performed using fluorescent in situ hybridization (FISH) with investigational DNA probes specific for the centromeric region of the X chromosome (Vysis CEP X-alpha probe set). Random sections of the slide were examined by two independent readers. Adequate signal for analysis was seen over the majority of the hybridization area. Results indicated over 85% of cells contained two signals for the X chromosome consistent with two copies of the X. No evidence was seen of X chromosome aneuploidy.

MOLECULAR CYTOGENETIC DIAGNOSIS: nuc ish Xcen(DXZ1x2)

Mary Ann Perle, Ph.D.
Director, Cytogenetics Laboratory

Note: Since this is an in vitro test, accuracy may be limited by technical or cultural artefacts.

APPENDIX 3

BREAST CANCER PILOT PROJECTS AWARDED
1995 - 1998

1995 GRANT YEAR

Pamela Cowin, Ph.D. Assistant Professor Cell Biology	"The Role of Plakoglobin in Breast Cancer" ($30,000)
Xiao-Hong Sun, Ph.D. Assistant Professor Cell Biology	"The Role of ID Proteins in Breast Cancer" ($28,450)
Mary Ann Perle, Ph.D. Assistant Professor Pathology	"Chromosomes 7, 18, 20 and X in Mammogram Detected Atypical Ductal Hyperplasia and Ductal Carcinoma in situ" ($8,950)

1996 GRANT YEAR

Sandra Reynolds, Ph.D. Res. Assistant Professor Dermatology	"Peptide Epitopes Recognized by CD8+ T Cells in Patients with Breast Cancer" ($10,000)
Herbert Samuels, M.D. Professor Medicine	"Retinoid-Regulated Genes and Breast Cancer" ($25,000)
Jan Sap, Ph.D. Assistant Professor Pharmacology	"Receptor Protein Tyrosine Phosphatases and Breast Cancer" ($20,000)
Kenichi Takeshita, M.D. Assistant Professor Medicine	"9-cis Retinoic Acid and Retinoid X Receptor RXR in Breast Cancer" ($20,000)
Stephen Tomlinson, Ph.D. Assistant Professor Pathology	"The Role of Complement Inhibitors in Tumorigenicity" ($10,000)
Stanislav Vukmanovic, MD,PhD Assistant Professor Pathology	"Effector Function of Vaccine Induced CD8+ Cells" ($10,000)

1997 GRANT YEAR

Harry Ostrer, M.D. (P.I.) Professor Pediatrics Ruth Oratz, M.D. (Co-P.I.) Assistant Professor Medicine	"Genetic Susceptibility to Breast Cancer" ($15,000)

W. Fraser Symmans, M.D. (P.I.) Assistant Professor Pathology Matthew Volm, M.D. (Co-P.I.) Instructor Medicine	"A Response Biomarker for Paclitaxel Chemotherapy in Patients with Breast Cancer" ($29,875)
Carolyn Wasserheit, M.D. (P.I.) Assistant Professor Medicine Kenichi Takeshita, M.D. (Co-P.I.) Assistant Professor Medicine	"Biological Correlates of 9-Cis Retinoic Acid and Tamoxifen" ($15,000)

1998 GRANT YEAR

Ruben Abagyan, Ph.D. Associate Professor Biochemistry	"Toward A New Chemotherapy for Breast Cancer: Rational Design of A Retinoid X Receptor-Selective Agonist" ($29,968)
Alan Frey, Ph.D. Assistant Professor Cell Biology	"Translational Arrest of IL-2 mRNA in Human Breast Cancer Tumor Infiltrating Lymphocytes" ($30,000)
Giorgio Inghirami, M.D. Associate Professor Pathology	"Molecular Characterization of BRCA1" ($30,000)
Carole Oddoux, Ph.D. Assistant Professor Pediatrics	"Heritable Susceptibility to Invasive and Non-Invasive Breast Cancer" ($15,000)