AWARD NUMBER: W81XWH-16-1-0045

TITLE: Discoidin Domain Receptors: Novel Targets in Breast Cancer Bone Metastasis

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REPORT DATE: February 2019

TYPE OF REPORT: Annual

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

REPORT DOCUMENTATION PAGE

Form Approved
OMB No. 0704-0188

1. REPORT DATE	2. REPORT TYPE	3. DATES COVERED
FEB 2019	Annual	1 Feb 2018 – 31 Jan 2019

4. TITLE AND SUBTITLE

Discoidin Domain Receptors: Novel Targets in Breast Cancer Bone Metastasis

5a. CONTRACT NUMBER
W81XWH-16-1-0045

5b. GRANT NUMBER
BC150621

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)

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5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

7. PERFORMING ORGANIZATION NAME(S) AND ADDRESS(ES)

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8. PERFORMING ORGANIZATION REPORT NUMBER

9. SPONSORING / MONITORING AGENCY NAME(S) AND ADDRESS(ES)

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

10. SPONSOR/MONITOR'S ACRONYM(S)

11. SPONSOR/MONITOR'S REPORT NUMBER(S)

12. DISTRIBUTION / AVAILABILITY STATEMENT

Approved for Public Release; Distribution Unlimited

13. SUPPLEMENTARY NOTES

14. ABSTRACT Here we report major findings for our project aimed at studying the expression of Discoidin Domain Receptors (DDRs) in breast cancer (BrCa) tissues and their functional contribution to the formation of BrCa bone metastases. We also aim at testing the feasibility of targeting DDRs for the treatment BrCa bone metastases. During the current funding period, we identified and classified pair matched tissues of primary invasive BrCa cases and corresponding bone metastases, from which 12 cases were analyzed for DDR1 expression by immunohistochemistry (IHC). These analyses revealed expression of membranous DDR1 in both primary and metastases of ductal carcinomas. Lobular carcinomas displayed mostly cytoplasmic DDR1 staining in both primary and metastatic tumors. We generated human MDA-MB-231 BrCa cells with ectopic expression of wild type and kinase dead DDR1b and wild type DDR2. The DDR1b-expressing cells were tested in a model of intraosseous tumor growth in mice in a preliminary experiment. The promising results of this study led to a second experiment with a larger group of mice. X-ray analyses suggest that DDR1b may diminish formation of osteolytic lesions. However, confirmation of this potential role of DDR1b in bone metastases awaits analyses of tumor burden and bone response by histomorphometry, which are ongoing. In the next period, we plan to conduct additional IHC studies with the human samples and complete the analyses of the bones from the mice inoculated with the MDA-MB-231 cells..

15. SUBJECT TERMS
Breast cancer, bone metastasis, discoidin domain receptors, kinases, targeted therapies, immunohistochemistry

16. SECURITY CLASSIFICATION OF:			17. LIMITATION OF ABSTRACT	18. NUMBER OF PAGES	19a. NAME OF RESPONSIBLE PERSON
a. REPORT	b. ABSTRACT	c. THIS PAGE			USAMRMC
U	U	U	UU	14	19b. TELEPHONE NUMBER *(include area code)*

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1. INTRODUCTION

Different treatments are currently used to treat bone metastasis, the main cause of morbidity and mortality in patients with advanced breast cancer (BrCa). However, although currently available therapies can be effective to relieve pain, prevent complications, and improve quality of life in these patients, are not curative. The identification of novel molecules involved in the establishment and expansion of BrCa metastatic cells within the bone is, therefore, crucial for the development of new prognostic biomarkers and therapeutic agents to prevent and/or inhibit skeletal metastases. Discoidin domain receptors (DDRs) are expressed in invasive BrCa and represent the only receptor tyrosine kinases (RTKs) that uniquely signal in response to collagen, a major organic component of the bone microenvironment. Based on these facts, the purpose of the research proposed in this application is to test our hypothesis that DDRs mediate the survival of metastatic BrCa cells within the skeletal niche and consequently represent promising targets for intervention in BrCa patients with bone metastasis. The scope of research involves the analysis of DDR expression in primary tumor and bone metastatic tissues from BrCa patients, the evaluation of therapeutic efficacy of DDR inhibition in a preclinical model of intraosseous BrCa growth, and the study of tumor-derived DDRs' role in the regulation of BrCa pro-osteolytic programs using *in vitro* systems.

2. KEYWORDS

Discoidin domain receptors, breast cancer, bone metastasis, receptor tyrosine kinases, collagen, biomarkers, targeted therapy.

3. ACCOMPLISHMENTS

- **What were the major goals of the project?**

Specific Aim 1. To conduct a histopathological analysis of DDR expression in samples of primary BrCa tissues with different subtypes and their matching bone metastasis.

Task 1: Select BrCa tissues for analyses and construct tissue microarrays (TMAs).

Task 2: Analyses of DDR expression.

Specific Aim 2. To evaluate the therapeutic efficacy of DDR inhibition in a preclinical xenograft model of intraosseous BrCa growth.

Task 1: Analyze DDR expression/activation and generate modified BrCa cell lines.

Task 2: Conduct animal studies to evaluate the role of DDRs in intraosseous tumor growth.

Specific Aim 3. To investigate the role of tumor-derived DDRs in regulation of BrCa pro-osteolytic programs in cell culture systems.

Task 1: Evaluate role of DDRs in regulation of pro-osteolytic factors.

Task 2: Conduct *in vitro* osteoclastogenesis studies.

- **What was accomplished under these goals?**

1) Major activities:

Specific Aim 1.

Task 1: Select BrCa tissues (primary and bone metastases) for immunohistochemical (IHC) analyses of DDR expression and **Task 2**: Analyze DDR expression in the tissues.

In the last funding period, as we reported, we identified, classified, and retrieved BrCa cases of primary and metastatic tumors from the University of Michigan surgical pathology files, which were descrbed in **Table I** below:

Case	Type	Age at primary	Age at met	Histological type	Tumor Grade	Site of Bone Met	Type of specimen	
1	Metastasis	42	51	Lobular		paraspinal	bx	po
	Primary			Lobular			exc bx	
	Primary			Lobular			bx	n
2	Metastasis	61	72	Ductal		Bone	BM aspirate	n
	Primary			Ductal	2		ex bx	po
3	Metastasis	72	77	Lobular		Left ileum	bx	
	Primary			Lobular	2		lump	
	Primary			Lobular			bx	pos
4	Metastasis	43	43	Ductal		Left iliac	bx	po
	Primary			Ductal			bx	po
	Primary			Micropapillary	3		mastectomy	
5	Metastasis	62	62	Ductal		Left sacrum	bx	po
	Primary			Ductal	2			Pos
6	Metastasis	74	78	Ductal		R. ilium	bx	po
	Primary			Mucinous	2		ex bx	pos
7	Metastasis	41	67	Ductal	2	Left 5th rib	bx	po
8	Metastasis	49	51			BM	BM bx	po
9	Metastasis	49	73	Ductal		L. femoral head	res	Po
10	Metastasis	58	60	Ductal		T9 vertebrae	bx	Po
	Primary			Ductal	3		Lump	Po
	Primary			Ductal			bx	Po
11	Metastasis	42	44	Ductal		Lumbar vertebrae	bx	Po
	Primary			Ductal	3		Mastectomy	
	Primary			Ductal	3		Lump	
	Primary			Ductal	3		bx	Po
12	Metastasis	38	40	Ductal		R. femoral head	bx and res	po
	Primary			Ductal	3		Lump	
13	Metastasis	49	49			L. distal humerous	bx	
	Primary			Ductal	1		bx	Po
14	Metastasis	44	52	Lobular		Bone	BM bx	
	Primary			Lobular	1		Mast	
15	Metastasis	31	31			bone	BM bx	
	Primary			Ductal	3		bx, mast	Po
16	Metastasis	57	69			R. ilium	bx	
	Primary			Ductal	2		Lump	
17	Metastasis	49	61			R. Ilium	bx	Po
	Primary							
18	Metastasis	65	65	Lobular		R. Ileum	bx	po
	Primary			Lobular	1		bx	po
19	Metastasis	65	65			T10	bx	po
	Primary			Micropapillary	1		lump	po
20	Metastasis	73	73	Lobular		R. Ileum	bx	po
	Primary			Lobular			bx (chest wall)	Po
21	Metastasis	68	75	Lobular		L. iliac	bx	
	Primary			Lobular	2			
22	Metastasis	57	57			R. iliac	bx	Po
23	Metastasis	57	57			R. Iliac	bx	Po
	Primary			Ductal and lobular	1		bx	Po
24	Metastasis	69	69			T9	bx	
	Primary			Ductal and lobular			bx	Po
	Primary			Micropapillary	3		mast	

From these cases, sections on Plus slides were cut from 12 matched primary breast cancers and their corresponding distant metastasis (**Table II**) and processed for IHC analyses at Wayne State University using a highly specific monoclonal antibody that recognizes only human DDR1 (a gift from Dr. Prunotto, Roche). The tissues were processed for DDR1 IHC using a protocol developed in our lab. The stained slides were evaluated blindly to clinical and pathological information. Specifically, we looked at DDR1 level of expression and subcellular localization.

Table II

Case	Cancer type	DDR1 expression M_membranous C_cytoplasm	Grade	Bone met	Type of specimen	ER	PR	HER2 status	Primary size	Lymph node mets	Other mets	Age of primary	Age of met	Survival	Last seen	Slides	Blocks	Number	Notes
SU-14-8532	Lobular	M negative _increased C and N		Left ileum	bx	pos	focally pos	neg			peritoneum	72	77	Deceased	9/19/15	Y	Y	A1	
BE-09-9872	Lobular	M negative _increased C	2		lump				4.3 cm	7 (0.8 cm)						Y	Y	2E-2F, 3B	
SU-15-41418	Ductal	M low		Left iliac	bx	pos (90%)	pos (90%)	equivocal (2+)			ovary	43	43	Alive-on c	Nov-17	Y	Y	A1	Scant
SU-15-38386	Ductal with micropapillary and lobular feat	M positive	3		mastectomy			Neg (by fish)	4.6 cm	16 (1.1cm)						Y	Y	A2, A13	
SU-15-72386	Ductal	M low_ C increased		T9 vertebrae	bx	Pos (99%)	Neg (0%)	Neg (0+)				58	60	live	1-Nov	Y	Y	A1	
SU-14-15876	Ductal	M low_ C increased	3		Lump	Pos (95%)	Pos (5%)	Neg (0+)	0.9 cm	38 (>2cm)						Y	Y	B4, A14	
SU-13-24588	Ductal	M low and negative areas _heterogeneous		R. femoral head	bx and res	pos (90%)	pos (20%)	pos (3+)			Possible lung	38	40	Went to C	2016	Y	Y	B2	
BE-11-28920	Ductal	M low	3		Lump	Pos	Pos	Neg (2+, fish n	2.6, 0.9, 0.2		2					Y	Y	1B, 1E, 7D, 9F	
SU-14-28105		M high		Epidural/T-6	res	pos (95%)	Few week 1.3%	Neg (1+)			Bone, liver, lung, a	41	50	Alive	1-Dec	Y	Y	A3	
BE-05-15918	Ductal	heterog_ M high and low_ C increased	2		lump	Pos	Pos	neg	1.7 cm	2 (0.4cm)						Y	Y	1C, 1F-1G	
Su-14-29413		M high		R. iliac	bx	Pos (80%0	Neg (0%)	Neg (1+)				57	57	Alive	Jun-17	Y	Y	A1	Scant
SU-14-30052	Ductal with lobular features	M high	1		bx	Pos (99%)	Neg (1%)	Equivocal (2+)								Y	Y	A1, B1	
SU-15-43142		M high		T9	bx						Melanoma and me	69	69	Alive	Apr-17	Y	Y	A1	
SU-15-50449	Ductal with lobular features	M high			bx	Pos (99%0	Neg (1%)	Neg (1+)								Y	Y	A1	
SU-14-30538		M low_C increased		Left ilium	Bx	Pos (90%)						59	59	Alive	Nov-17	Y	Y	A1	
SU-14-27290	Lobular	M low_C increased	1		bx	Pos (95%)	Pos (15%)	Neg (1+)	2.6 (imaging	1 (FNA)						Y	Y	A1	
SU-15-32662		negative		bone	BM bx	Pos	Neg	Neg			Bone, liver, peritor	63	63	Deceased	May-17	y	Y	A1	
BE-13-06193	Lobular	negative	2		bx	Pos (88%)	Pos (65%)	Neg (1+)								Y	Y	A1	scant
SU-14-71591	Lobular	M negative _C increased		L1	bx	Pos						57	57	Alive	Nov-17	Y	Y	A1	Super scar
SU-14-65607	Lobular ***NOTE LN DISSECTION***	M negative	1			Pos (98%)	Pos (79%)	Neg (1+)		12 (1.5 cm)						Y	Y	A2	
SU-14-1573	Lobular	M negative		R. ileum	bx	Neg	Neg	Neg (0+)				38	39	Deceased	Mar-14	Y	Y	A1	Scant
BE-13-34846	Lobular	M negative	3		lump	Neg	Neg	Neg (1+)	3.2 cm		9					Y	Y	1I	
SU-13-33783	Lobular	M low _C increased		R. ileum	bx	pos (90%)	neg (0%)	neg			Brain, liver, bone,	73	73	Deceased	Oct-14	Y	Y	A1	
SU-13-27858	Lobular	M low _C low			bx (chest wall)	Pos (30%)	neg (0%)	neg (1+)								Y	Y	A1	

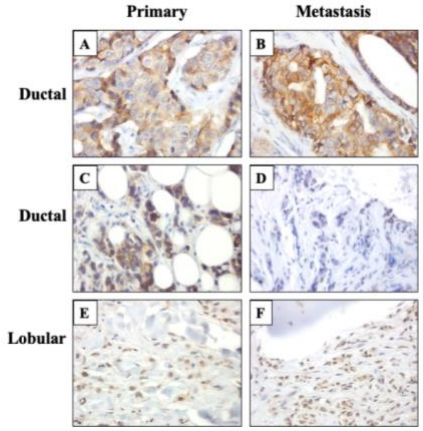

Figure 2. Representative images of DDR1 staining in BrCa samples of primary and metastatic tumors. **A**. Primary IDC with cytoplasmic and some membrane staining (BE-05-15918). **B**. Metastatic IDC with membrane expression (SU14 28105). **C**. IDC with DDR1 at the membrane (SU 15 38386). **D**. Metastasis, mainly negative, in cytoplasm (SU 15 41418). **E**. ILC with cytoplasmic DDR1 (no membrane) (SU 13 27858). F. Metastatic ILC with cytoplasmic DDR1 (no membrane) (SU 13 33783).

Preliminary Results for Task 2:

Of the 12 primary invasive carcinomas, 6 were Invasive Ductal Carcinomas (IDC) and 6 were Invasive Lobular Carcinomas (ILC) (**Table II**). All metastases were to the bone. The IHC staining showed the following findings:

1. The primary and metastatic carcinoma have a similar expression level and pattern of DDR1 expression. Thus, in those samples, levels of DDR1 do not appear to be different between primary and metastatic tumors (**Fig. 2**).

2. Invasive ductal carcinomas (including ductal with lobular features) have frequent positive (or high) membrane expression in both the primary and the metastasis. This is consistent with DDR1 being a cell surface receptor (**Fig. 2A-D**).

3. Invasive lobular carcinomas tend to have low membrane, and increased cytoplasmic expression both in the primary and the metastasis (**Fig. 2E-F**).

4. The normal breast lobules around the invasive carcinoma and the DCIS are positive for DDR 1 in the membrane (data not shown)

5. The pattern of expression in the primary tumors is similar with what we see with E-cadherin (membrane in ductal, reduced or cytoplasmic in lobular carcinomas).

Specific Aim 2.

Task 1: Analyze DDR expression/activation and generate modified BrCa cell lines.

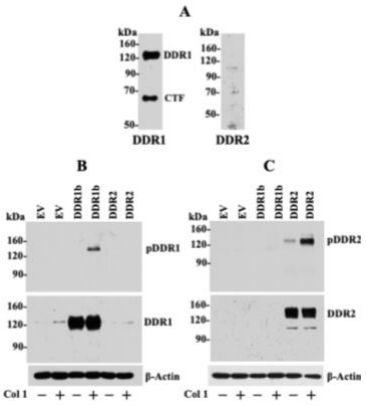

Figure 1. **A**. Expression of DDRs in parental MDA-MB-231 cells (Long exposure blots). **B and C**. Expression and collagen I-dependent activation of DDR1b (A) and DDR2 (B) in stable transfectants of MDA-MB-231 cells. Note that the blot in panel B is a short exposure and thus the endogenous DDR1 is not detectable. EV, control cells transfected with empty vector. pDDR, phosphorylated DDR.

As we reported previously, we characterized expression of DDRs in multiple breast cancer cell lines with the focus on identifying cells with DDR1 expression and capable of growing within bone in mouse models. During this period, we also conducted studies with human MDA-MB-231 cells (referred here as MDA), which are triple-negative breast cancer cells that are known to grow within bone and generate osteolytic lesions. As proposed in the original application, we used this cell line to investigate the role of DDRs in intraosseous tumor growth upon intratibial inoculation of the cells. Human MDA cells, obtained from the American Type Culture Collection, express undetectable levels of DDR2 and low levels of DDR1 (Fig. 1A). Please note, levels of DDR1 are very low in MDA cells but the blots of Fig. 1A are a long-exposure blots, therefore it appears that DDR1 levels are high. Based on these results and the known ability of MDA to grow within bone tissues, we decided to overexpress human DDR1b (wild type and kinase dead) and DDR2 in MDA cells. The kinase dead (KD) DDR1b is a valuable construct to assess the biological effects of DDR1b that are mediated by its kinase activity. To this end, we generated stable transfects and collected pooled populations for analyses of receptor expression and activation. These analyses demonstrated that the pooled populations of stable transfectants expressed the corresponding recombinant proteins (only wild type DDR1 and DDR2 are shown). The wild type DDR1 and DDR2 were activated in response to collagen I, DDR1 ligand (Fig. 1B-C).

Task 2: Conduct animal studies to evaluate the role of DDRs in intraosseous tumor growth.

In the previous funding period (2017-2018) we examined the effect of a DDR1 kinase inhibitor (referred to as Compound A) on intraosseous tumor growth using human BrCa MCF7-Luc cells. Mice were supplemented with estrogen to stimulate cell growth as MCF7 cells are estrogen receptor positive and require estrogen supplementation for growth. As we reported last year, these inhibitor studies with the MCF7-Luc cells were unfortunately unsuccessful. We indicated that although the tibiae of mice inoculated with the MCF7-Luc cells developed intraosseous DDR1-positive tumors infiltrating into the bone marrow, the bones revealed significant areas of dense

bone tissue with constricted bone marrow spaces. This effect, we speculated, possibly limited tumor expansion due to the generation of new bone in the presence of estrogen, a known inducer of bone formation. Importantly, treatment of mice with Compound A showed no evidence of anti-tumor effect when compared to untreated mice, as determined by quantitative imaging of tumors and histomorphometry. From these studies, we were unable to determine whether DDR1 plays a role in intraosseous tumor growth, under the experimental conditions used. Therefore, we decided to focus on the MDA cell system, described in the section Task 1, to address the role of DDRs in intraosseous tumor growth. This was also part of the original application.

In the period of this report (2018-2019), we conducted two major animal studies with the DDR1-overexpressing MDA cell lines. First, we wished to confirmed the ability of the cells to grow within bone. Although it is well established that MDA cells are able to form tumors when inoculated intratibially, it was important to us to confirm this ability with the cells on hand in a limited set of mice.

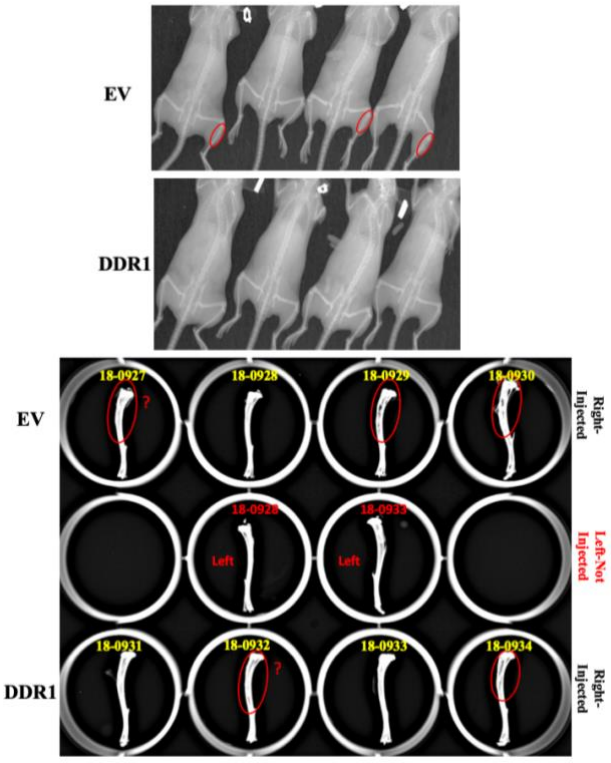

Figure 3. Upper picture: X-Ray imaging of whole mouse body after 4 weeks post-inoculation of MDA-EV and MDA-DDR1 cells. Lower picture: Ex-vivo X-Ray imaging of tibiae isolated from euthanized mice. Red circles show areas of osteolysis. Question marks indicate unclear response.

Experiment #1: We tested the ability of MDA cells, EV (empty vector) and DDR1 expressing cells to develop radiographically detected bone response upon inoculation into the tibiae of female SCID mice. Briefly, $2X10^5$ cells of MDA-EV and MDA-DDR1b cells were inoculated into the tibiae of mice (n=4 per group). After inoculation, the mice were imaged every week by X-ray using a Bruker's In-Vivo Xtreme optical and x-ray small animal imaging system. At 4 weeks post inoculation, based on the radiographic findings, the mice were euthanized. Tibiae were isolated and subjected to ex-vivo X-ray imaging.

Results: As shown in **Fig. 3**, X-ray imaging showed clear bone osteolysis in 3 out of 4 mice with cells inoculated with MDA-EV cells (Upper picture). In contrast, mice inoculated with MDA-DDR1 cells showed no clear bone response (Middle picture). When tibiae were imaged ex-vivo (Lower picture), presence of osteolytic were readily seen in 2/4 mice inoculated with MDA-EV cells whereas one mice showed unclear response. The MDA-DDR1 bones showed 2/4 osteolytic regions. Based on these results, we concluded that the MDA cell variants obtained are capable of growing within the bone and produced radiographically detectable osteolytic lesions and thus they are appropriate for the conduct of a larger experiments with more mice. Interestingly, these results further suggested that expression of DDR1b in MDA cells may reduce intraosseous tumor growth and/or diminish osteolysis. Thus, DDR1 may elicit

an inhibitory effect on development of osteolytic metastases. However, to test this possibility we conducted Experiment #2.

Experiment # 2: Based on the results of Experiment #1, we designed a second experiment utilizing a larger number of mice as follow: MDA-EV (n=9), MDA-DDR1b WT (n=10), and MDA-DDR1b KD (n=10). In this experiment we also included the inactive KD of DDR1b. Mice were inoculated intratibially with $2X10^5$ cells. As in experiment #1, mice were imaged every week by X-ray. Based on these data, mice were euthanized on week 3. From the results of the X-ray images of whole mice and ex-vivo tibiae, we cannot make an educated conclusion as to the extent of bone osteolysis between the three groups, unfortunately. Therefore, the results of this experiment await the conduct of the histomorphometry analyses, which are time consuming.
For these analyses, ex-vivo tibiae were fixed in 4% paraformaldehyde and imbedded in paraffin blocks. Paraffin sections (5 μm) derived from bone tumors were immunostained with Pan-cytokeratin and counterstained with hematoxylin. The histomorphometry analyses are ongoing, and thus at the time of this submission of the report (Feb 2019), these data are not yet available. Thus, we are unable to provide an assessment of tumor burden and bone response. These histomorphometry analyses, and additional IHC analyses of DDR expression, bone remodeling markers, will be completed in the next months.

Specific Aim 3.

Task 1 and Task 2

Nothing to report. These studies are on hold until we obtain the data from the mouse studies.

2) Specific objectives:

The objectives during the period covered by this report were:

a. Use primary invasive breast carcinomas cases with matching bone metastases for analyses of DDR expression.

b. Generate stable transfectant of MDA-MB-231 cells with recombinant expression of DDRs. Characterize expression, collagen-dependent activation and effect on cell proliferation.

c. Conduct animal studies of intraosseous tumor growth to determine the role of DDRs in the MDA-MB-231 cell system of triple negative BrCa.

3) Significant results or key outcomes:

Specific Aim 1, Tasks 1 and 2:

DDR1 is expressed in primary and bone metastatic lesions of IDC and ILC of BrCa. There is a differential subcellular localization of DDR1 in IDC vs. ILC tumors, with a more prominent membrane expression in IDC than in ILC.

Specific Aim 2, Tasks 1 and 2:

Generated MDA cells with overexpressed wild type DDR1 and DDR2 and kinase dead DDR1. Demonstrated expression and functionality of receptor.

Expression of DDR1 in MDA cells appears to be associated with reduced osteolytic response. However, these preliminary outcome needs careful evaluation by histomorphometry analyses and analyses of bone remodeling markers, which will be completed in the next months.

Specific Aim 3, Task 1 and Task 2:

No outcomes to report.

4) Other achievements:

Nothing to report.

- **What opportunities for training and professional development has the project provided?**

Nothing to report.

- **How were the results disseminated to communities of interest?**

Nothing to report.

- **What do you plan to do during the next reporting period to accomplish the goals?**

This award is under a non-cost extension period, in this period, we plan to perform the following studies, as per the SOW:

Specific Aim 1, Tasks 1 and 2: Conduct the IHC for DDR1 and DDR2 expression in the samples obtained and evaluate the levels and subcellular expression of the receptor and their association with available histopathological and clinical markers. As we indicated, a set of 12 cases was already analyzed for DDR1 expression.

Specific Aim 2.

We plan to conclude the histomorphometry analyses of Experiment #2.

Specific Aim 3.

Task 1 and 2: We will follow with the studies proposed in SOW examining the role of DDR activation on the expression of pro-osteolytic factors in the BrCa cells.

4. IMPACT

- **What was the impact on the development of the principal discipline(s) of the project?**

Nothing to report.

- **What was the impact on other disciplines?**

Nothing to report.

- **What was the impact on technology transfer?**

Nothing to report.

- **What was the impact on society beyond science and technology?**

Nothing to report.

5. CHANGES/PROBLEMS

The studies of Aim 3 have been delayed due to the need to conduct the studies with mice, as described in Aim 2 of the SOW. In the non-cost extension period, we will conduct a limited set of experiments to address the role of DDRs in regulation of osteolytic factors.

Actual or anticipated problems or delays and actions or plans to resolve them

We do not anticipate major problems for the remining of the non-cost extension period.

- **Changes that had a significant impact on expenditures**

Nothing to report.

- **Significant changes in use or care of human subjects, vertebrate animals, biohazards, and/or select agents**

Nothing to report.

- **Significant changes in use or care of human subjects**

Nothing to report.

- **Significant changes in use or care of vertebrate animals.**

Nothing to report.

- **Significant changes in use of biohazards and/or select agents**

Nothing to report.

6. PRODUCTS

- **Publications, conference papers, and presentations**

Nothing to report.

- **Website(s) or other Internet site(s)**

Nothing to report.

- **Technologies or techniques**

Nothing to report.

- **Inventions, patent applications, and/or licenses**

Nothing to report.

- **Other Products**

Nothing to report.

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

- **What individuals have worked on the project? See Note below Table**

Award W81XWH-16-1-0046: Dr. Fridman

Name	Project Role	Nearest Person Months Worked	Contribution to the Project	Funding Support
Rafael Fridman	Initiating PI (9/17-present) Partnering PI (prior to 9/17)	0.72	Design of experiments and data analyses	W81XWH-16-1-0046
Allen Saliganan	Research Assistant	12	Animal studies, tissue processing immunohistochemistry	W81XWH-16-1-0046
Anjum Sohail	Research Scientist	3.0	Animal studies In vitro studies	W81XWH-16-1-0046

Benjamin Wasinski	Research Assistant	4.8	In vitro studies	W81XWH-16-1-0046

Award W81XWH-16-1-0045: Dr. Kim (since 09/17)

Name	Project Role	Nearest Person Months Worked	Contribution to the Project	Funding Support
Hyeong-Reh Kim	Partnering PI (9/17-present)	0.72	Design of experiments and data analyses	W81XWH-16-1-0045
Anjum Sohail	Research Scientist	1.8	In vitro studies	W81XWH-16-1-0045

Subcontract to Award W81XWH-16-1-0045: Dr. Kleer (Co-I, University of Michigan)

Celina Kleer	Co-I	0.6	Pathology analyses and tissue supplies	W81XWH-16-1-0045 (subcontract)
Maria E. Gonzalez	Research Associate	4.8	Processing of pathological tissues	W81XWH-16-1-0045 (subcontract)

- **Has there been a change in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period?**

Rafael Fridman, Initiating PI in this grant:

Nothing to report.

Dr. Hyeong-Reh Kim, Partnering PI

Nothing to report.

Celina Kleer, Co-Investigator in this grant:

Nothing to report.

- **What other organizations were involved as partners?**

Organization Name: Hoffmann-La Roche
Location of organization: Basel, Switzerland
Partner's contribution to the project: Supplied antibodies for DDR1 and a small molecule inhibitor for DDR1.

8. SPECIAL REPORTING REQUIREMENTS

Nothing to report.

9. APPENDICES

Nothing to report.