Award Number: W81XWH-19-1-0729

TITLE: Targeting Fibroblast Growth Factor Receptors in Preclinical Patient-Derived Xenograft Models of CRPC

PRINCIPAL INVESTIGATOR: Mark Labrecque

CONTRACTING ORGANIZATION: University of Washington

SEATTLE WA 98195-0001

REPORT DATE: October 2020

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
Oct 2020	Annual	30 Sept 2019 – 29 Sept 2020

4. TITLE AND SUBTITLE

Targeting Fibroblast Growth Factor Receptors in Preclinical Patient-Derived Xenograft Models of CRPC

5a. CONTRACT NUMBER

5b. GRANT NUMBER
W81XWH-19-1-0729

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)
Mark Labrecque, PhD

E-Mail: mlabrec@uw.edu

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

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

UNIVERSITY OF WASHINGTON
GRANT & CONTRACTS DIVISION
4333 BROOKLYN AVE NE
SEATTLE WA 98195-0001

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

In this reporting period, the didactic coursework and training opportunities that were proposed for this project were completed. However, significant disruptions to research activities were encountered due to the ongoing COVID-19 pandemic response and ACURO approval delays. These factors led to the application and approval of a no cost extension to complete all proposed work. Nevertheless, all study approvals (IACUC, ACURO, HRPO) have been obtained and experiments using fibroblast growth factor receptor and androgen receptor inhibitors in patient derived xenograft models are underway. The approved project objectives and scope will not change and the work to be completed in the extension period will include all experiments and analyses related to Specific Aim 1.

15. SUBJECT TERMS
Castration-resistant prostate cancer, Patient-derived xenografts, Fibroblast growth factor receptor, Androgen receptor

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	8	19b. TELEPHONE NUMBER *(include area code)*

Table of Contents

Introduction

Early stage prostate cancer (PCa) cells typically require androgen receptor (AR) activity for survival and proliferation. Thus, standard PCa treatments include anti-androgen therapies that suppress AR function. However, the majority of PCa recur as castration-resistant prostate cancer (CRPC) that is resistant to AR-directed therapies. Observations in clinical CRPC specimens and *in vitro* studies using CRPC cell lines suggest that activation of the fibroblast growth factor (FGF) pathway is one mechanism to overcome AR pathway blockade [1, 2]. In addition, FGF pathway activity is critical for cell survival and proliferation in advanced AR-null mCRPC without neuroendocrine features (i.e. double-negative prostate cancer; Ref. [3]). While the FGF pathway has been nominated as a therapeutic target in treatment-resistant mCRPC, rigorous preclinical testing in in vivo models has yet to be conducted.

The Department of Urology at the University of Washington has generated one of the largest collections of mCRPC patient-derived xenograft (PDX) models. The LuCaP PDX series is comprised of over 40 different models that represent diverse molecular phenotypes of mCRPC [4, 5]. In addition, LuCaP PDX models retain patient tumor characteristics and serve as patient avatars for preclinical testing. In a pilot study using AR-expressing LuCaP PDX models, we determined that combination treatment with an FGF receptor (FGFR) inhibitor and AR inhibitor blocked tumor growth *in vivo* warranting further study. The objective of the proposed studies is to further examine FGFR inhibition in LuCaP PDX models and focus on (i) the efficacy of combination FGFR and AR pathway inhibitors in a larger preclinical PDX study and (ii) delineate the mechanisms of response and resistance to treatment *in vivo*. We hypothesize that AR pathway inhibition in AR-expressing CRPC leads to FGF pathway activation as a requirement for tumor cell survival and that combination FGFR and AR therapy will block tumor cell survival and proliferation.

These studies will provide insights on the molecular mechanisms of response and resistance to combined FGFR and AR pathway inhibition in AR-expressing models of CRPC in vivo. In addition, significant therapeutic responses observed in the proposed studies would strongly support rapidly translating combination FGFR and AR pathway inhibition to clinical trials in men with treatment-resistant mCRPC.

Body

Key Training and Research Accomplishments

As indicated in the statement of work, year 1 (months 0-12) of this PCRP-Early Investigator Research Award, training milestones consisted of didactic coursework and conference/workshop/seminar attendance; and research milestones included *in vivo* studies using FGFR and AR inhibitors in AR-expressing LuCaP PDX models. While significantly diminished due to restrictions imposed for the COVID 19 pandemic, accomplishments include:

- Completed *BIOST 509 - Introduction to R for Data Analysis in Health Sciences* to gain experience in R programming language.
- Completed *RNAseq Analysis in R* prepared by Combine Australia to learn methods for analyzing RNAseq experiments relevant to this proposal.
- Attended the AACR Translational Cancer Research For Basic Scientists Workshop
- Obtained all study approvals (i.e. IACUC, HRPO and ACURO)
- Obtained an ACURO amendment to use erdafitinib (FDA approved) instead of CH5183284 for *in vivo* testing.

Reportable Outcomes

To summarize the research accomplishments to date, the tasks described in the proposed Statement of Work (revised March 2020) are itemized here with a brief update for each task.

Specific Aim: Evaluate the efficacy of combination erdafitinib and enzalutamide in a PDX preclinical trial (months 1-12).

Major Task 1: Preclinical PDX Study

Subtask 1.1: Apply for IACUC, ACURO and HRPO approvals (Months 1-3). **Completed.** All study approvals were successfully obtained. In addition, an ACURO amendment was submitted and approved to use the FGFR inhibitor erdafitinib (FDA approved) instead of CH5183284 for *in vivo* testing.

Task 1.2: Enroll five PDX lines and implant one cell line. Monitor survival and tumor growth in mice. Three treatment groups per line - control, enzalutamide, combination enzalutamide and erdafitinib (Months 3-10). **In Progress.** Significant challenges were encountered due to delays acquiring the study approvals as well as due to the ongoing COVID-19 pandemic response. As such, *in vivo* testing using enzalutamide and erdafitinib in LuCaP PDX models have been initiated but have not reached data reporting endpoints at the time of this annual report.

Major Task 2: Molecular and Transcriptomic Characterization of Tumors from PDX Preclinical Trial

Subtask 2.1: Assess molecular features of response and emergent resistance mechanism(s) from tumor specimens obtained from the PDX preclinical trial using IHC, qPCR and immunoblot analyses (months 6-10). Not yet started.

Subtask 2.2: Identify features of response and emergent resistance mechanism(s) to treatment in the PDX tumors using RNAseq and GSEA (months 8-10). Not yet started.

Conclusions

The project was significantly impacted by the ongoing COVID-19 pandemic response and the longer than anticipated approval process for ACURO and ACURO amendments. A key factor leading to the study amendment (i.e. erdafitinib *in vivo* instead of CH5183284) was due to the recent FDA approval of erdafitinib for metastatic bladder cancer therapy. Thus, positive results from the proposed studies could be translated more rapidly to clinical trials in men with treatment-resistant mCRPC. However, the significant disruptions to study timelines prevented completion of this project in the proposed 12-month window period. To remedy these outcomes, a no cost extension to the grant timeline was recently approved. While there continues to be on-going COVID-related research disruptions, the initial PDX studies are already underway and all analyses and reporting should be completed within the extension period.

Publications

Title – "Targeting Fibroblast Growth Factor Receptors in Castration-Resistant Prostate Cancer"

Labrecque MP, Brown LG, Coleman IM, Lakely B, Nguyen HM, Corey E, Nelson PS and Morrissey M.

1. Poster presentation at AACR Advances in Prostate Cancer Research (2020), Denver, CO, USA *cancelled due to COVID-19*

Inventions, Patents and Licenses

None

References

1. Armstrong K, Ahmad I, Kalna G, Tan SS, Edwards J, Robson CN, et al. Upregulated FGFR1 expression is associated with the transition of hormone-naive to castrate-resistant prostate cancer. British journal of cancer. 2011;105(9):1362-9. doi: 10.1038/bjc.2011.367. PubMed PMID: 21952621; PubMed Central PMCID: PMC3241546.
2. Ozen M, Giri D, Ropiquet F, Mansukhani A, Ittmann M. Role of fibroblast growth factor receptor signaling in prostate cancer cell survival. Journal of the National Cancer Institute. 2001;93(23):1783-90. PubMed PMID: 11734594.
3. Bluemn EG, Coleman IM, Lucas JM, Coleman RT, Hernandez-Lopez S, Tharakan R, et al. Androgen Receptor Pathway-Independent Prostate Cancer Is Sustained through FGF Signaling. Cancer cell. 2017;32(4):474-89 e6. doi: 10.1016/j.ccell.2017.09.003. PubMed PMID: 29017058; PubMed Central PMCID: PMC5750052.
4. Nguyen HM, Vessella RL, Morrissey C, Brown LG, Coleman IM, Higano CS, et al. LuCaP Prostate Cancer Patient-Derived Xenografts Reflect the Molecular Heterogeneity of Advanced Disease an--d Serve as Models for Evaluating Cancer Therapeutics. The Prostate. 2017;77(6):654-71. doi: 10.1002/pros.23313. PubMed PMID: 28156002; PubMed Central PMCID: PMC5354949.
5. Labrecque MP, Coleman IM, Brown LG, True LD, Kollath L, Lakely B, et al. Molecular profiling stratifies diverse phenotypes of treatment-refractory metastatic castration-resistant prostate cancer. J Clin Invest. 2019;130:4492-505. Epub 2019/07/31. doi: 10.1172/JCI128212. PubMed PMID: 31361600; PubMed Central PMCID: PMCPMC6763249.

Appendix

None