AWARD NUMBER: W81XWH-18-1-0157

TITLE: Overcoming PARP Inhibitor Resistance of BRCA-Deficient Ovarian Cancers

PRINCIPAL INVESTIGATOR: Michael J. Birrer, MD, PhD

CONTRACTING ORGANIZATION: The University of Alabama at Birmingham
Birmingham, AL

REPORT DATE: June 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**
June 2020	Annual	15May2019-14May2020

4. TITLE AND SUBTITLE
Overcoming PARP Inhibitor Resistance of BRCA-Deficient Ovarian Cancers

5a. CONTRACT NUMBER

5b. GRANT NUMBER
W81XWH-18-1-0157

5c. PROGRAM ELEMENT NUMBER

6. AUTHOR(S)
Michael J. Birrer, MD, PhD

E-Mail: mbirrer@uams.edu

5d. PROJECT NUMBER

5e. TASK NUMBER

5f. WORK UNIT NUMBER

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

University of Alabama at Birmingham
701 S. 20th Street
Birmingham, AL 35294-0001

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
BRCA1 and BRCA2 are critical DNA repair proteins involved in the maintenance of DNA by allowing cells to overcome stress during DNA duplication. Indeed, BRCA1/2 will stabilize the DNA through the disabled replication process and allow DNA repair by the enzyme called Poly ATR-ribose polymerase (PARP). Once the DNA is repaired it can continue its replication process and the cell will keep living. *BRCA1/2* mutations are prevalent in ovarian cancer, leading to genomic instability and providing a unique opportunity for targeted therapy. Due to their inability to cope with replication stress, BRCA1/2-deficient cancer cells are highly sensitive to inhibitors of PARP (PARPi), which specifically interfere with the repair of DNA single-strand breaks which leads to the formation of DNA double strand breaks and consequent cell death. Three PARPi (olaparib, niraparib, rucaparib) have been approved by the FDA for the treatment of advanced ovarian cancer patients with BRCA mutations and shown significant efficacy. However, the vast majority of patients eventually developed resistance and relapsed. Therefore, a better understanding of how BRCA-deficient ovarian cancer cells acquire resistance to PARPi and how to overcome the resistance is urgently needed. We have gathered preliminary results suggesting that PARPi resistance of BRCA-deficient ovarian cancer cells is caused by an enzyme called ATR which is a master checkpoint of DNA metabolism. In this project, we will systematically address three key aspects of the hypothesis. In Aim 1, we will identify the specific alterations in DNA repair pathways driving PARPi resistance. In Aim 2, we will elucidate how ATR regulates DNA replication. In Aim 3, we will test the efficacy of the ATRi-PARPi combination therapy in ovarian cancer patients resistant to PARPi.

15. SUBJECT TERMS
PARP, ATR, resistance, ovarian cancer

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)*

Standard Form 298 (Rev. 8-98)
Prescribed by ANSI Std. Z39.18

Table of Contents

Page

1. INTRODUCTION:

The vast majority of patients with advanced stage epithelial ovarian cancer eventually suffer relapse and ultimately develop drug resistant disease. Initial recurrence after a long progression free interval is frequently sensitive to platinum and is treated with a platinum doublet. Patients can suffer multiple recurrences during the natural history of ovarian cancer. As the disease free interval becomes shorter, ovarian cancer becomes platinum resistant. For platinum resistant disease, there are relatively few therapeutic options and patients have a median survival of 11 months. Thus, the search for new effective agents for recurrent ovarian cancer is critical and highly significant.

PARP inhibitors (PARPi) are a new class of drugs recently approved for the treatment of ovarian cancer. They work by inhibiting the DNA repair protein PARP1 resulting in the accumulation of DNA single-stranded breaks (SSBs), which ultimately gives rise to DNA double-stranded breaks (DSBs) during DNA replication. DSBs are lethal to cells unless repaired by a BRCA1/2-dependent pathway. Thus, tumors arising from germ-line or somatic mutation in *BRCA1/2* are particularly sensitive to PARP inhibition, while cells with wild-type *BRCA1/2* are resistant. This phenomenon, known as "synthetic lethality", is precisely why these agents are effective for *BRCA1/2* disease (homologous recombination defect, HRD) with minimal toxicity. Recent data suggests that HRD can be found in up to 40-50% of high-grade serous cancers.

Three different PARPi (olaparib, rucaparib, niraparib) have recently been approved for the treatment or maintenance of platinum sensitive ovarian cancer. They demonstrate impressive response rates and prolongation of disease free intervals. However, most if not all patients will eventually develop PARPi resistance. It is likely that in a short period of time there will be vast numbers of ovarian cancer patients who have PARPi resistant disease. The mechanisms(s) of PARPi resistance are poorly understood. Reversion mutations have been identified in some cases, but there is evidence that other, yet unknown, mechanisms are also involved. The identification of mechanisms of resistance to PARPi is a critical effort with high clinical significance. This project, aimed at developing therapeutic interventions to overcome PARPi resistance, is focused precisely on this effort. If successful, it will provide the next generation of drugs for these patients.

2. KEYWORDS:

PARP, ATR, Resistance, Ovarian Cancer

3. ACCOMPLISHMENTS:

- **What were the major goals of the project?** The major goals of the project include: 1.) the determination of the alterations of DDR pathways leading to PARPi resistance in ovarian cancer; 2.) the understanding of the dependency of PARPi resistance on ATR in ovarian cancer; and 3.) overcoming the PARPi resistance of BRCA-deficient ovarian cancer patients. The first two goals involve both cell line work and animal experiments. The latter goal involves a human clinical trial.

Major Task 1: Regulatory Compliance for human studies
Subtask 1: Material transfer agreements (MTAs) or clinical trial agreements (CTAs) approval
Subtask 5: IRB protocol submission
Subtask 6: 2nd level IRB review (ORP/HRPO)
Subtask 7: Amendments, adverse events and protocol deviations as needed
Subtask 8: Annual IRB report for continuing review
Milestone Achieved: IRB and HRPO approvals
Major Task 2: Regulatory Compliance for animal studies
IACUC and ACURO approval of animal studies

Milestones achieved: animal studies approved

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

This DOD grant was awarded in June 2017 while the PI was on faculty at Massachusetts General Hospital. Due to his recruitment to UAB, the grant was not processed and activated at MGH but instead the grant was held until the PI was established at UAB. This resulted in the grant not being activated by the regulatory office at UAB till 2018. Within 2 months, the process started to established the subcontract with Dr Zou at MGH. Over the next 5 months of negotiation, Dr Zou felt that the animal studies could not be done at MGH. As such it was determined that the cell line experiments would be done at MGH and the animal experiments would be done at UAB. Over the subsequent two months an animal protocol was developed and submitted to the IAUCU at UAB. Considerable discussion occurred over the next several months to determine the optimal division of experiments to efficiently accomplish the goals of the project.

In parallel to this there was considerable effort to move forward the clinical trial portion of the grant. An initial phone call was placed to the suppliers of the therapeutic agents. For the ATRi, we discussed with the supplier (company) that the oral form appeared to be the preferred drug sine the IV form did not achieve high enough levels in the blood stream without toxicity. As such we had another phone conference to ascertain whether this would be acceptable to DOD and the conclusion was we should proceed with the oral form of the drug. After these phone conferences, the PI and team developed the appropriate clinical trial documents.

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

Dr. Arend is the nested young investigator on this grant. Dr. Birrer performed the role of her mentor on the development of AIM 3 through regular one-on-one meetings together. He guided her through writing the protocol, securing contracts with all necessary parties, submitting to the IRB and the FDA. Dr. Arend is completing her masters in clinical translational research under Dr. Birrer's mentorship. In addition, she has attended multiple conferences such as ASCO, NRG, ESMO, AACR, SGO and has now become the co-leader of the annual SGO clinical trial development workshop for young investigators.

Finally, she is also a full member of the DOD academy for which Dr Birrer is also her mentor. Both mentor and investigator have participated in all events in the academy.

- **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?**

In the next reporting we plan to do the following:

- Dr Zou's laboratory will begin the work characterizing the alterations in the DDR pathways in cell lines (AIM 1A, B) and defines the dependency of PARP resistance on ATR (AIM 2A, B, and C)). This will be done based upon a continuing collaboration and will eventually require a sub-contract.
- Dr Birrer's laboratory will begin on similar work focused on using PDX models (AIM 1A, B and AIM 2A,B and C)
- Dr Birrer and Dr Arend will develop a protocol and submit it to the IRB
- Dr Birrer and Dr Arend will establish contracts with the appropriate industrial partners to enable the trials

3. **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

4. **CHANGES/PROBLEMS:**

   Animal experiments

   - **Changes in approach and reasons for change**

     While there are no substantive changes in objectives or scope, the experiments described in the original project will now be done at UAB and not MGH

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

     As pointed out above there was a significant delay in the opening of the grant and ultimately the decision as to where the animal experiments would be performed. This has resulted in considerable time to open the animal protocol. However, despite delays an animal protocol has been developed and submitted. After several versions, the protocol has been approved.

   - **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.**

     See above

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

     Nothing to report

5. **PRODUCTS**

- **Publications, conference papers, and presentations**
  Report only the major publication(s) resulting from the work under this award.
  - **Journal publications.**

    Nothing to report
  - **Books or other non-periodical, one-time publications.**

    Nothing to report
  - **Other 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

6. **PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS**
   - **What individuals have worked on the project?**
     -

Name:	Michael Birrer
Project Role:	PI
Researcher Identifier (e.g. ORCID ID):	0000-0001-6464-4225
Nearest person month worked:	2
Contribution to Project:	Ms. Smith has performed work in the area of combined error-control and constrained coding.
Funding Support:	The Ford Foundation (Complete only if the funding support is provided from other than this award).

Name:	Rebecca Arend
Project Role:	Clinician
Researcher Identifier (e.g. ORCID ID):	1234567
Nearest person month worked:	5
Contribution to Project:	Ms. Smith has performed work in the area of combined error-control and constrained coding.
Funding Support:	The Ford Foundation (Complete only if the funding support is provided from other than this award).

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

  Nothing to report

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

  Nothing to report

  - **Organization Name:**
  - **Location of Organization:** *(if foreign location list country)*
  - **Partner's contribution to the projec**t *(identify one or more)*
    - **Financial support;**
    - **In-kind support** *(e.g., partner makes software, computers, equipment, etc., available to project staff)*;
    - **Facilities** *(e.g., project staff use the partner's facilities for project activities)*;
    - **Collaboration** *(e.g., partner's staff work with project staff on the project)*;
    - **Personnel exchanges** *(e.g., project staff and/or partner's staff use each other's facilities, work at each other's site)*; and
    - **Other.**

7. **SPECIAL REPORTING REQUIREMENTS**
   - **Nothing to report**
8. **APPENDICES:** *none*