AWARD NUMBERS: W81XWH-14-1-0518

TITLE: Development of a New Class of Drugs to Inhibit All Forms of Androgen Receptor in Castration-Resistant Prostate Cancers

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CONTRACTING ORGANIZATION: University of Minnesota Minneapolis, MN 55455

REPORT DATE: October 2015

TYPE OF REPORT: Annual Report

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

DISTRIBUTION STATEMENT: Approved for Public Release; Distribution Unlimited

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# Development of a New Class of Drugs to Inhibit All Forms of Androgen Receptor in Castration-Resistant Prostate Cancers

**Scott Dehm, Paul Rennie, Daniel Gewirth**

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14. ABSTRACT
Prostate cancer is the most frequently diagnosed male cancer and second leading cause of male cancer death. Management of patients with advanced-stage disease relies on inhibiting the androgen receptor (AR) with conventional endocrine targeting therapies, and more recently with second-generation endocrine targeting therapies designed to block AR activity that re-emerges during castration. However, despite a growing armamentarium of drugs targeting the androgen/AR signaling axis, progression of castration-resistant prostate cancer (CRPC) remains a major clinical challenge that undermines survival and quality of life for prostate cancer patients. The proposed research is focused on the pre-clinical development of VPC14228, a drug-like small molecule that targets the AR:DNA interaction. During the first year of this award, we have made progress in investigating the functional effects of VPC14228 on DNA interaction and transcriptional activation mechanisms for AR, developing a definitive experimental and structural characterization of the VPC14228 interaction with the AR DBD, and conducting pre-clinical evaluation of VPC14228.

15. SUBJECT TERMS

16. SECURITY CLASSIFICATION OF:
   a. REPORT
      Unclassified
   b. ABSTRACT
      Unclassified
   c. THIS PAGE
      Unclassified

17. LIMITATION OF ABSTRACT
   Unclassified

18. NUMBER OF PAGES
   Unclassified

19a. NAME OF RESPONSIBLE PERSON
    USAMRMC

19b. TELEPHONE NUMBER (include area code)

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

Prostate cancer is the most frequently diagnosed male cancer and second leading cause of male cancer death. Management of patients with advanced-stage disease relies on inhibiting the androgen receptor (AR) with conventional endocrine targeting therapies, and more recently with second-generation endocrine targeting therapies designed to block AR activity that re-emerges during castration. However, despite a growing armamentarium of drugs targeting the androgen/AR signaling axis, progression of castration-resistant prostate cancer (CRPC) remains a major clinical challenge that undermines survival and quality of life for prostate cancer patients. The proposed research is focused on the pre-clinical development of VPC14228, a drug-like small molecule that targets the AR:DNA interaction. The goals of this research are to investigate the functional effects of VPC14228 on DNA interaction and transcriptional activation mechanisms for AR, develop a definitive experimental and structural characterization of the VPC14228 interaction with the AR DBD, and conduct pre-clinical evaluation of VPC14228.

2. KEYWORDS
Androgen receptor, castration-resistant prostate cancer, DNA binding domain, androgen response element, AR inhibitor, chromatin, x-ray crystallography, pre-clinical

3. ACCOMPLISHMENTS

a. Major goals of the project as outlined in the approved Statement of Work (SOW):

1.1. Assay luciferase reporters in prostate cancer cell lines (months 1-12).
1.2. Determine effects of VPC14228 on AR binding to genomic AREs (months 13-24)
1.3. Chromatin immunoprecipitation (ChIP) assays with lentivirus-infected cells to test AR-V binding to genomic AREs (months 13-24)
1.4. Obtain approval for mouse xenograft studies from U of M IACUC and USAMRMC ACURO (months 1-6).
1.5. Establish xenografts with prostate cancer cell lines to generate tissue for chromatin fractionation and ChIP-seq assays (months 7-36)
1.6. Perform chromatin fractionation assays with prostate cancer cell lines and xenografts (months 1-6).
1.7. Perform ChIP-seq with prostate cancer cell lines and xenografts
2.1. Form crystals of ARdbd in complex with ARE DNA (Months 1-6).
2.2. Optimize ligand soaking conditions (Months 7-18).
2.3. Collect native and Zn- or Br-anomalous X-ray diffraction data from VP14228-soaked crystals using synchrotron radiation (Months 18-21).
2.4. Obtain phases by molecular replacement and single wavelength anomalous dispersion (Months 18-21).
2.5. Identify the ligand and its binding site, build the VP14228-ARdbd-DNA model, and refine the structure (Months 21-24).
2.6. Crystallize ARdbd or ARtnd in the presence and absence of VP14228 without DNA (Months 1-20).
2.7. Soak VP14228 into apo ARdbd or ARtnd crystals (Months 12-20).
2.8. Collect native and Zn-anomalous X-ray diffraction data from co-crystallized, soaked, and apo ARdbd or ARtnd crystals, with the final data sets collected using synchrotron radiation (Months 20-24).

2.9. Obtain initial phases by molecular replacement and single wavelength anomalous dispersion (Months 24-26).

2.10. Build the model and refine the structure (Months 26-30).

3.1. Molecular modeling of derivatives of our current lead VPC-14228 (months 1-30)

3.2. Synthesis of derivatives of our lead compounds (months 6-30).

3.3. Experimental evaluation of the developed synthetic derivatives (months 4-30)

3.4. Selection of several lead compounds for pharmacological development

b. Accomplishments under these goals:

Major Activities for University of Minnesota Site (Dehm, PI)
In the first reporting period, the major activities consisted of a) Testing the effects of VPC14228 and the derivative VPC14449 on AR transcriptional activity in prostate cancer cell lines using luciferase reporter assays; b) Testing the effects of VPC14228 and the derivative VPC1449 on AR chromatin binding and transcriptional output in prostate cancer cell lines using ChIP and quantitative RT-PCR approaches; c) Testing the effects of VPC14228 on AR-V chromatin binding in prostate cancer cell lines using lentivirus infection and ChIP; d) applying for and securing approval from IRB and ACURO authorities to perform mouse studies; e) Testing the effects of VPC14228 and the derivative VPC14449 on AR chromatin binding using chromatin fractionation techniques.

Major Activities for Hauptman Woodward Institute Site (Gewirth, PI)
In the first reporting period, the major activities consisted of a) expressing and purifying the ARdbd protein, b) forming the ARdbd-DNA complex, c) carrying out crystallization procedures and trials on the ARdbd and ARdbd-DNA complex, d) carrying out crystallization trials on ARdbd protein and ARdbd protein mixed with VP14228 ligand, and e) soaking VP14228 ligand into crystals of ARdbd-DNA complexes.

Major Activities for Vancouver Prostate Centre Site (Rennie, PI)
In the first reporting period, the major activities consisted of a) design and synthesis of new chemical derivatives based on VPC-14228; b) testing of these new molecules in biological assays; and c) analysis of their metabolic stability for further pharmacological development.

Specific Objectives for University of Minnesota Site (Dehm, PI)
The specific objectives for this reporting period were to a) assess the effects of VPC14228 on AR DNA binding and transcriptional activity in reporter assays; b) assess the effects of VPC14228 on AR and AR-V binding and transcriptional regulation of endogenous target genes; c) obtain approvals for mouse studies; d) assess the effects of VPC14228 on AR chromatin binding. These objectives correspond to Tasks 1.1, 1.2, 1.3, 1.4, and 1.6 as described in the approved SOW.
Specific Objectives for Hauptman Woodward Institute Site (Gewirth, PI)
The specific objectives for this reporting period were to a) produce ARdbd protein, b) form ARdbd-DNA complexes, c) grow diffracting crystals of the ARdbd-DNA complex, d) collect diffraction data from the ARdbd-DNA complex, e) identify conditions that produce crystals of ARdbd, and f) evaluate the stability of ARdbd-DNA crystals when soaked with VP14228 ligand. These objectives correspond to Tasks 2.1, 2.2, 2.6, and 2.8 as described in the approved SOW.

Specific Objectives for Vancouver Prostate Centre Site (Rennie, PI)
The specific objectives for this reporting period was to a) identify weak structural points in the structures of lead compounds VPC-14228 and VPC-14449 then b) use medicinal chemistry and molecular docking analysis to design new compounds that would overcome these structural problems, and c) testing these new compounds to select those that retain a low nanomolar activity while having an improved metabolic stability. These objectives correspond to Tasks 3.1, 3.2, 3.3, and 3.4 as described in the approved SOW.

Key Outcomes for University of Minnesota Site (Dehm, PI)

Key Outcome 1: VPC14228 inhibits activity of PSA-LUC and MMTV-LUC reporters in LNCaP cells (Fig. 1), but paradoxically activates a 4XARE-LUC reporter in R1-AD1 cells (Fig. 2). The Vancouver site has also observed cell line variability in luciferase reporter readouts following VPC14228 treatments. This finding indicates that luciferase-based studies should be de-emphasized because this approach is more prone to artifacts than studies with endogenous genes.

Key Outcome 2: Using ChIP, we have found that VPC14228 represses AR binding to the PSA enhancer in LNCaP cells. Similarly, VPC14228 also represses AR-V7 binding to the PSA enhancer in lentivirus-infected LNCaP cells. In line with this repressive effect, VPC14228 inhibited DHT-mediated induction of PSA mRNA in LNCaP cells.

Key Outcome 3: Using chromatin fractionation, we have found that VPC14228 inhibits AR binding to chromatin in LNCaP cells at 1uM concentration, but this effect is reduced at 10uM concentration (Fig. 3). The Vancouver site has also observed this effect in their studies, and have concluded it is due to VPC14228 functioning as an AR agonist by binding to the AR ligand binding domain at higher drug concentrations. This finding supports the future inclusion of VPC14449 in functional assays, as this derivative does not have this AR LBD binding property.
Fig. 1. LNCaP cells were transfected with A) PSA-LUC or B) MMTV-LUC reporters and treated with combinations of mibolerone (Mib) and VPC14228 at the indicated concentrations. Ethanol (ETH) and DMSO served as vehicle controls for mibolerone and VPC14228, respectively.
**Fig. 2.** R1-AD1 cells were transfected with 4XARE-LUC and treated with combinations of mibolerone (Mib) and VPC14228 at the indicated concentrations. Ethanol (ETH) and DMSO served as vehicle controls for mibolerone and VPC14228, respectively.
Fig. 3. LNCaP cells were treated with combinations of VPC14228 and dihydrotestosterone (DHT) at the indicated concentrations and subjected to chromatin fractionation. Chromatin fractions were subjected to western blot analysis with antibodies specific to AR and histone H3 (chromatin fraction control).
**Key Outcomes for Hauptman Woodward Institute Site (Gewirth, PI)**

Key Outcome 1: We have successfully produced ARdbd protein, the ARdbd-DNA complex, and have grown crystals of ARdbd in complex with DNA. We have collected diffraction data from these crystals and calculated an electron density map. This work has revealed the structure of the previously determined protein-DNA complex. This is a necessary control step prior to the ligand soaking experiments.

Key Outcome 2: We have carried out ligand soaking experiments with ARdbd-DNA crystals and the VP14228 ligand. This work has established that the soaking of the ligand does not disrupt the crystal lattice, an important benchmark for the future viability of this approach.

Key Outcome 3: We have tested VP14228-soaked ARdbd-DNA crystals for diffraction and anomalous scattering. Because the VP14228 ligand contains 2 bromine atoms, we carried out a fluorescence scan at the Bromine edge. This scan was positive for Bromine, confirming that the ligand had been soaked into the crystal. We also collected X-ray diffraction data at the Bromine edge from VP-soaked crystals. We observed the presence of a strong anomalous diffraction signal. This confirms the incorporation of the VP ligand into the ordered crystalline lattice, suggesting that the ligand was indeed bound to the protein-DNA complex.

Key Outcome 4: We have carried out preliminary crystallization screening operations with the ARdbd protein alone and in the presence of the VP14228 ligand. We have identified a positive crystal hit for the apo-protein and 4 positive crystal hits for the protein-ligand complex. This is a key step in the determination of the crystal structure of the protein alone.

**Key Outcomes for Vancouver Prostate Centre Site (Rennie, PI)**

Key outcome 1: *Using our in-silico model we identified and characterized 8 novel small molecule compounds inhibitor of the AR with an IC$_{50}$ under 500 nM in transcription assays.*

A new round of medicinal chemistry was performed using the original X-ray structure from Dr Gewirth's laboratory (Shaffer et al, PNAS 2004) and structural docking information from compounds VPC-14228 and VPC-14449. Weak structural points in their stability was also taken into account. The virtual molecular derivatives were docked into the DBD binding site using Glide and eHiTS program. Compounds that received moderate to higher score by Glide SP were selected and re-docked using the eHiTS docking protocol. Selected docked ligands were subjected to additional on-site scoring using the Ligand Explorer (LigX) program and the pKi predicting module of the Molecular Operating Environment (MOE). With this information, a cumulative scoring of four different predicted parameters (Glide score, eHiTS score, LigX score and pKi predicted by the MOE) were generated with each molecule, receiving a binary 1, 0 score for every “top 20% appearance”. The final cumulative vote resulted in about 22 compounds that consistently demonstrated high predicted binding affinity toward the DBD site.

All the 22 compounds were then screened for their ability to inhibit AR transcriptional activity using a nondestructive, cell-based enhanced green fluorescent protein (eGFP) AR transcriptional assay. All compounds were subjected to concentration-dependent titration to establish their
corresponding IC$_{50}$ values. To ensure these values are true positive hits in the AR transcriptional eGFP assay, we validated their activity by a second transcription assay, based on light detection instead of fluorescence, by quantifying their effect on the production of the prostate specific antigen (PSA) in prostate cancer cell lines. As expected, hit compounds induced a equivalent decrease in PSA levels in LNCaP cells as the IC$_{50}$ values found with the eGFP assay. Table 1 below summarize compounds that were found to have an IC$_{50}$ below 1 µM. For comparison purposes, in this assay, gold standards Enzalutamide and Bicalutamide show IC$_{50}$ of 100 nM and 600 nM respectively. Importantly, seven new compounds showed excellent activity under 500 nM.

Table 1: Compounds that exhibited an IC$_{50}$ under 0.5 µM in transcription assays (eGFP and PSA)

<table>
<thead>
<tr>
<th>Internal Number</th>
<th>IC$_{50}$ eGFP (µM)</th>
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<tr>
<td>14534</td>
<td>0.04</td>
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Key outcome 2: These new compounds are effective to reduce the growth of prostate cell lines including an Enzalutamide-resistant cell line. They also show no direct interaction with the AR by Biolayer Interferometry experiments.

To determine the translational potential of the most potent DBD inhibitors listed in Table 1, we evaluated their ability to reduce growth of prostate cell lines stimulated by the androgen R1881. The cell viability was assessed after 3 days of incubation with the test compound in a concentration dependent manner. Table 2 shows the effectiveness of our selected compounds to inhibit the growth of LNCaP cells, with their IC$_{50}$ values. These compound also did not show any effect on AR independent PC3 cell lines, confirming their AR-specific activity. Moreover, these compound were also very effective in inhibiting the growth of MR49F cells resistant to Enzalutamide at an IC$_{50}$ similar to what was observed in LNCaP cells.
Table 2: Effect on proliferation of LNCaP cells with effective DBD compounds

<table>
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Biolayer interferometry (BLI) studies can demonstrate if a direct reversible interaction between compounds and a purified AR ligand binding domain (LBD) occurs in a dose dependent manner. As our compounds are predicted to bind to the DBD we should not be able to detect such interaction. Figure 4a confirms that there is indeed no interaction of the lead compound VPC-14518 with the LBD of AR, compared to the LBD binding positive control VPC 13688 (Fig 4b).

**Figure 4: Analysis of DBD compounds interaction with AR LBD determined by Biolayer Interferometry analysis**

a) ![Graph](image1)

b) ![Graph](image2)
Key outcome 3: These compounds are effective to inhibit AR-V7 variants and can reduce the growth of prostate cell lines driven by the AR variant V7.

Because most AR splice variants retain the DBD domain they should be vulnerable to inhibition by our DBD compounds. We tested for inhibition of the transcriptional activity of variant AR-V7 using a luciferase reporter assay under the control of an AR promoter. The activity of transiently expressed AR-V7 was reduced with increasing concentrations of VPC-14449 without altering AR-V7 expression (Fig. 5). Control experiments with enzalutamide showed no effect on AR-V7 activity (Fig. 5) and are consistent with the absence of the LBD from this variant. Notably, the compounds did not achieve complete inhibition and were less effective against the transcriptional activity of AR-V7 (IC$_{50}$ 4–8 µM) when compared with inhibition of the full-length receptor. All compounds under 500 nM were tested in this system.

Figure 5: Effect of DBD compounds VPC-14449 on the activity of AR-V7 variant

Furthermore, we investigated the capability of these compounds to inhibit the growth of a prostate cancer cell line 22rv1 that is known to be driven by an AR variant (AR V7). Lead compounds VPC-14449 and VPC-14518 were effective to inhibit the growth of this cell line, albeit at a higher concentration (around 10 µM) than with prostate cell lines driven by the full length AR (Figure 6).
To further validate the site of action of AR-DBD binders, we introduced point mutations at residues that are predicted to interact with the lead compounds. Two positions (Tyr-594 and Gln-592) that were identified in the region with amino acid differences among related nuclear receptors. We are still working on the characterization of the effects of these mutations on the interaction of our various DBD compounds. This task will be continued in year 2.

Key outcome 4: Half-life of these compounds in metabolic conditions were improved, as few of the very active compounds initially had a half-life under 20 minutes in microsome studies.

All compounds that were shown to be effective in in vitro assays were evaluated for their stability. All compounds tested were soluble up to 50 µM in media (not shown). Preliminary data using microsome studies have shown that many of our previous compounds did not show a great half-life in metabolic stability. For example, compounds VPC-14228 and 14449 displayed 18 and 30 minutes of half-life in microsome experiments respectively. However, our new lead compound VPC-14518 was found to have a half-life of 263 minutes in microsome experiment, up from 30 minutes for 14449. All the novel compounds will be eventually tested using this technology.

c. Opportunities for training and professional development:

Nothing to report

d. Results disseminated to communities of interest:

Nothing to report
e. Plans for next reporting period:

**University of Minnesota Site (Dehm, PI)**
Based on our findings that VPC14228 actively inhibits AR chromatin binding in prostate cancer cells, but has a liability of AR agonist activity at higher concentrations, we will add VPC14449 to assays planned in Year 2. These assays will include ChIP and electrophoretic mobility shift assays (EMSAs) to determine effects of VPC14228 and VPC14449 on AR and AR variant binding to androgen response elements (AREs), ongoing chromatin fractionation assays with VPC14228 and VPC14449 treated cell lines and xenografts, and optimization of conditions for ChIP-seq assays.

**Hauptman Woodward Institute Site (Gewirth, PI)**
Having established that ARdbd-DNA crystals can incorporate ordered VP14228 ligand, we will next test soaking conditions in order to optimize the diffraction from the soaked crystals. This involves varying the concentration, soaking duration, temperature, and solvent of the VP14228 compound. We will test soaked crystals for anomalous diffraction and collect diffraction data from appropriately diffracting specimens. We will also carry out optimization of crystallization conditions for the apo ARdbd protein and for the ARdbd-VP14228 complex with the objective of growing suitable specimens for diffraction testing.

**Vancouver Prostate Centre Site (Rennie, PI)**
Having characterized new lead compounds with a much improved metabolic stability we will undergo *in-vivo* experiment with mice. Pharmacokinetics of these compounds will be evaluated, to determine their best option for delivery (oral gavage or intravenous injection). We will evaluate their toxicity using a series of dose escalation studies in mice. Pharmacokinetic and tissue distribution studies will be completed with non-toxic lead compounds in LNCaP tumour bearing mice to establish the drugs levels anticipated in efficacy studies. Finally with each lead compounds the initial in vivo screening for tumour growth inhibition will be done using LNCaP tumour xenografts, Enzalutamide-resistant MR49F cells, as well as genome-engineered R1-D567 cells from the Dehm Lab. In parallel other medicinal chemistry avenues are being explored to design even better compounds which will also be tested *in vitro* in year 2.

4. IMPACT

a. **Impact on the development of the principal discipline(s) of the project?:**
University of Minnesota Site: Nothing to Report
Hauptman Woodward Institute Site: Nothing to Report
Vancouver Prostate Centre Site: Nothing to Report

b. **Impact on other disciplines:**
University of Minnesota Site: Nothing to Report
Hauptman Woodward Institute Site: Nothing to Report
c. **Impact on technology transfer:**

University of Minnesota Site: Nothing to Report  
Hauptman Woodward Institute Site: Nothing to Report  
Vancouver Prostate Centre Site: Nothing to Report

**d. Impact on society beyond science and technology:**

University of Minnesota Site: Nothing to Report  
Hauptman Woodward Institute Site: Nothing to Report  
Vancouver Prostate Centre Site: Nothing to Report

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5. **CHANGES/PROBLEMS**

a. **Changes in approach and reasons for change**

University of Minnesota Site: Nothing to Report  
Hauptman Woodward Institute Site: Nothing to Report  
Vancouver Prostate Centre Site: Nothing to Report

b. **Changes that had a significant impact on expenditures**

University of Minnesota Site: Nothing to Report  
Hauptman Woodward Institute Site: Nothing to Report  
Vancouver Prostate Centre Site: Nothing to Report

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

University of Minnesota Site: Nothing to Report  
Hauptman Woodward Institute Site: Nothing to Report  
Vancouver Prostate Centre Site: Nothing to Report

6. **PRODUCTS**

a. Publications, conference papers, and presentations

University of Minnesota Site: Nothing to Report  
Hauptman Woodward Institute Site: Nothing to Report  
Vancouver Prostate Centre Site: Abstracts presented at the AACR annual meeting in Philadelphia, 2015:

   1) Selectively targeting the DNA-binding domain of the androgen receptor as a prospective therapy for prostate cancer
2) Structure-based study to overcome cross-reactivity of novel androgen receptor inhibitors

Kush Dalal, Mani Roshan-Moniri, Aishwariya Sharma, Huifang Li, Fuqiang Ban, Mohamed D. Hassona, Michael Hsing, Kriti Singh, Eric LeBlanc, Scott Dehm, Emma Tomlinson Guns, Artem Cherkasov, Paul S. Rennie. 1Vancouver Prostate Centre, Vancouver, BC, Canada; 2Masonic Cancer Center, University of Minnesota, Minneapolis, MN

7. PARTICIPANTS & OTHER COLLABORATING ORGANIZATIONS

a. Individuals who have worked on the project:

University of Minnesota Site (Dehm, PI)

<table>
<thead>
<tr>
<th>Name</th>
<th>Dr. Scott Dehm</th>
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<tr>
<td>Project Role</td>
<td>PI</td>
</tr>
<tr>
<td>Researcher Identifier</td>
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<tr>
<td>Person months worked</td>
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<tr>
<td>Contribution to project</td>
<td>Dr. Dehm provided project oversight, supervised research carried out by Dr. Chan, secured regulatory approvals, managed grant budget and reporting, and coordinated with co-PIs Rennie and Gewirth.</td>
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Funding support

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<tr>
<th>Name</th>
<th>Dr. Siu Chiu Chan</th>
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<tr>
<td>Project Role</td>
<td>Research Associate</td>
</tr>
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<td>Contribution to project</td>
<td>Dr. Chan collected and analyzed data under supervision of Dr. Dehm</td>
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Hauptman Woodward Institute Site (Gewirth, PI)

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<td>Contribution to project</td>
<td>Dr. Gewirth carried out oversight of the project, provided guidance and consultation to Dr. Que, and assisted with the collection of diffraction data.</td>
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<th>Dr. Nanette Que</th>
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**Vancouver Prostate Centre Site (Rennie, PI)**

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<td>Contribution to project</td>
<td>Dr. Rennie provided project oversight, and supervised research carried out by Dr. Dalal for the characterization of derivatives using biochemical assays.</td>
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<tr>
<th>Name</th>
<th>Dr. Artem Cherkasov</th>
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</thead>
<tbody>
<tr>
<td>Project Role</td>
<td>Collaborator</td>
</tr>
<tr>
<td>Researcher Identifier</td>
<td></td>
</tr>
<tr>
<td>Person months worked</td>
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</tr>
<tr>
<td>Contribution to project</td>
<td>Dr Cherkasov oversees all aspects of computational drug design, molecular modeling and bioinformatics.</td>
</tr>
<tr>
<td>Funding support</td>
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<table>
<thead>
<tr>
<th>Name</th>
<th>Dr. Emma Guns</th>
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<tbody>
<tr>
<td>Project Role</td>
<td>Collaborator</td>
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<tr>
<td>Researcher Identifier</td>
<td></td>
</tr>
<tr>
<td>Person months worked</td>
<td>1.2</td>
</tr>
<tr>
<td>Contribution to project</td>
<td>Dr. Guns supervised the work of Dr Hessein with the analysis of metabolic stability of compounds</td>
</tr>
<tr>
<td>Funding support</td>
<td></td>
</tr>
</tbody>
</table>
b. Changes in the active other support of the PD/PI(s) or senior/key personnel since the last reporting period:

University of Minnesota Site (Dehm, PI)

The following grants have been activated since negotiation and setup of the current award:

MNP #14.37 02/01/2015-02/28/2017
MN Partnership for Biotechnology and Medical Genomics
“Abiraterone Acetate Pharmacogenomics
The goal of this proposal is to identify and overcome mechanisms of resistance to AR-targeted therapy in prostate cancer via targeting the AR TAU5/WHTLF domain.
Role: Principal Investigator with co-PI Manish Kohli, M.D., Mayo Clinic
Overlap: None.

W81XWH-15-1-0220 07/01/2015-06/30/2015
DOD PCRP Exploration-Hypothesis Development Award
“Molecular Profiling of EPI-001: An Inhibitor of Androgen Receptor Signaling With a Disputed Mechanism of Action”
The goal of this project is to identify proteomic targets of the AR N-terminal antagonist EPI-001.
Role: Co-I (PI: Dan Harki, Ph.D.)
Overlap: None.
DOD PCRP Synergistic Idea Development Award
“Abiraterone Acetate Pharmacogenomics”
The goal of this proposal is to leverage the resources of an existing clinical trial at Mayo Clinic to investigate the evolution of metastatic tumor subclonal architecture in patients and patient-derived xenografts treated with abiraterone acetate. Role: PI with co-PIs Manish Kohli, M.D. and Liewei Wang, Ph.D., Mayo Clinic
Overlap: None.

DOD PCRP Synergistic Idea Development Award
“Androgen Receptor Gene Rearrangements in EpCAM-Positive and -Negative Circulating CTCs”
The goal of this proposal is to compare paired genomic and transcriptomic profile of EpCAM positive and negative CTCs for AR pathway alterations as potential biomarkers of resistance to AR signaling inhibitors. The total direct costs for this grant are $750,000.
Role: PI with co-PI Joshua Lang, M.D., University of Wisconsin
Overlap: None.

Movember / Prostate Cancer Foundation Challenge Award
“Targeting Aberrant AR-FL and AR-V Expression and Activity to Overcome Therapy Resistance in Metastatic Castration-Resistant Prostate Cancer”
The goal of this project is to develop antisense technologies to extinguish expression and activity of all forms of the full-length AR and truncated AR variants in prostate cancer. I will be responsible for TALEN-based gene editing to develop AR- and AR-V-driven prostate cancer models with impaired function of PSA enhancer RNA (eRNA).
Role: PI with co-PIs Haojie Huang, Ph.D., Mayo Clinic, and Martin Gleave M.D., Vancouver Prostate Centre.
Overlap: None.

Hauptman Woodward Institute Site (Gewirth, PI)
The following grant has been activated as of September 1, 2015
NIH/NCI
“Endoplasmic Reticulum Chaperones in Cancer Biology and Therapy”
Role on project: PI of Project 3; co-I of Project 1
This multi-PI PPG examines the structure, function, mechanism, and drug targeting of the major ER chaperone Grp94.
Overlap: none
Movember / Prostate Cancer Foundation Challenge Award
“Targeting Aberrant AR-FL and AR-V Expression and Activity to Overcome Therapy Resistance in Metastatic Castration-Resistant Prostate Cancer”
The goal of this project is to develop antisense technologies to extinguish expression and activity of all forms of the full-length AR and truncated AR variants in prostate cancer. I will be responsible for TALEN-based gene editing to develop AR- and AR-V-driven prostate cancer models with impaired function of PSA enhancer RNA (eRNA).
Role: PI with co-PIs Haojie Huang, Ph.D., Mayo Clinic, and Martin Gleave M.D., Vancouver Prostate Centre.
Overlap: None.

Principal Investigator : Butler M
Prostate Cancer Canada
Small molecule inhibition of ETS factors in prostate cancer.
The goal of this project is to develop small molecule inhibitors to inhibit ETS factors such as ERG in prostate cancer.
Role: Paul Rennie, Co-Investigator
Overlap: None.

c. What other organizations were involved as partners?

This is a Synergistic Idea Development Award being conducted at partnering organizations University of Minnesota, Hauptman Woodward Medical Research Institute, Inc., and the Vancouver Prostate Centre.

8. SPECIAL REPORTING REQUIREMENTS
This report is for a COLLABORATIVE AWARD, and was prepared jointly by the three study PIs. The tasks are clearly marked with the responsible PI and research site.

9. APPENDICES
NONE