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14. ABSTRACT There is an urgent need to better understand the biology contributing to the worse prognosis observed in African American (AA) prostate cancer (PCa) patients in order to 1) predict which men are more likely to suffer from an aggressive form of the disease and 2) develop more patient-specific treatments. Because AA men are less likely to be included in key PCa studies and clinical trials, we miss many other potential genetic indicators that steer molecular mechanisms, disease outcomes, and drug response. For example, human epidermal growth factor receptor-2 (HER2) overexpression has been correlated with advanced stage, treatment resistance, and worse outcome for PCa patients. Phase II clinical trials targeting HER2 were initiated, but major limitations were the lack of enrollment of study participants with HER2+ tumors as well as poor efficacy with anti-HER2 single agents. PCa patients were not stratified by race, and racial differences in HER2 expression were not evaluated. In this study, I aim to establish that HER2 expression promotes cellular growth, clonogenicity, and migration in a racially diverse panel of PCa cell lines and tumor tissue specimens. Thus far, results suggest that the targeting of HER2 with trastuzumab inhibits viability of RC-77T/E, an AA PCa cell line, but not PC3, a European American (EA) cell line. I will also confirm whether HER2 overexpression is positively correlated with West African ancestry (WAA) in tumor tissue specimens. Preliminary RNAseq analysis of prostate tissue suggested a moderate positive correlation between HER2 gene expression and WAA (r=0.1593). Additional genotyping analysis of isolated genomic DNA that is patient-matched to a 456 Case Race Disparity TMA is underway to support these prior findings. I will also determine if HER2 overexpression correlates with disease stage, clinical features, treatments, and outcomes in PCa patients. Preliminary HER2 membrane staining of AA FFPE specimens with IHC showed 30% (3/10) scored as 2+ or equivocal. In-progress IHC analysis of 400 FFPE specimens acquired from the COHCCC Frozen Tissue Bank as well as 456 Case Race Disparity TMA acquired from the DOD PCBN will provide additional insight into the correlation with HER2 overexpression and clinical variables associated with aggressive PCa and worse outcomes. Timely completion of this and a subsequent clinical trial would have a major near-term impact in lessening the burden of PCa mortality in AA men and would provide an additional treatment option for others with HER2+ metastatic PCa.					
15. SUBJECT TERMS prostate cancer, HER2, African American, health disparities, mCRPC, West African ancestry					
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INTRODUCTION: African American (AA) men suffer from increased prostate cancer (PCa) incidence and mortality.¹ Gene expression profiles confirm prominent racial differences in tumor biology, and a variety of pro-tumorigenic genes are differentially expressed in AA men.²⁻⁵ Human epidermal growth factor receptor-2 (HER2) is a tyrosine kinase involved in the promotion of cellular division and suppression of apoptosis and is correlated with worse prognosis and treatment resistance in PCa patients, but this gene has not been evaluated in AA men.^{6,7} Clinical trials targeting HER2 were previously initiated in PCa patients. There is no indication that racial differences in HER2 expression were evaluated.⁸⁻¹¹ There is an urgent need to better understand the biology contributing to the worse prognosis observed in AA PCa patients in order to 1) predict which men are more likely to suffer from lethal disease and 2) develop more patient-specific treatments. In this study, we seek to establish that HER2 expression promotes cellular growth, clonogenicity, and migration in a racially diverse panel of PCa cell lines; confirm whether HER2 overexpression is positively correlated with West African ancestry (WAA) in tumor tissue specimens; and determine if HER2 overexpression correlates with disease stage, clinical features, treatments, and outcomes in PCa patients.

KEYWORDS: prostate cancer, HER2, African American, health disparities, mCRPC, West African ancestry

BODY

ACCOMPLISHMENTS:

What were the major goals of the project?

Specific Aim 1: Establish that HER2 expression promotes cellular growth, clonogenicity, and migration in a racially diverse panel of PCa cell lines.

Major Task 1: Quantify basal HER2 gene and protein levels.

Target Dates: 1-24 months

Percentage of Completion: 50%

Major Task 2: Evaluate whether HER2 overexpression alters cell growth, clonogenicity, and migration.

Target Dates: 12-24 months

Percentage of Completion: 0%

Major Task 3: Evaluate whether HER2 blockade alters cell growth, clonogenicity, and migration.

Target Dates: 9-24 months

Percentage of Completion: 40%

Specific Aim 2: Confirm whether HER2 overexpression is positively correlated with WAA in tumor tissue specimens.

Major Task 1: Access racially diverse tumor tissue specimens.

Target Dates: 1-15 months

Percentage of Completion: 100%

Major Task 2: Quantify HER2 in tumor tissue specimens.

Target Dates: 12-24 months

Percentage of Completion: 100%

Major Task 3: Calculate genetic ancestry of patients.

Target Dates: 12-18 months

Percentage of Completion: 60%

Specific Aim 3: Determine HER2 correlation with PCa disease stage, clinical features, treatments, and outcomes in patients.

Major Task 1: Stratify tumor tissue specimens.

Target Dates: 12-24 months

Percentage of Completion: 100%

Major Task 2: Correlate HER2 expression with disease stage, clinical features, treatments, and outcomes.

Target Dates: 12-24 months

Percentage of Completion: 50%

What was accomplished under these goals?

Specific Aim 1: Establish that HER2 expression promotes cellular growth, clonogenicity, and migration in a racially diverse panel of PCa cell lines.

Major Task 1: Quantify basal HER2 gene and protein levels.

Subtask 1: Examine HER2 mRNA levels in cancer cells using qPCR.

Cell lines used: MDA-PCa-2b, PC3, DU145, 22Rv1, RC-77T/E

I repeated qPCR with in-lab equipment (Applied Biosystems) to establish HER2 mRNA transcript levels in all cell lines. All cell lines were grown in a humidified incubator with 5% CO₂ at 37°. PC3 (Cat:CRL-1435), DU145 (Cat:HTB-81), 22Rv1 (Cat:CRL-2505) and MDA-PCa-2b (Cat:CRL-2422) cell lines were purchased from the American Type Culture Collection (ATCC). PC3, DU145, and 22Rv1 was cultured in RPMI 1640 medium (Corning, Cat:10-040-CV) supplemented with 10% fetal bovine serum (Corning, Cat:35010CV), penicillin-streptomycin (Corning, Cat:30001CI), and gentamicin (Gibco, Cat:15710064) as recommended by the supplier. MDA-PCa-2b (Cat:CRL-2422) cell line was cultured in F-12K medium (ATCC®, Cat:30-2004) supplemented with 20% fetal bovine serum (Corning, Cat:35010CV), cholera toxin (Sigma-Aldrich, Cat:C8052), epidermal growth factor (Sigma-Aldrich, Cat:E4127), *o*-phosphoethanolamine (Sigma-Aldrich, Cat:P0503), hydrocortisone (Sigma-Aldrich, Cat:H0888), selenious acid (ACROS Organics, Cat:AC19887), bovine insulin (Sigma-Aldrich, Cat:I6634), and penicillin-streptomycin as recommended by the supplier. The RC-77T/E cell line was provided by C.A. Casiano's laboratory with permission from J.S. Rhim. RC-77T/E was grown in collagen-coated treated culture dishes in keratinocyte serum-free medium in (K-SFM) supplemented with bovine pituitary extract and recombinant epidermal growth factor (Gibco, Cat:17-005-042) used for growing and maintaining the cells. 0.2% Normocin (Invivogen, Cat:ANT-NR-1) was added to the medium for all cell lines to prevent contamination by mycoplasma, bacteria, or fungi. In accordance with NIH guidelines concerning the authentication of key biological resources, I verified the identity and purity of the cell lines by performing mycoplasma testing (**Table 1**) using MycoProbe Mycoplasma Detection Kit (Cat: CUL001B) in addition to short tandem repeat (STR) profiling provided by ATCC (**Tables 2-6**) within the first six months of receipt. For STR profiling, seventeen STR loci plus the gender determining locus, Amelogenin, were amplified using the commercially available PowerPlex® 18D Kit from Promega. The cell line sample was processed using the ABI Prism® 3500xl Genetic Analyzer. Data were analyzed using GeneMapper® ID-X v1.2 software (Applied Biosystems). Appropriate positive and negative controls were run and confirmed for each sample submitted.

PC3	-0.0054
DU145	0.0028
22Rv1	-0.00075
MDA	-0.0088
RC77T	-0.0073

Table 1. Mycoplasma testing O.D. Values (Calculated) to verify absence of mycoplasma. <0.05 is considered negative with no mycoplasma detected. 0.05-0.10 is considered inconclusive with sample being suspect for mycoplasma. >0.10 is considered positive with mycoplasma detected.



Test Results for Submitted Sample				ATCC Reference Database Profile			
Locus	Query Profile: PC3			Database Profile: PC-3; Prostate Adenocarcinoma; Human (Homo sapiens)			
D3S1358	16						
TH01	6	7		6	7		
D21S11	29	31.2					
D18S51	14	15					
Penta_E	10	13	17				
D5S818	13			13			
D13S317	11			11			
D7S820	8	11		8	11		
D16S539	11			11			
CSF1PO	11			11			
Penta_D	9	11					
Amelogenin	X			X			
vWA	17			17			
D8S1179	13						
TPOX	8	9	11	8	9		
FGA	22	24					
D19S433	14						
D2S1338	18	20					
Number of shared alleles between query sample and database profile:							12
Total number of alleles in the database profile:							12
Percent match between the submitted sample and the database profile:							100
<i>The allele match algorithm compares the 8 core loci plus amelogenin only, even though alleles from all loci will be reported when available.</i>							
NOTE: Loci highlighted in grey (8 core STR loci plus Amelogenin) can be made public to verify cell identity. In order to protect the identity of the donor, please do not publish the allele calls from all the STR loci tested. Electropherograms showing raw data are attached.							

Explanation of Test Results

Cell lines with 80% match are considered to be related; i.e., derived from a common ancestry. Cell lines with between a 55% to 80% match require further profiling for authentication of relatedness.

- The submitted sample profile is human, but not a match for any profile in the ATCC STR database.
- The submitted profile is an exact match for the following ATCC human cell line(s) in the ATCC STR database (8 core loci plus Amelogenin):
- The submitted profile is similar to the following ATCC human cell line(s): CRL-1435
- An STR profile could not be generated.

Table 2. STR profiling performed by ATCC to verify that the PC3 sample profile provided matches (100%) the human cell line CRL-1435.



Test Results for Submitted Sample					ATCC Reference Database Profile			
Locus	Query Profile: DU145				Database Profile: DU 145; Prostate Carcinoma; Human (Homo sapiens)			
D3S1358	16							
TH01	7				7			
D21S11	28	30	31	33				
D18S51	12	17						
Penta_E	12	13	14					
D5S818	10	13			10	13		
D13S317	12	13	14		12	13	14	
D7S820	7	10	11		7	10	11	
D16S539	11	13			11	13		
CSF1PO	10	11			10	11		
Penta_D	9	11	13					
Amelogenin	X	Y			X	Y		
vWA	17	18			17	18	19	
D8S1179	13	14						
TPOX	11				11			
FGA	22	24						
D19S433	13	15						
D2S1338	16	19						
Number of shared alleles between query sample and database profile:								18
Total number of alleles in the database profile:								19
Percent match between the submitted sample and the database profile:								95
<i>The allele match algorithm compares the 8 core loci plus amelogenin only, even though alleles from all loci will be reported when available.</i>								
NOTE: Loci highlighted in grey (8 core STR loci plus Amelogenin) can be made public to verify cell identity. In order to protect the identity of the donor, please do not publish the allele calls from all the STR loci tested. Electropherograms showing raw data are attached.								

Explanation of Test Results

Cell lines with 80% match are considered to be related; i.e., derived from a common ancestry. Cell lines with between a 55% to 80% match require further profiling for authentication of relatedness.

- The submitted sample profile is human, but not a match for any profile in the ATCC STR database.
- The submitted profile is an exact match for the following ATCC human cell line(s) in the ATCC STR database (8 core loci plus Amelogenin):
- The submitted profile is similar to the following ATCC human cell line(s): HTB-81
- An STR profile could not be generated.

Table 3. STR profiling performed by ATCC to verify that the DU145 sample profile provided matches (95%) the human cell line HTB-81.



Test Results for Submitted Sample					ATCC Reference Database Profile			
Locus	Query Profile: 22RV1				Database Profile: 22Rv1; Prostate Carcinoma; Human (Homo sapiens)			
D3S1358	15							
TH01	6	9.3			6	9.3		
D21S11	30							
D18S51	13	14						
Penta_E	5	13						
D5S818	11	13			11	12	13	
D13S317	9	12			9	12		
D7S820	9	10	11		9	10	11	
D16S539	12				12			
CSF1PO	10	11			10	11		
Penta_D	9	12						
Amelogenin	X	Y			X	Y		
vWA	15	21			15	21		
D8S1179	12	13	14					
TPOX	8				8			
FGA	20	23						
D19S433	13	14						
D2S1338	17	18						
Number of shared alleles between query sample and database profile:								17
Total number of alleles in the database profile:								18
Percent match between the submitted sample and the database profile:								94
<i>The allele match algorithm compares the 8 core loci plus amelogenin only, even though alleles from all loci will be reported when available.</i>								
NOTE: Loci highlighted in grey (8 core STR loci plus Amelogenin) can be made public to verify cell identity. In order to protect the identity of the donor, please do not publish the allele calls from all the STR loci tested. Electropherograms showing raw data are attached.								

Explanation of Test Results

Cell lines with 80% match are considered to be related; i.e., derived from a common ancestry. Cell lines with between a 55% to 80% match require further profiling for authentication of relatedness.

- The submitted sample profile is human, but not a match for any profile in the ATCC STR database.
- The submitted profile is an exact match for the following ATCC human cell line(s) in the ATCC STR database (8 core loci plus Amelogenin):
- The submitted profile is similar to the following ATCC human cell line(s): CRL-2505
- An STR profile could not be generated.

Table 4. STR profiling performed by ATCC to verify that the 22Rv1 sample profile provided matches (94%) the human cell line CRL-2505.



Test Results for Submitted Sample				ATCC Reference Database Profile			
Locus	Query Profile: MDA-PCa-2b			Database Profile: MDA PCa 2b; Prostate Adenocarcinoma; Human (Homo sapiens)			
D3S1358	14	17					
TH01	8	9		8	9		
D21S11	33.2						
D18S51	16	17					
Penta_E	8	14					
D5S818	12	13		12	13		
D13S317	11	12		11	12		
D7S820	8	10		8	10		
D16S539	10	11		10	11		
CSF1PO	10	12		10	12		
Penta_D	11	13					
Amelogenin	X	Y		X	Y		
vWA	16			16			
D8S1179	13	14	15				
TPOX	8	11		8	11		
FGA	20	21	22				
D19S433	13.2	14.2	16				
D2S1338	22	24					
Number of shared alleles between query sample and database profile:							17
Total number of alleles in the database profile:							17
Percent match between the submitted sample and the database profile:							100
<i>The allele match algorithm compares the 8 core loci plus amelogenin only, even though alleles from all loci will be reported when available.</i>							
NOTE: Loci highlighted in grey (8 core STR loci plus Amelogenin) can be made public to verify cell identity. In order to protect the identity of the donor, please do not publish the allele calls from all the STR loci tested. Electropherograms showing raw data are attached.							

Explanation of Test Results

Cell lines with 80% match are considered to be related; i.e., derived from a common ancestry. Cell lines with between a 55% to 80% match require further profiling for authentication of relatedness.

- The submitted sample profile is human, but not a match for any profile in the ATCC STR database.
- The submitted profile is an exact match for the following ATCC human cell line(s) in the ATCC STR database (8 core loci plus Amelogenin): CRL-2422
- The submitted profile is similar to the following ATCC human cell line(s):
- An STR profile could not be generated.

Table 5. STR profiling performed by ATCC to verify that the MDA-PCa-2b sample profile provided matches (100%) the human cell line CRL-2422.



Test Results for Submitted Sample				ATCC Reference Database Profile			
Locus	Query Profile: RC77T/E			Database Profile:			
D3S1358	15	18					
TH01	8	9					
D21S11	28	30					
D18S51	15	17					
Penta_E	12	14					
D5S818	12	13					
D13S317	12	14					
D7S820	9	10					
D16S539	11	13					
CSF1PO	9	11					
Penta_D	2.2	5					
Amelogenin	X	Y					
vWA	15						
D8S1179	13	15					
TPOX	9	10					
FGA	22						
D19S433	13	14.2					
D2S1338	19	22					
Number of shared alleles between query sample and database profile:							NA
Total number of alleles in the database profile:							NA
Percent match between the submitted sample and the database profile:							NA
<i>The allele match algorithm compares the 8 core loci plus amelogenin only, even though alleles from all loci will be reported when available.</i>							
NOTE: Loci highlighted in grey (8 core STR loci plus Amelogenin) can be made public to verify cell identity. In order to protect the identity of the donor, please do not publish the allele calls from all the STR loci tested. Electropherograms showing raw data are attached.							

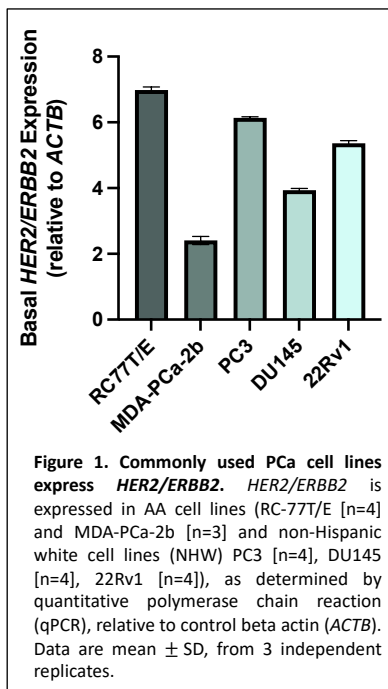
Explanation of Test Results

Cell lines with 80% match are considered to be related; i.e., derived from a common ancestry. Cell lines with between a 55% to 80% match require further profiling for authentication of relatedness.

- The submitted sample profile is human, but not a match for any profile in the ATCC STR database.
- The submitted profile is an exact match for the following ATCC human cell line(s) in the ATCC STR database (8 core loci plus Amelogenin):
- The submitted profile is similar to the following ATCC human cell line(s):
- An STR profile could not be generated.

Table 6. STR profiling performed by ATCC to verify that the RC-77T/E sample profile provided is human, but not a match for any profile in the ATCC STR database. RC-77T/E is not available through ATCC. As such, I expected to see no match.

Following cell line authentication and mycoplasma testing, total RNA was extracted from cells using the PureLink RNA Mini Kit (Thermo Fisher Scientific, Cat. 12183025). RNA yield and quality were determined by the absorbance ratio at 260/280 nm using the NanoDrop One Microvolume UV-Vis Spectrophotometer (Thermo Fisher Scientific, Cat. ND-ONE-W). Total RNA (1 μ g) was reverse transcribed into cDNA using the RevertAid First Strand cDNA Synthesis Kit (Thermo Fisher Scientific, Cat. K1622) and random hexamer primers following the manufacturer's protocol. Primers for *HER2/ERBB2* (L: gggaaacctggaactcaccta, R: ccctgcacctctggata) and beta-actin (*ACTB*) commercially synthesized by Integrated DNA Technologies were optimized using the $\Delta\Delta$ CT method to establish HER2 mRNA transcript levels in all cell lines. qPCR was performed using the PowerUp SYBR Green Master Mix (Thermo Fisher Scientific, Cat. A25742) following the manufacturer's fast cycling mode protocol. qPCR amplification was performed using the QuantStudio 3 Real-Time PCR System (Thermo Fisher Scientific, Cat. A28137). Reactions were performed in duplicates for three independent experiments and data was normalized to the reference gene beta-actin (**Figure 1**). *HER2/ERBB2* is expressed in AA cell lines (RC-77T/E and MDA-PCa-2b) and non-Hispanic white (NHW) cell lines (PC3, DU145, and 22Rv1) with the highest expression observed in RC-77T/E.



Subtask 2: Examine HER2 protein expression in cancer cells using Western blot.

Cell lines used: MDA-PCa-2b, PC3, DU145, 22Rv1, RC-77T/E

To quantify HER2 protein expression in all cell lines, I originally proposed to use primary antibodies against HER2 and β -actin as a control for Western blot analysis with in-lab equipment (Invitrogen) using ECL chemiluminescence. As an alternative protocol, Western blot optimization using LI-COR with Total Protein Stain were underway. **Due to the transition of the lab from City of Hope in Duarte, CA to Morehouse School of Medicine in Atlanta, GA, experiments for Subtask 2 were temporarily halted in June 2022. Due to construction at Morehouse School of Medicine, all lab equipment and supplies have been contained in storage units in Atlanta, GA until lab space became available. The Burnham lab space became available in January 2023. Lab equipment and supplies will be moved into the Burnham lab over the next few weeks with experiments expected to resume full-time in February 2023.*

Briefly, equal amounts of protein from whole cell lysates (10 μ g) will be separated using sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE, Bolt NuPAGE 4–12%, Thermo Fisher Scientific, Cat: NW04120BOX) and transferred onto polyvinylidene difluoride membranes (Millipore, Cat: IPFL00010). Western blot normalization will be performed with LI-COR Revert 700 Total Protein Stain for Western Blot Normalization (Cat: 926-11011). Membranes will be blocked in LI-COR Intercept TBS Blocking Buffer (Cat: 926-35010). Membranes will then be probed individually with rabbit anti-HER2 antibody (1:1000) (Cat: 50-190-774) and LI-COR IRDye 800CW goat anti-rabbit secondary antibody (1:15,000) (Cat: 926-35010) and washed several times with TBS-T between each antibody application. Membranes will be imaged with the Odyssey Imaging System in the 700 and 800 nm channels. Protein bands from at least 3 independent experiments will be quantified using ImageJ Software. The ratios of anti-HER2 protein bands to the Total Protein Stain will be normalized.

Major Task 2: Evaluate whether HER2 overexpression alters cell growth, clonogenicity, and migration.

Subtask 1: Generate HER2-overexpressing PC3 and RC-77T/E cell lines using lentiviral transfection method.

Cell lines used: PC3, RC-77T/E

To complete Major Task 2, Subtask 1, I hired a postdoctoral fellow (Nicole Mavingire, Ph.D.) in June 2022. **Due to the transition of the lab from City of Hope in Duarte, CA to Morehouse School of Medicine in Atlanta, GA, experiments for Major Task 2 were temporarily halted in June 2022. Due to construction at Morehouse School of Medicine, all lab equipment and supplies have been contained in storage units in Atlanta, GA until lab space became available. The Burnham lab space became available in January 2023. Lab equipment and supplies will be moved into the Burnham lab over the next few weeks with experiments expected to resume full-time in February 2023.* Dr. Mavingire and a newly accepted graduate student, Ms. Janelle Moore, will generate HER2-overexpressing (HER2-OE) PC3 (European American (EA)) and RC-77T/E (AA) cells using lentiviral transfection method. Briefly, cells will be plated in 6-well plates at a density of $1 - 5 \times 10^5$ cells/mL to produce 60% confluency in 24 hours. Pre-packaged lentiviral particles—either human HER2 (G&P Biosciences) or empty vector control (G&P Biosciences)—with puromycin selection marker will be added to cell medium. Gene integration will be verified through qPCR and Western blot.

Subtask 2: Evaluate HER overexpression on cell growth with CellTiter-Glo Viability assay.

Cell lines used: PC3, RC-77T/E

To complete Major Task 2, Subtask 2, Dr. Mavingire and Ms. Moore will evaluate whether HER2-OE alters cell growth compared to empty vector control. They will use CellTiter-Glo Viability assay which determines the number of viable cells by quantifying adenosine triphosphate (ATP). Briefly, cells will be plated in a 96-well plate and the CellTiter-Glo reagent added. This will be repeated at 0, 24, 48, and 72 hours. The plate will be read on a departmental microplate reader available through Morehouse School of Medicine core facilities.

Subtask 3: Evaluate HER2 overexpression on clonogenicity.

Cell lines used: PC3, RC-77T/E

To complete Major Task 2, Subtask 3, Dr. Mavingire and Ms. Moore will determine if HER2-OE alters clonogenicity. HER2-OE and empty vector control cells will be plated, and number and size of colonies will be visualized with light microscopy (10X) using equipment provided by the Morehouse School of Medicine Image Analysis Lab and quantified at 3 days with ImageJ software. Differences between empty vector control and HER2-OE cells as well as AA vs. EA cells will be analyzed using unpaired Student's t-test with GraphPad Prism 8. P values below 0.05 will be considered statistically significant.

Subtask 4: Evaluate HER2 overexpression on migration.

Cell lines used: PC3, RC-77T/E

To complete Major Task 2, Subtask 4, Dr. Mavingire and Ms. Moore will evaluate migration of empty vector control and HER2-OE cells. A wound healing assay will be performed. Cell migration will be tracked with light microscopy (10X) at 0, 24, and 48 hours, and the wound recovery rate obtained in 6 independent experiments will be measured using equipment provided by the Morehouse School of Medicine Image Analysis Lab and quantified with ImageJ

software. The wound area at 24 hours will be compared to the wound area at 0 hours for empty vector control cells and HER2-OE cells to determine the % wound recovery. Differences between empty vector control and HER2-OE cells as well as AA vs. EA cells will be analyzed using unpaired Student's t-test with GraphPad Prism 8. P values below 0.05 will be considered statistically significant.

Major Task 3: Evaluate whether HER2 blockade alters cell growth, clonogenicity, and migration.

Subtask 1: Treat cells with trastuzumab to inhibit HER2.

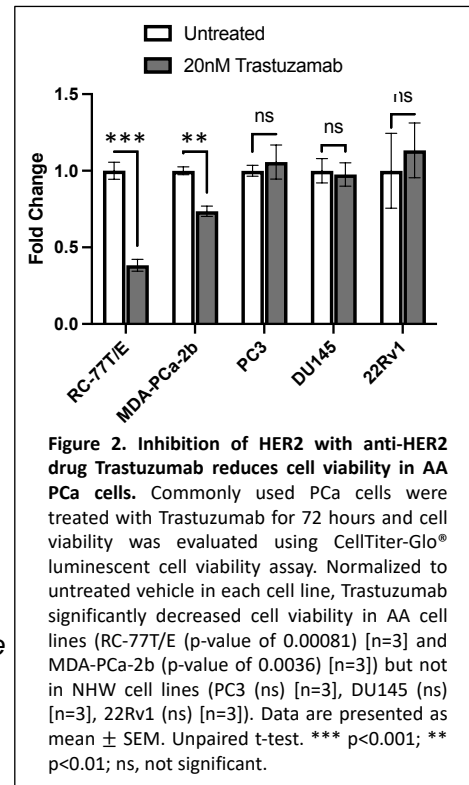
Cell lines used: PC3, RC-77T/E

We have inhibited HER2 pharmacologically with trastuzumab (Cat: 50-187-4657) reconstituted in PBS in RC-77T/E, MDA-PCa-2b, PC3, DU145, and 22Rv1 cells. To determine an optimal concentration of trastuzumab treatments, initial dose-dependent (0 nM to 5 μ M) experiments were conducted and evaluated with CellTiter-Glo Viability (Cat: PR-G7572) at 72 hrs. 20 nM trastuzumab at 72 hrs was selected for subsequent cell viability analysis. We normalized to untreated vehicle in each cell line. We observed that trastuzumab significantly decreased cell viability in AA cell lines (RC-77T/E and MDA-PCa-2b) but not in NHW cell lines (PC3, DU145, and 22Rv1) (**Figure 2**).

Subtask 2: Generate HER2 transient knockdown PC3 and RC-77T/E cell lines using siRNA.

Cell lines used: PC3, RC-77T/E

We were in the process of inhibiting HER2 genetically with small interfering RNA (siRNA) (Cat:6283) in PC3 and RC-77T/E cells. We were optimizing knockdown efficiency with a dose-dependent (50 nM to 100 nM oligo) and time-dependent (48 hr to 72 hr) experimental approach. **Due to the transition of the lab from City of Hope in Duarte, CA to Morehouse School of Medicine in Atlanta, GA, experiments for Subtask 2, 3, and 4 were temporarily halted in June 2022. Due to construction at Morehouse School of Medicine, all lab equipment and supplies have been contained in storage units in Atlanta, GA until lab space became available. The Burnham lab space became available in January 2023. Lab equipment and supplies will be moved into the Burnham lab over the next few weeks with experiments expected to resume full-time in February 2023. We will verify efficient knockdown with Western blot analysis.*



Subtask 3: Evaluate HER blockade on cell growth with CellTiter-Glo Viability assay.

Cell lines used: PC3, RC-77T/E

To evaluate whether HER2 blockade arrests cell growth, Dr. Mavingire and Ms. Moore will perform CellTiter-Glo Viability assay and compare results in untreated vs. treated cells as well as in siSD (scrambled control) cells vs. siHER2 (HER2 knockdown) cells in PC3 and RC-77T/E cells. For statistical analyses, we will use unpaired Student's t-test with GraphPad Prism 8. P values below 0.05 will be considered statistically significant.

Subtask 4: Evaluate HER2 blockade on clonogenicity.
Cell lines used: PC3, RC-77T/E

To evaluate whether HER2 blockade arrests clonogenicity, Dr. Mavingire and Ms. Moore will perform clonogenic assay and compare results in untreated vs. treated cells as well as in siSD (scrambled control) cells vs. siHER2 (HER2 knockdown) cells in PC3 and RC-77T/E cells. For statistical analyses, we will use unpaired Student's t-test with GraphPad Prism 8. P values below 0.05 will be considered statistically significant.

Subtask 5: Evaluate HER2 blockade on migration.
Cell lines used: PC3, RC-77T/E

To evaluate whether HER2 blockade arrests migration, Dr. Mavingire and Ms. Moore will perform wound healing assay and compare results in untreated vs. treated cells as well as in siSD (scrambled control) cells vs. siHER2 (HER2 knockdown) cells in PC3 and RC-77T/E cells. For statistical analyses, we will use unpaired Student's t-test with GraphPad Prism 8. P values below 0.05 will be considered statistically significant.

Specific Aim 2: Confirm whether HER2 overexpression is positively correlated with WAA in tumor tissue specimens.

Major Task 1: Access racially diverse tumor tissue specimens.

Subtask 1: Complete IRB approval to access tumor tissue specimens.

We received IRB (Protocol 20619) approval from City of Hope (COH) (see below) to access tumor tissue specimens for this study from the COH Comprehensive Cancer Center (CCC) Frozen Tissue Bank as well as the Department of Defense (DOD) Prostate Cancer Biorepository Network (PCBN).

**Institutional Review Board
Notification of Determination that Activity is Not Human Subjects Research**

Date: November 04, 2020

To: Leanne Burnham, Ph.D, Principal Investigator
City of Hope - Cncr Genetics - Epigenetics-BRI

From: Milda Plioplys, Director *Milda Plioplys*
Clinical Research Protections

COH Protocol #/Ref #: 20619 / 199945

Protocol Title: HER2 expression in men with prostate cancer

Regulatory Sponsor: City of Hope, U.S. Department of Defense (DOD)

Sponsor #:

Action Date: 11/03/2020

Action: NOT HUMAN SUBJECTS RESEARCH

The information provided for the above submission was evaluated and determined to not meet the definition of human subjects research as set forth at (45 CFR 46.102 (e)). Accordingly, IRB approval and continuing review is not required.

Please note that if any changes occur on this project, notification to the IRB is required to determine if the status of this project should be updated to be under the purview of the IRB.

Please note the following:

1. If this project is receiving funding from the Department of Defense (DOD), it will require an additional determination made by their Human Research Protections Office (HRPO) before proceeding by submitting to dha.ncr.dha-cs-mgt.mbx.hrpp@mail.mil.
2. The IRB protocol application form has been corrected. The participation of Antelope Valley and Santa Clarita have been removed as these two sites are not approved by COH to begin research at this time.

If you have questions or concerns about this submission, please contact Gwen Jorgensen or IRBeSubmit@coh.org.

Subtask 2: Access FFPE.

FFPE: 400 patients (100 biopsy negative, 100 lymph node negative, 100 lymph node positive, 100 mCRPC) [COHCCC Frozen Tissue Bank]

COH Honest Broker/Research Informatics identified a dataset of clinical FFPE specimens from the COH Comprehensive Cancer Center (CCC) Frozen Tissue Bank previously collected from an equal amount Black/AA and non-Hispanic white male patients at different disease stages (Group 1: normal prostate [normal prostate removed as part of cystoprostatectomy]; Group 2: prostate cancer, prostatectomy, and TNM status of T any, N0, Mx; Group 3: prostate cancer, prostatectomy, and TNM status of T any, N1, Mx; and Group 4: diagnosis of prostate cancer where tissue is NOT prostate, tissue might be bone or lymph node, lung or liver). FFPE specimens for Groups 1, 2, and 3 were pulled by COH Pathology Core for IHC analysis. FFPE specimens for Group 4 are currently being pulled by COH Pathology Core for IHC analysis.

Subtask 3: Access TMA and patient-matched DNA.

TMA: 456 Case Race Disparity TMA [DOD]

We received approval from the DOD PCBN and accessed the 456 Case Race Disparity tissue microarray (TMA) (please see below) as well as patient-matched DNA. As an alternative to purchasing the patient-matched DNA, the DOD PCBN negotiated with Washington University in St. Louis to be able ship patient-matched frozen cell pellets to me to extract genomic DNA. An MTA agreement between Washington University in St. Louis and COH was settled (please see below), and 456 frozen cell pellets were shipped to my lab (Date of Receipt 12/08/21).



PROSTATE CANCER BIOREPOSITORY NETWORK

Tissue, Fluids, and Derived Biospecimens Request Application Form
Form No: 001

Application #

SECTION A: Applicant Details		
Principal Investigator (PI) Name Leanne Burnham, Ph.D.	PI Telephone 626-256-4673 Ext. 81631	PI Email lburnham@coh.org
Institution City of Hope Comprehensive Cancer Center		Department Population Sciences
Institution Address 1500 E. Duarte Rd., Duarte, CA 91010 Rick Kittles Laboratory Shapiro Building 096 Room 1038 Attn: Leanne Burnham,		Institution Type <input type="checkbox"/> Academic/Government <input type="checkbox"/> Commercial <input checked="" type="checkbox"/> Non-profit
Contact Person Leanne Burnham, Ph.D.	Contact Telephone 626-256-4673 Ext. 81631	Contact Email lburnham@coh.org
Legal Contact Person (MTA purposes) Sylvia Campos	Legal Contact Telephone (MTA purposes) 626-256-4673 Ext. 81811	Legal Contact Email (MTA purposes) scampos@coh.org

SECTION B: Billing Information		
Billing Contact Person Claudio Carbajal	Billing Contact Telephone 626-256-4673 Ext. 85488	Billing Contact Email ccarbajal@coh.org
Billing Address 1500 E. Duarte Rd. Duarte, CA 91010	<input type="checkbox"/> Same as Institution Address	Payment Details <input checked="" type="checkbox"/> Purchase Order <input type="checkbox"/> Credit Card <input type="checkbox"/> Other
Funding Source Department of Defense Prostate Cancer Research Program Early Investigator Award	Grant ID # W81XWH2110038	Grant End Date 31 Dec 2022

SECTION C: Project Information
Project Title HER2 Expression in African American Men with Prostate Cancer
Hypothesis HER2 overexpression is positively correlated with West African ancestry and contributes to worse clinical features, treatment response, and survival outcomes.

<p>Specific Aims</p> <p>Aim 1: Establish that HER2 expression promotes cellular growth, clonogenicity, and migration in a racially diverse panel of prostate cancer cell lines.</p> <p>Aim 2: Confirm whether HER2 overexpression is positively correlated with West African ancestry in tumor tissue specimens.</p> <p>Aim 3: Determine if HER2 overexpression correlates with disease stage, clinical features, treatments, and outcomes in prostate cancer patients.</p>		
<p>Protocol / Method to be used</p> <p>Aim 1: Establish that HER2 expression promotes cellular growth, clonogenicity, and migration in a racially diverse panel of prostate cancer (PCa) cell lines. Step 1.1 Quantify basal HER2 gene and protein levels in a racially diverse panel of cell lines. Step 1.2 Evaluate whether HER2 overexpression differentially alters cell growth, clonogenicity, and migration. Step 1.3 Evaluate whether HER2 blockade differentially arrests cellular growth, clonogenicity, and migration.</p> <p>Aim 2: Confirm whether HER2 overexpression is positively correlated with West African ancestry (WAA) in tumor tissue specimens. Step 2.1 Access racially diverse tumor tissue specimens from the COHCCC Frozen Tissue Bank and the Department of Defense Prostate Cancer Biorepository Network (DOD PCBN). We will access FFPE primary PCa specimens from the COHCCC Frozen Tissue Bank previously collected from a racially/ethnically diverse cohort of male patients at different disease stages (Group 1: biopsy negative [50 African American (AA), 50 European American (EA)], Group 2: lymph node negative at radical prostatectomy [50 AA, 50 EA], Group 3: lymph node positive at radical prostatectomy (RP) [50 AA, 50 EA], and Group 4: mCRPC [50 AA, 50 EA]). We will also access the 459 Case Race Disparity tissue microarray (TMA) (153 AA, 306 EA) collected at RP through the DOD PCBN. Step 2.2 Quantify HER2 gene and protein expression in tumor tissue specimens. We will conduct IHC membrane staining on City of Hope Comprehensive Cancer Center (COHCCC) FFPE and DOD PCBN TMA specimens with the assistance of COHCCC Pathology Core. Tumors with absent or weak membrane HER2 staining (scored as 0 or 1+) will be regarded as HER2 negative. Tumors with intense and complete membrane HER2 staining (scored as 3+) will be regarded as HER2+. Tumors with moderate membrane HER2 staining (scored as 2+) will be regarded as HER2 equivocal and reflexively tested by fluorescent in situ hybridization (FISH) with the assistance of COHCCC Pathology Core to assess for HER2 gene amplification. A dual-probe HER2/ chromosome enumeration probe 17 (HER2/CEP17) ratio >2.0 or average HER2 copy number >6.0 signals per cell will be used to classify tumors as HER2+. Data generated from each self-reported race/ethnicity will allow for detection of an effect size of 0.249 at 80% power (t-test, $\alpha = 0.05$) for intra- and inter-group differential expression analysis of 200 FFPE specimens and 0.246 at 80% power (t-test, $\alpha = 0.05$) for 456 Case Race Disparity TMA specimens. Step 2.3 Calculate genetic ancestry proportion of study patients' DNA using validated ancestry informative markers (AIMs) through single nucleotide polymorphism (SNP) genotyping. To calculate genetic ancestry, we will obtain patient-matched DNA from COHCCC and DOD PCBN (Step 2.1). We will use a panel of AIMs developed to determine proportions of West African, European, and Indigenous American ancestry and to distinguish populations of different biogeographic origins. We will genotype isolated genomic DNA using the Sequenom MassARRAY platform with iPLEX chemistry. Individual admixture estimates will be calculated using a model-based clustering method as implemented in STRUCTURE 2.3. Step 2.4 Compare HER2 expression to WAA proportion in the AA tumor tissue specimens. For statistical analyses, we will conduct linear regression with an 80% power to detect an effect size of 0.027 ($\alpha = 0.05$) to determine the relationship between HER2 expression (Step 2.2) and WAA (Step 2.3) in the 353 (200 FFPE, 153 TMA) (Step 2.1) AA tumor tissue specimens.</p> <p>Aim 3: Determine if HER2 overexpression correlates with disease stage, clinical features, treatments, and outcomes in PCa patients. Step 3.1 Stratify tumor tissue specimens by disease stage, clinical features, treatments, and outcomes. We will stratify FFPE primary PCa specimens from the COHCCC Frozen Tissue Bank (Step 2.1) by disease stage (Group 1: biopsy negative [100], Group 2: lymph node negative at radical prostatectomy ([100], Group 3: lymph node positive at radical prostatectomy [100], and Group 4: mCRPC [100]). The Institutional Review Board (IRB)-approved biobank includes clinical annotation; data will be abstracted related to relapse and survival. For the 456 Case Race Disparity TMA obtained through the DOD PCBN, we will request available corresponding matched clinical data upon approval for similar analysis. Step 3.2 Evaluate HER2 expression correlation with disease stage, clinical features, treatment, and outcomes of PCa patients. For statistical analyses, we will conduct logistic regression and multivariable Cox regression with $\alpha = 0.05$ to determine the relationship between HER2 overexpression (Step 1.2) and disease stage (Step 3.1) as well as clinical features (i.e. PSA, Gleason score, lymph node status), treatments (i.e. primary hormone therapy and radiation), and outcomes (i.e. biochemical recurrence and overall survival) of PCa patients.</p>		
<p>Name of Pathologist associated with the study (if TMA or tissue slides require microscopic examination)</p> <p style="text-align: center;">Zhirong Yin, M.D.</p>		
<p>IRB Approval Type</p> <p><input checked="" type="checkbox"/> Full <input type="checkbox"/> Expedited <input type="checkbox"/> Exempt</p>	<p>IRB Approval Number</p> <p style="text-align: center;">20619</p>	<p>IRB Approval Dates</p> <p style="text-align: center;">Nov. 4, 2020 Apr. 2, 2021</p>

SECTION D: Biospecimen Information		
Biospecimen Type	# Cases	Justification for # Cases Requested (Statistical Power Analysis)
<p>Patient-matched DNA (matched to 456 Case Race Disparity TMA)</p>	<p>456</p>	<p>Data generated from each self-reported race/ethnicity will allow for intra- and inter-group differential expression analysis for HER2 detection of an effect size of 0.246 at 80% power (t-test, $\alpha = 0.05$) for 456 Case Race Disparity TMA specimens. For statistical analyses, we will conduct linear regression with an 80% power to detect an effect size of 0.027 ($\alpha = 0.05$) to determine the relationship between HER2 expression (Step 2.2) and WAA (Step 2.3) in the 353 (200 FFPE, 153 TMA) (Step 2.1) AA tumor tissue specimens.</p>

Case Characteristics (optional information if you have specific requirements)

Procedure Type									
<input type="checkbox"/> Radical Retropubic Prostatectomy <input type="checkbox"/> Robot-Assisted Prostatectomy <input type="checkbox"/> Biopsy <input type="checkbox"/> Transurethral Resection <input type="checkbox"/> Autopsy									
Age at Surgery		Neo-adjuvant Treatment		Adjuvant Treatment		pStage		Gleason Score	
Min	Max	<input type="checkbox"/> Yes	<input type="checkbox"/> No	<input type="checkbox"/> Yes	<input type="checkbox"/> No	Min	Max	Min	Max


Sample Preparation Details (optional information if you have specific requirements)

For Tissue			
<input type="checkbox"/> FFPE Section	<input type="checkbox"/> Frozen Section	<input type="checkbox"/> Scroll / Ribbon	<input type="checkbox"/> Macrodissection
<input type="checkbox"/> Core	<input type="checkbox"/> Touch Prep	<input type="checkbox"/> Other	
<input type="checkbox"/> Malignant	<input type="checkbox"/> Benign	<input type="checkbox"/> Normal	<input type="checkbox"/> Tumor /Benign Pairs
<input type="checkbox"/> Other			
For Body Fluids			
Biospecimen Minimum Volume			
		<input type="checkbox"/> mL	<input type="checkbox"/> uL
For Derived Biospecimens (DNA/RNA/Protein)			
Origin			
<input type="checkbox"/> Frozen Tissue	<input type="checkbox"/> Fixed Tissue	<input type="checkbox"/> Seminal Vesicle	<input type="checkbox"/> Prostatic Fluid
<input type="checkbox"/> Plasma	<input type="checkbox"/> Serum	<input checked="" type="checkbox"/> Buffy Coat	<input type="checkbox"/> Seminal Vesicle Fluid
Biospecimen Minimum Amount (e.g. 100 ng total RNA)			
800ng genomic DNA at 10ng/uL			
Additional Requirements			

SECTION E: Shipping Information

Shipping Address	<input checked="" type="checkbox"/> Same as Institution Address
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We/I have read, understood and agree with the Tissue Access Policy and Conditions of Use for Tissue And Data Bank Resources. We/I agree that the samples provided by PCBN will be used for the research work detailed in the attached proposal. The material will not be used for other studies, or distributed to third parties. Tissues and their products will not be used for commercial purposes. We/I realize that there is the potential that this human biological material may contain infectious agents and, therefore, will handle it appropriately.

Principal Investigator Signature 	Date 9-15-21	Submit
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**Letter Agreement for the Transfer of Human Specimens
The Washington University**

Whereas, The Washington University, One Brookings Drive, St. Louis, MO, 63130, USA (PROVIDER) possesses coded human tissue samples (MATERIAL); and

Whereas, the non-profit institution listed below (RECIPIENT) has requested from PROVIDER's investigator, Dr. Bettina Drake, the MATERIAL further described as: Frozen Cell Pellets (200um) from 456 Cases from Race Disparity TMA and HER2 IHC data on the TMA set for use in the following not-for-profit research project (PROJECT):

This pilot study is the first to investigate HER2 (ERBB2) and androgen receptor (AR/NR3C4) status in mCRPC patients inclusive of African American men. We anticipate that circulating HER2 and AR as well as ERBB2 and NR3C4 gain will be increased in African American men. Successful completion of this study will generate preliminary data to support applications for future funding to 1) fully evaluate whether HER2 contributes to the worse prognosis experienced by African American men and 2) to develop more effective treatment strategies for advanced PCa patients with increased HER2 and AR.

In response to RECIPIENT's request for the MATERIAL the PROVIDER and the RECIPIENT agree to the following before the RECIPIENT receives the MATERIAL:

1. The above MATERIAL is the property of the PROVIDER and is made available as a service to the research community. MATERIAL provided pursuant to this Agreement was collected or will be collected in accordance with the standard patient informed consent procedures of the PROVIDER in effect at the time of collection and subject to approval by the PROVIDER's IRB. MATERIAL provided to RECIPIENT by PROVIDER will not contain personally identifiable patient information and will not include "Protected Health Information" ("PHI") as defined in 45 C.F.R. Section 160.103. The parties agree that the human data and/or biospecimens transferred under this Agreement may be subject to NIH Policy NOT-OD-17-109 (the "Policy") and therefor is deemed under the Policy to be issued a Certificate of Confidentiality. Accordingly the RECIPIENT is required to adhere to the Policy and protect the privacy of the individuals from whom the data and/or biospecimens were collected in accordance with the Policy and subsection 301(d) of the Public Health Service Act.
2. THIS MATERIAL IS NOT FOR USE IN HUMAN SUBJECTS.
3. The MATERIAL will be used for not-for-profit research purposes in conduct of the above PROJECT only.
4. The MATERIAL will not be further distributed to others, outside of RECIPIENT's Scientist and RECIPIENT'S employees, medical staff, agents and representatives having a need for access to the MATERIAL in order to conduct the PROJECT, without the PROVIDER'S written consent. The RECIPIENT shall refer any request for the MATERIAL to the PROVIDER.
5. The RECIPIENT agrees to acknowledge the source of the MATERIAL in any publications and presentations reporting use of the MATERIAL.
6. The RECIPIENT agrees that it will not have or seek access to any identifiable information (such as the key to the code) under any circumstances. The RECIPIENT agrees to never use the MATERIAL or any substance derived from the MATERIAL (e.g. DNA, RNA or any information provided with the MATERIAL) to attempt to ascertain the identity of the individual from whom the MATERIAL was obtained.
7. Any MATERIAL delivered pursuant to this Agreement is understood to be experimental in nature and may have hazardous properties and may carry transmissible infectious agents. THE PROVIDER MAKES NO REPRESENTATIONS AND EXTENDS NO WARRANTIES OF ANY KIND, EITHER EXPRESSED OR IMPLIED. THERE ARE

NO EXPRESS OR IMPLIED WARRANTIES OF MERCHANTABILITY OR FITNESS FOR A PARTICULAR PURPOSE, OR THAT THE USE OF THE MATERIAL WILL NOT INFRINGE ANY PATENT, COPYRIGHT, TRADEMARK, OR OTHER PROPRIETARY RIGHTS. Unless prohibited by law, RECIPIENT assumes all liability for claims for damages against it by third parties which may arise from the use, storage or disposal of the MATERIAL except that, to the extent permitted by law, the PROVIDER shall be liable to the RECIPIENT when the damage is caused by the gross negligence or willful misconduct of the PROVIDER.

- 8. The RECIPIENT agrees to use the MATERIAL in compliance with all applicable U.S. federal, California state and local statutes and regulations.
- 9. The MATERIAL is provided according to the amount indicated in the provided PCBN Cost Recovery Structure document and the RECIPIENT will be further responsible for all shipping costs.

The RECIPIENT and RECIPIENT SCIENTIST must sign this letter and return one signed copy to the PROVIDER. Upon receiving this fully executed agreement, the PROVIDER will then send the MATERIAL.

RECIPIENT INFORMATION and AUTHORIZED SIGNATURE


Recipient Scientist: Dr. Leanne Burnham

Recipient Organization: City of Hope National Medical Center and Beckman Research Institute of the City of Hope, each a California nonprofit, public benefit corporation.


Address: 1500 E. Duarte Rd. Duarte, CA 91010

Name of Authorized Official: Benjamin Diamond

Title of Authorized Official: Senior Contract Administrator

Signature of Authorized Official:  Date: 12/2/2021

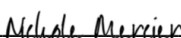
Certification of Recipient Scientist: Although not a legal party to this Agreement, I have read and understood the conditions outlined in this Agreement.

Recipient Scientist:  Date: December 2, 2021

PROVIDER AUTHORIZED SIGNATURE

Name of Authorized Official: Nichole R. Mercier, PhD

Title of Authorized Official: Assistant Vice Chancellor & Managing Director

Signature of Authorized Official:  Date: December 2, 2021

Major Task 2: Quantify HER2 in tumor tissue specimens.

Subtask 1: Perform IHC to detect HER2.

FFPE: 400 patients [COHCCC Frozen Tissue Bank]

TMA: 456 Case Race Disparity TMA [DOD]

The COHCCC Pathology Core will perform HER2 IHC membrane staining on COHCCC FFPE specimens that have been identified and recently pulled. Dr. Zhirong Yin is the research pathologist who will stain and score the COH specimens. Washington University in St. Louis has completed HER2 IHC membrane staining and scoring of the DOD PCBN TMA specimens are currently being analyzed by Dr. Mavingire and Dr. Deyana Lewis at Morehouse School of Medicine. Both COH and Washington University in St. Louis will use and have used the Ventana staining protocol optimized and drafted by Dr. Yin (please see below). Data generated from each self-reported race/ethnicity will allow for detection of an effect size of 0.249 at 80% power (t -test, $\alpha = 0.05$) for intra- and inter-group differential expression analysis of 400 FFPE specimens and 0.246 at 80% power (t -test, $\alpha = 0.05$) for 456 Case Race Disparity TMA specimens.

**Pathology Research Services Core
Beckman Research Institute of City of Hope**

HER2 Antibody VALIDATION REPORT:

Antibody: Her 2	Species: Rabbit Monoclonal	Clone: 4B5
Titer/Concentration: pre-diluted dispenser	Catalog #: 790-2991	Localization: membrane

Status & validation from Ventana:

The Ventana Medical Systems, Inc.'s (Ventana) PATHWAY anti-HER-2/*neu* (4B5) is a FDA/CE-IVD approved HER2 IHC assays, and it has been used as reference standard method. PATHWAY anti-HER-2/*neu* is a rabbit monoclonal antibody (clone 4B5) against the internal domain of the c-erbB-2 oncoprotein (HER2).

Clone 4B5 has been shown to react with a 185 kD protein from SK-BR-3 cell lysates via Western blotting. SK-BR-3 is a breast carcinoma cell line, which has a 128-fold over expression of HER2 mRNA. The size of the band identified correlates well with that reported by Akiyama et al for HER2 protein (185 kD). Staining results in normal tissues, neoplastic tissues, and 322 cases of breast carcinoma with PATHWAY HER2 (4B5) were evaluated by Ventana.

Material and Methods:

All the specimen was fixed for 24-48 h in 10% neutral buffered formalin. The paraffin embedded tissue sections in 4µM thickness were prepared in Pathology Core at City of Hope.

PATHWAY anti-HER-2/*neu* (4B5) was used to detect HER2 antigen in sections of formalin-fixed, paraffin-embedded normal and prostate cancer tissue on the VENTANA Discovery Ultra automated immunohistochemistry slide staining device. PATHWAY HER2 (4B5) antibody binds to HER2 in paraffin-embedded tissue sections. The specific antibody can be localized by a HQ conjugated secondary antibody formulation that recognizes rabbit immunoglobulins followed by the addition of a secondary antibody-HRP conjugate. The specific antibody-enzyme complex is then visualized with a precipitating enzyme reaction product (DISCOVERY ChromoMap DAB Kit). Each step is incubated for a precise time and temperature. At the end of each incubation step, the VENTANA automated slide stainer washes the sections to stop the reaction and to remove unbound material that would hinder the desired reaction in subsequent steps. The detail of protocol is as the following:

Ventana Discovery Ultra HER2 IHC staining protocol

1. Load slides and reagents onto Ventana Discovery Ultra stainer.
2. Deparaffinization in EZ prep 69°C 24 minutes.
3. Cell Conditioning using Conditioner #1, Standard CC1, 95°C 64 minutes.
4. Block with Inhibitor CM, 37°C 8 minutes.
5. Incubation with HER2 primary antibody for 16 minutes.
6. Apply one drop of Discovery anti-Rb HQ and incubate for 12 minutes.
7. Apply one drop of Discovery anti-HQ HRP and incubate for 12 minutes.
8. Apply DAB kit including DAB CM, H₂O₂ CM and Copper CM, incubate 8 minutes.
9. Counterstain with Hematoxylin, incubate for 16 minutes.
10. Post counterstain with Bluing Reagent, incubate for 4 minutes.
11. Slide Cleaning.
12. Coverslipper.

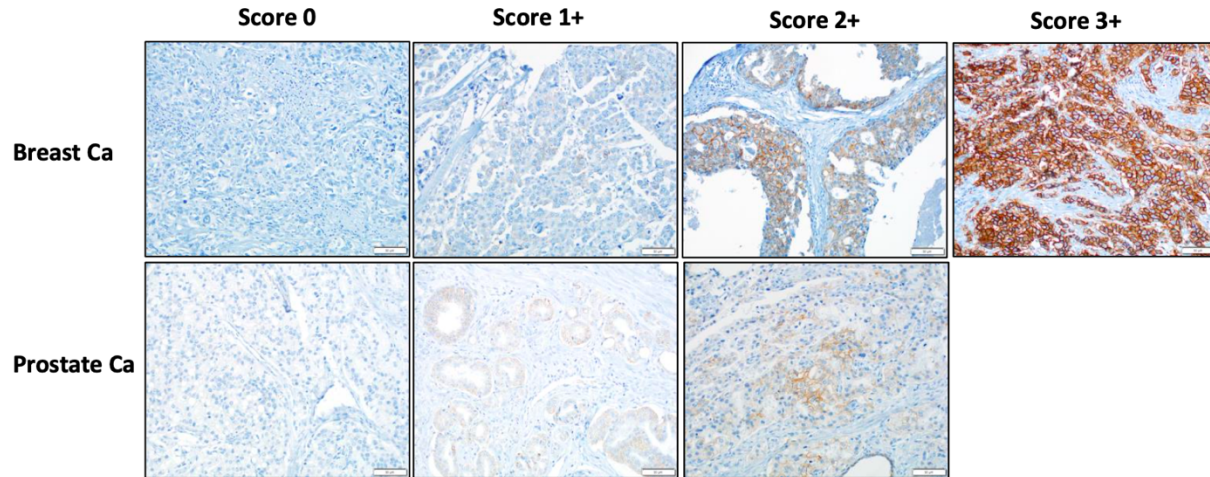
Quality Control

A TMA consisting of 31 tissues of breast cancer with known positive HER2 expression, serving as positive control tissue and 7 tissues of normal breast, serving as negative control tissue used for a preliminary validation of the processing method used for staining slides with PATHWAY HER2 (4B5), and for performance controls across staining runs. A negative reagent control was also run for every specimen to aid in the interpretation of results, in which CONFIRM Negative Control Rabbit Ig is used in place of the primary antibody to evaluate nonspecific staining.

In addition, another TMA was used for testing the PATHWAY HER2 (4B5) specificity. The results showed no specific membrane staining for tonsil (1/4, focal staining of surface epithelial cells), and weak membrane staining on lung cancer (1/2) and pancreatic cancer (1/2).

Scoring of PATHWAY HER2 (4B5) on representative images

Score	ER2 Staining Assessment	Staining Pattern
0	Negative	No membrane staining is observed
1+	Negative	Faint, partial staining of the membrane in any proportion of the cancer cells
2+	Weakly Positive	Weak complete staining of the membrane, greater than 10% of cancer cells
3+	Positive	Intense complete staining of the membrane, greater than 10% of cancer cells



Zhirong Yin, MD, Ph.D

Research Pathologist
 Pathology Cores & Biobanking| Shared Resources
 City of Hope
Office: Familian Sciences Bldg. (#84), Lower level-L001F
Phone: (626) 218- 6840 (ext. 86840)
Email: zhyin@COH.org
 1500 East Duarte Rd. Duarte, CA 91010

Subtask 2: Perform FISH to confirm overexpression in HER2 equivocal samples.
 FFPE: 400 patients [COHCCC Frozen Tissue Bank]

Tumors with moderate membrane HER2 staining (scored as 2+) will be regarded as HER2 equivocal and reflexively tested by fluorescent in situ hybridization (FISH) with the assistance of COHCCC Pathology Core to assess for HER2 gene amplification. A dual-probe HER2/chromosome enumeration probe 17 (HER2/CEP17) ratio >2.0 or average HER2 copy number >6.0 signals per cell will be used to classify tumors as HER2+. Data generated from each self-reported race/ethnicity will allow for detection of an effect size of 0.249 at 80% power (*t*-test, $\alpha = 0.05$) for intra- and inter-group differential expression analysis of 200 FFPE specimens.

Major Task 3: Calculate genetic ancestry of patients.

Subtask 1: Extract genomic DNA.
 FFPE: 400 patients [COHCCC Frozen Tissue Bank]
 TMA: 456 Case Race Disparity TMA frozen cell pellets [DOD]

We extracted and quantified genomic DNA from frozen cell pellets which were patient-matched with the 456 Case Race Disparity TMA from DOD PCBN (**see Appendix**). Dr. Mavingire and Ms. Moore will be extracting genomic DNA from FFPE specimens accessed from the COHCCC Frozen Tissue Bank. For genomic DNA extraction, we used the Invitrogen PureLink Genomic DNA Kit (Cat: K1820-01).

Subtask 2: SNP genotyping using a panel of validated AIMS.
FFPE: 400 patients [COHCCC Frozen Tissue Bank]
DNA: 459 Case Race Disparity TMA [DOD]

To calculate genetic ancestry, Dr. Rick Kittles and Dr. Jabril Johnson at Morehouse School of Medicine are using a panel of ancestry informative markers (AIMs) developed to determine proportions of West African, European, and Indigenous American ancestry and to distinguish populations of different biogeographic origins¹². They genotyped isolated genomic DNA using the Sequenom MassARRAY platform with iPLEX chemistry. Individual admixture estimates are calculated using a model-based clustering method as implemented in STRUCTURE 2.3.

Subtask 3: Compare HER2 expression to WAA.

For statistical analyses, Dr. Kittles, Dr. Johnson, and Dr. Lewis will conduct linear regression with an 80% power to detect an effect size of 0.027 ($\alpha = 0.05$) to determine the relationship between HER2 expression and WAA in the 553 (400 FFPE, 153 TMA) AA tumor tissue specimens.

Specific Aim 3: Determine HER2 correlation with PCa disease stage, clinical features, treatments, and outcomes in patients.

Major Task 1: Stratify tumor tissue specimens.
FFPE: 400 patients [COHCCC Frozen Tissue Bank]
TMA: 459 Case Race Disparity TMA [DOD]

FFPE PCa specimens from the COHCCC Frozen Tissue Bank have been stratified by COH Honest Broker/Research Informatics by disease stage (Group 1: normal prostate [normal prostate removed as part of cystoprostatectomy]; Group 2: prostate cancer, prostatectomy, and TNM status of T any, N0, Mx; Group 3: prostate cancer, prostatectomy, and TNM status of T any, N1, Mx; and Group 4: diagnosis of prostate cancer where tissue is NOT prostate, tissue might be bone or lymph node, lung or liver). The IRB-approved biobank includes clinical annotation; data will be abstracted related to relapse and survival. For the 456 Case Race Disparity TMA obtained through the DOD PCBN, corresponding matched clinical data has been made available to my lab.

Major Task 2: Correlate HER2 expression with disease stage, clinical features, treatments, and outcomes.

FFPE: 400 patients [COHCCC Frozen Tissue Bank]
TMA: 459 Case Race Disparity TMA [DOD]

For statistical analyses, Dr. Lewis will conduct logistic regression and multivariable Cox regression with $\alpha = 0.05$ to determine the relationship between HER2 overexpression and disease stage as well as clinical features (i.e., PSA, Gleason score, lymph node status), treatments (i.e., primary hormone therapy and radiation), and outcomes (i.e., biochemical recurrence and overall survival) of PCa patients at COHCCC. Dr. Mavingire has begun

preliminary analyses to correlate HER2 expression with disease stage, clinical features, treatments, and outcomes.

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

This funding opportunity as a DOD Prostate Cancer Research Program Early Investigator has greatly contributed to my professional development. In May 2021, I accepted a new appointment to Assistant Research Professor in the Department of Population Sciences Division of Health Equities at City of Hope. This funding opportunity has allowed me to transition to an independent tenure-track faculty position. In January 2022, I accepted a new appointment as assistant professor in the Department of Population Sciences Division of Health Equities at City of Hope. In the summer of 2022, I relocated my lab to Atlanta, GA upon my acceptance of a tenure-track appointment within the department of physiology at Morehouse School of Medicine. Because of my progress and continued success of my research program, I was recently awarded a National Institutes of Health National Cancer Institute Health Disparities Research Loan Repayment Program Award.

In terms of my training development, I have learned the intricacies of the IRB review process. I learned how to draft protocols, present the study, and receive endorsement from COH's Genitourinary Disease Team, and respond to IRB requests to ensure full approval. I also learned how to navigate the process of identifying samples through COH Honest Broker/Research Informatics as well as how to engage COH's Pathology Core for collaborative purposes. I have extended my collaborative network within the Morehouse School of Medicine Alliance and am currently expanding my research program in partnership with Emory University, Grady Health, and O'Neal Comprehensive Cancer Center. I meet monthly to share study results and discuss goals with my mentors, Dr Rick Kittles and Dr. Tanya Dorff. I also meet with Dr. Sean Kimbro twice a year for specific feedback and direction regarding the molecular biology techniques specific to this study.

Utilizing the combined skills I have learned as an Early Investigator, I was able to serve as an ad-hoc member of the NCI Connecting Underrepresented Populations to Clinical Trials (CUSP2CT) Review Panel. I was able to implement the grantsmanship and experimental design approaches I have been exposed to under the mentorship that the Prostate Cancer Research Program endorses and requires.

In terms of training development of others who worked on the project, I was able to hire a lab intern in August 2021 named Serene Dowiri. Ms. Dowiri has been able to learn and execute many wet lab techniques including cell culture, lysate collection, CellTiter-Glo Viability assay, genomic DNA extraction, and quantification of DNA using NanoDrop. In addition, I have hired and currently mentor a postdoctoral fellow, Dr. Nicole Mavingire, as well as a graduate student, Ms. Janelle Moore.

How were the results disseminated to communities of interest?

An overview of the goals of this study supported by this funding opportunity was disseminated to several communities of interest in virtual webinars including the following:

02/23/21 "Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community", OncLive State of the Science Summit (Sponsored by City of Hope)

- 04/15/21 “What is Special About Research at City of Hope”, Building Hope Toast of Hope Event, City of Hope
- 06/30/21 “Dine in for Hope”, City of Hope
- 07/31/21 “Clinical Trials: An Answer to Cancer?”, Cynthia Perry Ray Foundation Sistahs Can We Talk Monthly Nurses Series (Sponsored by City of Hope)
- 09/18/21 “Prostate Cancer in Black Men: The Latest in Screening and Treatment Options”, Prostate Cancer Foundation & The West Angeles Church of God in Christ
- 09/23/21 “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, City of Hope Community Engagement Community Advisory Board
- 12/10/21 “HER2 Expression in African American Men with Prostate Cancer”, City of Hope Health Equities Faculty Candidate Seminar
- 05/04/22 “Patient and Healthcare Provider Education on Advancing Inclusive Research”, Roche/Genentech AIR Site Alliance, San Francisco, CA
- 05/12/22 “Health Disparities Research: Viewpoints from a Survivor”, Universal Music Publishing Group and City of Hope Music, Film, and Entertainment Industry, Los Angeles, CA
- 05/24/22 “Health Disparities Research: Viewpoints from a Survivor”, Universal Music Publishing Group and City of Hope Music, Film, and Entertainment Industry, New York, NY
- 06/11/22 “Increasing Diversity in Clinical Trials: Multiple Solutions to Achieve Clinical Trial Equity”, ENDO 2022 Annual Conference, Atlanta, GA
- 06/25/22 “Advancing Inclusive Research at the Bench”, 2022 Coffey-Holden Prostate Cancer Academy Meeting, UCLA, Los Angeles, CA
- 09/12/22 “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, University of California Los Angeles, Los Angeles, CA
- 11/15/22 “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, Emerging 100 of Atlanta General Body Meeting, Atlanta, GA
- 10/28/22 “HER2 expression in African American men with prostate cancer”, 29th Prostate Cancer Foundation Annual Scientific Retreat

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

During the final reporting period, I plan on completing the following underlined tasks with the methodologies described previously.

Specific Aim 1: Establish that HER2 expression promotes cellular growth, clonogenicity, and migration in a racially diverse panel of PCa cell lines.

Major Task 1: Quantify basal HER2 gene and protein levels.

Target Dates: 1-24 months

Percentage of Completion: 50%

Major Task 2: Evaluate whether HER2 overexpression alters cell growth, clonogenicity, and migration.

Target Dates: 12-24 months

Percentage of Completion: 0%

Major Task 3: Evaluate whether HER2 blockade alters cell growth, clonogenicity, and migration.

Target Dates: 9-24 months

Percentage of Completion: 40%

Specific Aim 2: Confirm whether HER2 overexpression is positively correlated with WAA in tumor tissue specimens.

Major Task 1: Access racially diverse tumor tissue specimens.

Target Dates: 1-15 months

Percentage of Completion: 100%

Major Task 2: Quantify HER2 in tumor tissue specimens.

Target Dates: 12-24 months

Percentage of Completion: 100%

Major Task 3: Calculate genetic ancestry of patients.

Target Dates: 12-18 months

Percentage of Completion: 60%

Specific Aim 3: Determine HER2 correlation with PCa disease stage, clinical features, treatments, and outcomes in patients.

Major Task 1: Stratify tumor tissue specimens.

Target Dates: 12-24 months

Percentage of Completion: 100%

Major Task 2: Correlate HER2 expression with disease stage, clinical features, treatments, and outcomes.

Target Dates: 12-24 months

Percentage of Completion: 50%

We plan on presenting partial results of this study at the American Association for Cancer Research Annual Meeting 2023 as well as full results at the 16th American Association for Cancer Research Conference on The Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved. In addition, we plan on submitting peer-reviewed publications within 120 calendar days of the award performance end date.

REPORTABLE OUTCOMES

IMPACT

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

Metastatic prostate cancer (PCa) is a lethal disease and therapeutic options are limited for patients. Since there is no cure for advanced PCa, the pursuit of therapeutic strategies to

effectively treat this stage remains a critical goal. The ability to target non-androgenic growth factor signaling pathways that may promote metastasis represents a tangible and promising area of investigation. With the development of targeted therapeutic options optimized for a molecularly enriched population, there is potential to improve survival outcomes for men at high-risk for an advanced stage diagnosis. By providing an additional option to the current arsenal of available treatments, there is an opportunity to provide hope to late-stage patients and their families.

We need to better understand the biology contributing to the worse prognosis observed in African American (AA) PCa patients in order to 1) predict which men are more likely to suffer from aggressive disease and 2) develop more patient-specific treatments. Because the majority of biospecimens and patients evaluated in research studies are of European descent, we miss potential genetic indicators that steer molecular mechanisms, disease outcomes, and drug response.¹³ This experimental approach is counterintuitive as AA men are more likely to suffer disproportionate PCa incidence and mortality, but are less likely to be included in key studies and clinical trials. Since recent studies have revealed better treatment response and increased overall survival in AA men receiving PCa drug treatments, there is additional incentive to decrease the current racial disparity in mortality with precision medicine.

Given that our preliminary evidence suggests a potential increase in HER2 overexpression in AA men with PCa, we predict that AA patients with metastatic disease may especially benefit from HER2-targeted treatment. This study is the first to investigate HER2 status by race in PCa patients. Successful completion of this study will allow us to apply for future funding to support a Phase II race-stratified clinical trial with a focused AA target accrual to treat HER2 positive (HER2+) PCa patients with newer, more efficient anti-HER2 drugs. A precision medicine approach may allow for the repurposing of drugs currently approved to treat HER2+ breast cancer patients and potentially improve overall survival for currently incurable stages of PCa. The ability to repurpose currently FDA-approved HER2-targeted drugs for PCa patients would also drastically reduce the transition time of expanding the availability of effective treatment from clinical trial patients to the general population. Timely completion of this and a subsequent clinical trial would have a major near-term impact in lessening the burden of PCa mortality in AA men and would provide an additional treatment option for others with HER2+ metastatic PCa.

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?

The potential of improving racial/minority enrollment in clinical trials has a direct influence on survival outcomes in these populations. With the appropriate focus and commitment, inclusive research studies such as this will lead to greater potential for clinical trials that diverse and inclusive thereby increasing access to cutting-edge therapies for high-risk minority populations. Continued efforts to strategically increase minority enrollment in clinical trials will ultimately help eliminate health disparities by allowing for optimized plans of treatment based on observed and documented variations in treatment response.

CHANGES/PROBLEMS

Changes in approach and reasons for change

Nothing to Report

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

Due to the transition of the lab from City of Hope in Duarte, CA to Morehouse School of Medicine in Atlanta, GA, all wet lab experiments were temporarily halted in June 2022. Due to construction at Morehouse School of Medicine, all lab equipment and supplies have been contained in storage units in Atlanta, GA until lab space became available. The Burnham lab space became available in January 2023. Lab equipment and supplies are in the process of being moved into the Burnham lab (Medical Education Building Research Wing D213 and D217) over the next few weeks with experiments expected to resume full-time in February 2023. Dr. Nicole Mavingire was hired to the lab in June 2022 and Ms. Janelle Moore was accepted into the lab in January 2023. Dr. Mavingire brings a skillset that is inclusive of all experiments required for this study. The additional expertise aligned with the re-opening of the lab will allow for a streamlined completion of the specific aims of this study.

Due to the significant impact on expenditures (please see below), there was a substantial (6-month delay by City of Hope's pathology core to pull FFPE samples as they awaited confirmation of payment processing.

Changes that had a significant impact on expenditures

The relocation of my lab to Morehouse School of Medicine was decided and announced to City of Hope leadership in May 2022. Financial administration and leadership agreed to relinquish all awards, including the Department of Defense Prostate Cancer Research Program Early Investigator Award, back to the DoD in June 2022. City of Hope's Business Official confirmed the initiation of the relinquishing process in July 2022. My lab relocated to Morehouse School of Medicine in July 2022. I emailed my DoD Scientific Officer several times and was unaware that email correspondence was not deliverable between Morehouse School of Medicine and the DoD. In October 2022, the DoD provided instructions for Morehouse School of Medicine IT department to allow for email delivery from the DoD. Morehouse School of Medicine was unable to remedy the issue. I began corresponding with the DoD from my personal gmail account. In October 2022, City of Hope discussed keeping the award with the DoD and creating an adjunct assistant professor for me to complete the performance period. City of Hope asked me if I agreed. I agreed to the non-compensated visiting professor position. Morehouse School of Medicine was also in agreement with the proposed plan. In November 2022, City of Hope initiated the paperwork to create the adjunct position. In December 2022, City of Hope initiated a no-cost extension request to the DoD since the experiments had been halted since June 2022 and the institutional placement of the award remained ambiguous. On January 3, 2023, City of Hope informed me that "COH leadership has decided to relinquish your DoD award and help facilitate the transfer to Morehouse School of Medicine. Please note, our Sponsor Research Team has emailed Mr. Robert Doan to inquire about next steps and we will keep you informed on the status." Throughout June 2022 to January 2023, I and my team have not had access to the funds as we were initially told the funds would be transferred, and then told that the funds would stay at COH, and then told that the funds would be relinquished. We are prepared to

complete the specific aims as described in the original budget and would appreciate the opportunity to access the funds to honor our intentions.

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

PRODUCTS

Publications, conference papers, and presentations

Journal publications

Nothing to Report

Books or other non-periodical, one-time publications

Nothing to Report

Other publications, conference papers, and presentations

- | | |
|----------|--|
| 02/23/21 | “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, OncLive State of the Science Summit (Sponsored by City of Hope) |
| 06/25/22 | “Advancing Inclusive Research at the Bench”, 2022 Coffey-Holden Prostate Cancer Academy Meeting, UCLA, Los Angeles, CA |
| 09/12/22 | “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, University of California Los Angeles, Los Angeles, CA |
| 10/28/22 | “HER2 expression in African American men with prostate cancer”, 29 th Prostate Cancer Foundation Annual Scientific Retreat |

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

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

Name:	<i>Leanne Burnham, Ph.D.</i>
Project Role:	<i>PI, Assistant Professor</i>
Research Identifier (e.g., ORCID ID):	<i>0000-0003-0856-6386</i>
Nearest person month worked:	<i>12</i>
Contribution to Project:	<i>Dr. Burnham has performed experimental work for Specific Aim 1 and 2, completed IRB application through COH to obtain approval, secured DOD PCBN specimens through MTA between COH and Washington University in St. Louis.</i>
Funding Support:	<i>Current DOD W81XWH2110038</i>
Name:	<i>Serene Dowiri</i>
Project Role:	<i>Intern</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>4</i>
Contribution to Project:	<i>Ms. Dowiri has performed experimental work for Specific Aim 1 and 2.</i>
Funding Support:	<i>Other</i>
Name:	<i>Rick Kittles, Ph.D.</i>
Project Role:	<i>Collaborator</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>1</i>

Contribution to Project:	<i>Dr. Kittles has performed ancestral genotyping and analysis for Specific 2.</i>
Funding Support:	<i>Other</i>
Name:	<i>Jabril Johnson, Ph.D.</i>
Project Role:	<i>Collaborator</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>1</i>
Contribution to Project:	<i>Dr. Johnson has performed ancestral genotyping and analysis for Specific 2.</i>
Funding Support:	<i>Other</i>
Name:	<i>Nicole Mavingire, Ph.D.</i>
Project Role:	<i>Postdoctoral Fellow</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>6</i>
Contribution to Project:	<i>Dr. Mavingire has performed experimental work as well as analysis for Specific Aims 1, 2, and 3.</i>
Funding Support:	<i>Other</i>
Name:	<i>Huiqing Wu, M.D.</i>
Project Role:	<i>Collaborator</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>12</i>
Contribution to Project:	<i>Dr. Wu has pulled samples for Specific Aim 2.</i>
Funding Support:	<i>Pathology Core fees funded by current DOD W81XWH2110038</i>
Name:	<i>Zhirong Yin, M.D.</i>
Project Role:	<i>Collaborator</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>3</i>
Contribution to Project:	<i>Dr. Yin has stained and scored samples for Specific Aims 2.</i>

Funding Support:	<i>Pathology Core fees funded by current DOD W81XWH2110038</i>
Name:	<i>Tanya Dorff, M.D.</i>
Project Role:	<i>Collaborator</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>12</i>
Contribution to Project:	<i>Dr. Dorff has assisted with Specific Aims 2 and 3.</i>
Funding Support:	<i>Other</i>
Name:	<i>Mya Walker</i>
Project Role:	<i>Collaborator</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>3</i>
Contribution to Project:	<i>Ms. Walker has assisted with Specific Aim 1.</i>
Funding Support:	<i>Other</i>
Name:	<i>Bettina Drake, Ph.D.</i>
Project Role:	<i>Collaborator</i>
Research Identifier (e.g., ORCID ID):	<i>N/A</i>
Nearest person month worked:	<i>3</i>
Contribution to Project:	<i>Dr. Drake has assisted with Specific Aim 2 and 3.</i>
Funding Support:	<i>Other</i>

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

There has been no change in the following active support:

01/01/21 – 12/31/23 20YOUN04
Prostate Cancer Foundation Young Investigator Award (Burnham)
Prostate Cancer Foundation
“HER2 and Androgen Receptor Signaling in Prostate Cancer” (Mentors:

Rick Kittles, Ph.D. and Tanya Dorff, M.D.)
Role: PI

07/01/20 – 12/01/23 568866221
Pfizer Prostate Cancer Foundation Global Challenge Award (Dorff)
Pfizer and Prostate Cancer Foundation
“Identifying AR Characteristics that Define Populations of Patients with
CSPC who Benefit from Early PARP Inhibition Therapy with Talazoparib”
Role: Co-Investigator

New support:

07/01/22 – 06/30/24 NIH NCI Health Disparities Research Loan Repayment Program Award
(Burnham)
HER2 Expression in African American Men with Prostate Cancer
Role: PI

Support that has ended:

03/01/20 – 01/28/22 QueensCare Charitable Division (Kittles)
QueensCare Foundation
Community-Based Prostate Cancer Screenings Among Men in South LA
Co-Lead

11/01/21 – 10/31/22 Bank of America (Kittles)
Bank of America Charitable Foundation
Prostate Cancer Screenings in the San Gabriel Valley
Co-Lead

01/01/22 – 06/30/22 Genentech (Kittles)
Roche-Genentech/City of Hope AIR (Advancing Inclusive Research) Site
Alliance
Advancing Inclusive Research in Southern California: Challenges and
Opportunities
Co-Lead

What other organizations were involved as partners?

Organization Name: DOD PCBN, Washington University in St. Louis

Location of Organization: United States

Partner's contribution to the project

Financial support: Nothing to Report

In-kind support: 456 Case Race Disparity TMA; patient-matched frozen cell pellets

Facilities: Nothing to Report

Collaboration: Bettina Drake, Ph.D.

Personnel exchanges: Nothing to Report

Other: Nothing to Report

SPECIAL REPORTING REQUIREMENTS

COLLABORATIVE AWARDS: Nothing to Report

QUAD CHARTS: Nothing to Report

CONCLUSION

Metastatic PCa is a lethal disease and therapeutic options are limited for patients.¹⁴⁻¹⁷ Since there is no cure for advanced PCa, the pursuit of therapeutic strategies to effectively treat this stage remains a critical goal. The ability to target non-androgenic growth factor signaling pathways that may promote metastasis represents a tangible and promising area of investigation. With the development of targeted therapeutic options optimized for a molecularly enriched population, there is potential to improve survival outcomes for men at high-risk for an advanced stage diagnosis. Since recent studies have revealed better treatment response and increased overall survival in AA men receiving PCa drug treatments, there is additional incentive to decrease the current racial disparity in mortality with precision medicine.¹⁸⁻²⁰ Given that our preliminary evidence suggests a potential increase in HER2 overexpression in AA men with PCa, we predict that AA mCRPC patients may especially benefit from anti-HER2 drug targeting. Successful completion of this study will generate preliminary data to support applications for future funding to 1) fully evaluate whether HER2 contributes to the worse prognosis experienced by AA men and 2) to develop treatment strategies for PCa patients, targeting HER2 overexpression with newer generation drugs that have been proven effective in other HER2+ cancer types. A precision medicine approach may allow for the repurposing of drugs currently approved to treat HER2+ breast cancer patients and potentially improve overall survival for currently incurable stages of PCa. We anticipate that successful completion of this and a subsequent clinical trial would have a major impact in reducing lethal PCa in AA men and would provide an additional treatment option for others with HER2+ mCRPC.

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APPENDICES:

CURRICULUM VITAE
LEANNE WOODS-BURNHAM, Ph.D.
Assistant Professor
Department of Physiology
Morehouse School of Medicine
720 Westview Dr. SW
Atlanta, GA 30310

Date Prepared: January 9, 2023

I. EDUCATION

- 2012 University of Akron, B.S., *Cum Laude*, Biology
- 2018 Loma Linda University School of Medicine, Ph.D., Physiology
Graduate Degree Mentor: Carlos A. Casiano, Ph.D.

II. POSTDOCTORAL TRAINING

- 2018 – 2021 Beckman Research Institute, City of Hope, Rick Kittles, Ph.D.
- 2018 – 2020 NIH DNA Damage Response and Oncogenic Signaling (DNADRS) T32 Postdoctoral Fellow

III. PROFESSIONAL EXPERIENCE, POSITIONS & EMPLOYMENT

- 2009 – 2011 Physician Shadow, Cleveland Clinic Glickman Urology and Kidney Institute, Cleveland, OH
- 2009 – 2011 Research Intern, Janet Houghton Colon Cancer Laboratory, Cleveland Clinic Lerner Research Institute, Cleveland, OH
- 2011 – 2012 STEM Peer Mentor, University of Akron, Akron, OH
- 2012 – 2018 Graduate Student and Research Assistant, Carlos Casiano Prostate Cancer Laboratory, Center for Health Disparities and Molecular Medicine, Loma Linda University School of Medicine, Loma Linda, CA
- 2012 – 2018 ABC/UTP Mentor, Center for Health Disparities and Molecular Medicine, Loma Linda University School of Medicine, Loma Linda, CA
- 2018 – 2021 Postdoctoral Fellow, Rick Kittles Prostate Cancer Laboratory, Department of Population Sciences, Division of Health Equities, Beckman Research Institute, City of Hope, Duarte, CA
- 2019 – 2022 Community-Based Prostate Cancer Screening Co-Director, Division of Health Equities, Department of Population Sciences, City of Hope, Duarte, CA

- 2021 – 2022 Advancing Inclusive Research (AIR) Site Alliance Roche/Genentech-City of Hope Site Point-of-Contact, City of Hope, Duarte, CA
- 2021 – 2021 Assistant Research Professor, Department of Population Sciences, Division of Health Equities, City of Hope, Duarte, CA
- 2022 – 2022 Assistant Professor, Leanne Burnham Prostate Cancer Laboratory, Department of Population Sciences, Division of Health Equities, Beckman Research Institute, City of Hope, Duarte, CA
- 2022 – Present Assistant Professor, Leanne Burnham Prostate Cancer Laboratory, Department of Physiology, Morehouse School of Medicine, Atlanta, GA

Certifications

- 2018 Basic Workflow of R, Center for Informatics, City of Hope, Duarte, CA
- 2019 – 2022 Clinical Investigative Training Program (CITP), City of Hope, Duarte, CA
 CITP Clinical Trials (MEDONC531)
 Adverse Events in Protocol (MEDONC400)
 CITP Protocol Development (MEDONC550)
 CITP Protocol Management (MEDONC551)
 CITP Regulatory Processing (MEDONC562)
 CITP Clinical Trial Operations: Pragmatic Perspectives (MEDONC605)

IV. HONORS, SCHOLARSHIPS & AWARDS

- 1998 Chrysler Award, Stow, OH
- 1998 President, Jim Eliot Chapter of National Honors Society, Stow, OH
- 1998 Oscar Ritchie Scholar, Kent State University, Kent, OH
- 2008 Partners in Excellence Scholar, University of Akron, Akron, OH
- 2009 – 2012 STEM Scholar, University of Akron, Akron, OH
- 2009 – 2012 McNair Scholar, University of Akron, Akron, OH
- 2010 Verna Trushel Scholar, University of Akron, Akron, OH
- 2010 – 2012 Honors Scholar, University of Akron, Akron, OH
- 2011 Outstanding Presenter, McNair Undergraduate Research Symposium, University of Akron, Akron, OH
- 2012 Outstanding McNair Scholar of the Year, University of Akron, Akron, OH
- 2012 3rd Place, BURS Symposium Undergraduate Research, Akron, OH
- 2012 1st Place, UASIS Symposium Outstanding Undergraduate Student Research, Akron, OH
 Student-of-the-Month (Awarded by University of Akron President and Board of Trustees), Akron, OH
- 2012 – 2018 IMSD Fellowship, Center for Health Disparities and Molecular Medicine, Loma Linda University, Loma Linda, CA
- 2014 – 2016 Basic Science Dean's Award, Loma Linda University, Loma Linda
- 2016 AACR Scholar-in-Training, AACR Conference on the Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved, Fort Lauderdale, FL
- 2016 2nd Place Faculty Judge Award, Basic Science Symposium, Loma Linda University, Loma Linda, CA
- 2018 1st Place Student Judge Award, Basic Science Symposium, Loma Linda University, Loma Linda, CA
- 2018 – 2020 NCI T32 Fellow, City of Hope DNADRS Program, Duarte, CA
- 2019 Endocrine Society FLARE Fellow
- 2020 Health Equities Pilot Award, City of Hope, Duarte, CA
- 2020 Faces of *Cell*, Cell Press, Cambridge, MA
- 2020 – Present Prostate Cancer Foundation Young Investigator Award, Santa Monica, CA

2020 – Present Department of Defense Prostate Cancer Research Program Early Investigator Award,
Washington D.C.
2021 The Cancer Health 25: Black Lives Matter

V. ACTIVE GRANTS/RESEARCH SUPPORT

01/01/21 – 12/31/23 W81XWH2110038
Department of Defense Prostate Cancer Research Program Early Investigator Award (Burnham)
Department of Defense
“Her2 Expression in African American Men with Prostate Cancer” (Mentors: Rick Kittles, Ph.D. and Tanya Dorff, M.D.)
Direct
Role: PI

01/01/21 – 12/31/23 20YOUN04
Prostate Cancer Foundation Young Investigator Award (Burnham)
Prostate Cancer Foundation
“HER2 and Androgen Receptor Signaling in Prostate Cancer” (Mentors: Rick Kittles, Ph.D. and Tanya Dorff, M.D.)
Direct
Role: PI

07/01/20 – 12/01/23 568866221
Pfizer Prostate Cancer Foundation Global Challenge Award (Dorff)
Pfizer and Prostate Cancer Foundation
“Identifying AR Characteristics that Define Populations of Patients with CSPC who Benefit from Early PARP Inhibition Therapy with Talazoparib”
Direct
Role: Co-Investigator

ADDITIONAL SUPPORT

07/01/22 – 06/30/24 NIH NCI Health Disparities Research Loan Repayment Program Award (Burnham)
HER2 Expression in African American Men with Prostate Cancer
Role: PI

COMPLETED GRANTS/RESEARCH SUPPORT

01/01/22 – 06/30/22 Genentech (Kittles)
Roche-Genentech/City of Hope AIR (Advancing Inclusive Research) Site Alliance
Advancing Inclusive Research in Southern California: Challenges and Opportunities
Direct
Role: Co-Lead

11/01/21 – 10/31/22 Bank of America (Kittles)
Bank of America Charitable Foundation
Prostate Cancer Screenings in the San Gabriel Valley
Direct
Role: Co-Lead

- 03/01/20 – 02/28/23 QueensCare Charitable Division (Kittles)
QueensCare Foundation
Community-Based Prostate Cancer Screenings Among Men in South LA
Direct
Role: Co-Lead
- 01/10/20 – 12/31/20 City of Hope Research Initiative Health Equity Pilot Program
(Burnham/Hooker/Shuck/Termini) City of Hope
Advanced Glycation End Products as Diagnostic and Prognostic Indicators of
Prostate Cancer Disparities in Men of West African Ancestry
Direct
Role: Co-Investigator
- 12/01/18 – 11/30/21 5T32CA186895-04
DNA Damage Response and Oncogenic Signaling Fellow (Shen)
NIH/NHLBI
Training Fellowship
Role: Trainee
- 09/01/12 – 05/01/18 5R25GM060507-12
Initiative for Maximizing Student Development Fellow (De Leon)
NIH/NIGMS
Training Fellowship
Role: Trainee

VI. PUBLICATIONS

Publications (peer-reviewed) 7 Total

1. Basu A., **Woods-Burnham L.**, Ortiz G., Rios-Colon L., Figueroa J., Albesa R., Andrade L.E., Mahler M., Casiano C.A. (2015) Specificity of antinuclear autoantibodies recognizing the dense fine speckled nuclear pattern: Preferential targeting of DFS70/LEDGFp75 over its interacting partner MeCP2. *Clin Immunol*, 161(2):241-50. PMID: 26235378.
2. **Woods-Burnham L.**, Basu A., Cajigas-Du Ross C.K., Love A., Yates C., De Leon M., Roy S., Casiano C.A. (2017) The 22Rv1 prostate cancer cell line carries mixed genetic ancestry: Implications for prostate cancer health disparities research using pre-clinical models. *Prostate*, 77(16): 1601-1608. PMID: 29030865.
3. **Woods-Burnham L.**, Stiel L., Wilson C., Montgomery S., Duran A.M., Ruckle H.R., Thompson R.A., De Leon M., Casiano C.A. (2018) Physician consultations, prostate cancer knowledge, and PSA screening of African American men in the Era of Shared Decision-Making. *Am J Mens Health*, 12(4):751-759. PMID: 29658371.
4. Cajigas-Du Ross C.K., Martinez S.R., **Woods-Burnham L.**, Durán A.M., Roy S., Basu A., Ramirez J.A., Ortiz-Hernández G.L., Ríos-Colon L., Chirshv E., Sanchez-Hernandez E.S., Soto U., Greco C., Boucheix C., Chen X., Unternaehrer J., Wang C., Casiano C.A. (2018) RNA sequencing reveals upregulation of a transcriptomic program associated with stemness in metastatic prostate cancer cells selected for taxane resistance. *Oncotarget*. 9(54):30363-30384. PMID: 30100995.
5. **Woods-Burnham L.**, Cajigas-Du Ross C.K., Love A., Basu A., Durán A.M., Sanchez-Hernandez E.S., Martinez S.R., Ortiz-Hernandez G.L., Stiel L., Wilson C., Montgomery S., Roy S., Casiano C.A. (2018) Glucocorticoids induce stress oncoproteins associated with therapy-resistance in African American and European American prostate cancer cells. *Scientific Reports*, 8(1):15063. PMID: 30305646.
6. Hooker S.E., **Woods-Burnham L.**, Bathina M., Lloyd S.M., Gorjala P., Mitra R., Nonn L., Kimbro K.S., Kittles R.A. (2019) Genetic ancestry analysis reveals misclassification of commonly used cancer cell lines. *Cancer Epidemiol Biomarkers Prev*, 28(6):1003-1009. PMID: 30787054.

- Johnson J.R., Martini R.N., Yuan Y.C., **Woods-Burnham L.**, Walker M., Ortiz-Hernandez G.L., Galloway D., Davis M.B., Kimbro K.S., Murphy A.B., Kittles R.A. (2022) 1,25-dihydroxyvitamin D₃-mediated suppression of genes associated with cell cycle regulation and actin organization in a non-malignant African American prostate cell line. *Cancer Cell International* (under review).

Publications (review articles) 3 Total

- Woods-Burnham L.**, Stiel L., Martinez S.R., Sanchez-Hernandez E.S., Ruckle H.C., Almaguel F.G., Stern M.C., Roberts L.R., Williams D.R., Montgomery S., Casiano C.A. (2020) Psychosocial stress, glucocorticoid signaling, and prostate cancer health disparities. *Cancer Health Disparities*, 4:https://companyofscientists.com/index.php/chd/article/view/169/188. PMID: 35252767.
- Johnson J.R., **Woods-Burnham L.**, Hooker S., Batai K., Kittles R.A. (2021) Genetic contributions to prostate cancer disparities in men of West African descent. *Front Oncol.* 11.770500. PMID: 34820334.
- Johnson J.R., Mavingire N., **Woods-Burnham L.**, Hooker S., Galloway D., Kittles R.A. (2022) Prostate cancer disparities: Complex interplay of prostate cancer risk factors in African American men. *To be submitted December 2022.*

Editorials and Letters 2 Total

- Woods-Burnham L.** (2020) Not all champions are allies in health disparities research. *Cell*, 183(3):580-582. PMID: 33125884.
- Woods-Burnham L.**, Johnson J.R., Hooker S.E., Bedell F.W., Dorff T.B., Kittles R.A. (2021) The role of diverse populations in U.S. clinical trials. *Med*, 2(1):21-24. doi: 10.1016/j.medj.2020.12.009. PMID: 35590131.

VII. INVITED SEMINARS/LECTURES/FORUMS

Intramural

- | | |
|----------|--|
| 01/14/12 | “Pursuing STEM as a Nontraditional Student”, Rosa Parks Annual Event, University of Akron |
| 02/04/12 | “Pursuing STEM as a Nontraditional Student”, National TRiO Day, University of Akron |
| 04/09/12 | “Hedgehog Signaling in Colon Cancer”, Talent Dividend Forum, University of Akron |
| 02/05/18 | “Glucocorticoid Signaling in Black Men with Prostate Cancer”, Chalk Talk, Loma Linda University |
| 09/15/18 | “Prostate Cancer Education and Screening for Black Men”, Tabahani Book Circle Worship in Pink, Imperial Heights Community Church of the Brethren (Sponsored by City of Hope) |
| 11/10/18 | “Prostate Cancer Education and Screening for Black Men”, Men’s Day Breakfast/Health and Wellness Forum, Metropolitan Baptist Church (Sponsored by City of Hope) |
| 01/12/19 | “Prostate Cancer Education and Screening for Black Men”, CPAD Community Benefits Forum, City of Hope |
| 04/06/19 | “Prostate Cancer Education and Screening for Black Men”, 10 th Annual MAP Neighborhood Conference (Sponsored by City of Hope) |
| 04/11/19 | “Prostate Cancer Education and Screening for Black Men”, Learning and Professional Development Week, City of Hope |
| 06/01/19 | “Prostate Cancer Education and Screening for Black Men”, Omega Psi Phi and Charles R. Drew Blood Drive and Health Fair (Sponsored by City of Hope) |
| 09/21/19 | “Prostate Cancer Education and Screening for Black Men”, 32 nd Long Beach Jazz Festival Health and Wellness Pavilion (Sponsored by City of Hope) |
| 10/13/19 | “Prostate Cancer Education and Screening for Black Men”, First Ladies Los Angeles Chapter Annual Health Day (Sponsored by City of Hope) |

- 02/01/20 “Prostate Cancer Education and Screening for Black Men”, 39th Annual Orange County Black History Parade and Cultural Fair (Sponsored by City of Hope)
- 07/23/20 “All of Us: A Multidisciplinary Approach to Reducing Cancer Health Disparities”, KJLH Radio (Sponsored by City of Hope)
- 08/28/20 “My Career as a Postdoc”, Irell & Manella Graduate School of Biological Sciences Symposium, City of Hope
- 11/18/20 “Clinical Trial Recruitment for African Americans”, CISCRP Aware for All Webinar (Sponsored by City of Hope)
- 02/05/21 “Implications of Telemedicine for Prostate Cancer Patients”, Patient Empowerment Network and Diverse Health Hub (Sponsored by City of Hope)
- 02/23/21 “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, OncLive State of the Science Summit (Sponsored by City of Hope)
- 04/15/21 “What is Special About Research at City of Hope”, Building Hope Toast of Hope Event, City of Hope
- 06/30/21 “Dine in for Hope”, City of Hope
- 07/31/21 “Clinical Trials: An Answer to Cancer?”, Cynthia Perry Ray Foundation Sistahs Can We Talk Monthly Nurses Series (Sponsored by City of Hope)
- 09/23/21 “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, City of Hope Community Engagement Community Advisory Board
- 12/10/21 “HER2 Expression in African American Men with Prostate Cancer”, City of Hope Health Equities Faculty Candidate Seminar

Symposia Presentations

- 11/11/10 “GLI vs. Smoothened (SMO) as therapeutic targets in the Hedgehog (HH) signaling pathway”, ABRCMS Annual Meeting
- 01/06/10 “GLI vs. Smoothened (SMO) as therapeutic targets in the Hedgehog (HH) signaling pathway”, McNair Undergraduate Research Symposium at UC, Berkeley
- 07/22/11 “GLI vs. Smoothened (SMO) as therapeutic targets in the Hedgehog (HH) signaling pathway”, McNair Undergraduate Research Symposium at University of Akron
- 03/05/12 “GLI vs. Smoothened (SMO) as therapeutic targets in the Hedgehog (HH) signaling pathway”, Choose Ohio First STEMM Showcase at COSI
- 04/09/12 “GLI vs. Smoothened (SMO) as therapeutic targets in the Hedgehog (HH) signaling pathway”, UASIS Research Symposium
- 04/06/14 “Specificity of the human autoantibody response against the stress oncoprotein LEDGF/p75”, AACR Annual Meeting
- 11/16/14 “Glucocorticoid signaling in prostate cancer cells”, Loma Linda University Annual Basic Sciences Symposium
- 03/16/15 “Glucocorticoid signaling in prostate cancer cells”, Loma Linda University Annual Postgraduate Convention
- 05/13/15 “PSA Screening for Black Men in Southern California”, Loma Linda University Counseling and Family Sciences Conference
- 05/13/15 “Physician consultations, prostate cancer knowledge, and PSA screening in African American men: Are the conversations effective?”, Loma Linda University Counseling and Family Sciences Conference
- 09/14/16 “Glucocorticoid signaling in prostate cancer cells”, Loma Linda University Annual Health Disparities Research Symposium
- 09/26/16 “Glucocorticoid signaling in prostate cancer cells”, AACR Conference on the Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved
- 09/26/16 “Physician consultations, prostate cancer knowledge, and PSA screening in African American men: Are the conversations effective?”, AACR Conference on the Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved

- 09/15/17 “Glucocorticoid-mediated upregulation of stress oncoproteins associated with chemoresistance: Implications for prostate cancer health disparities”, Loma Linda University Annual Health Disparities Research Symposium
- 04/15/18 “Glucocorticoid-mediated upregulation of stress oncoproteins associated with chemoresistance: Implications for prostate cancer health disparities”, AACR Annual Meeting
- 11/03/18 “Genetic Ancestry Analysis Reveals Misclassification of Commonly Used Cancer Cell Lines”, AACR Conference on the Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved
- 09/21/19 “Vitamin D Signaling of Immune-Related Genes in Diverse Prostate Cancer Cell Lines”, AACR Conference on the Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved
- 08/10/21 “HER2 expression in African American men with prostate cancer”, Eugene and Ruth Roberts Summer Student Academy Symposium
- 10/07/21 “Connecting under-resourced populations: A community-based prostate cancer screening intervention”, 14th AACR Conference on The Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved
- 11/08/21 “HER2 and AR signaling in prostate cancer”, 28th Prostate Cancer Foundation Annual Scientific Retreat
- 09/18/22 “The Basic Science of Stress and Cancer Health Disparities: From Environment to Cancer Biology”, 15th AACR Conference on The Science of Cancer Health Disparities in Racial/Ethnic Minorities and the Medically Underserved, Philadelphia, PA (Session Co-Chair)
- 10/28/22 “HER2 expression in African American men with prostate cancer”, 29th Prostate Cancer Foundation Annual Scientific Retreat

Extramural Invited Seminars

- 10/15/19 “Science as a Career”, Career Day, Colony High School, Ontario, CA
- 10/05/20 “Dine in for Hope”, I Am The Voluntourist, City of Hope, Duarte, CA
- 10/15/20 “Science as a Career”, Career Day, Buchtel High School, Akron, OH
- 01/20/21 “Health Disparities: Past, Present, and Future”, Metropolitan State University, Saint Paul, MN
- 03/18/21 “Prostate Cancer in Black Men: A Personal and Professional Perspective”, The Patient Portal, Los Angeles, CA
- 04/27/21 “Cancer Research Perspective from a Cancer Patient”, CURE Magazine, Los Angeles, CA
- 09/18/21 “Prostate Cancer in Black Men: The Latest in Screening and Treatment Options”, Prostate Cancer Foundation & The West Angeles Church of God in Christ, Los Angeles, CA
- 05/04/22 “Patient and Healthcare Provider Education on Advancing Inclusive Research”, Roche/Genentech AIR Site Alliance, San Francisco, CA
- 05/12/22 “Health Disparities Research: Viewpoints from a Survivor”, Universal Music Publishing Group and City of Hope Music, Film, and Entertainment Industry, Los Angeles, CA
- 05/24/22 “Health Disparities Research: Viewpoints from a Survivor”, Universal Music Publishing Group and City of Hope Music, Film, and Entertainment Industry, New York, NY
- 06/11/22 “Increasing Diversity in Clinical Trials: Multiple Solutions to Achieve Clinical Trial Equity”, ENDO 2022 Annual Conference, Atlanta, GA
- 06/21/22 “Social Determinants of Technology”, City of Hope’s Closing the Care Gap–2022 California Coalition for Cancer Equity (C3E) Health Justice Summit, Grammy Museum, Los Angeles, CA (Session Moderator)

- 06/25/22 “Advancing Inclusive Research at the Bench”, 2022 Coffey-Holden Prostate Cancer Academy Meeting, UCLA, Los Angeles, CA
- 09/12/22 “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, University of California Los Angeles, Los Angeles, CA
- 10/26/22 “The Sociology of Disparities in Graduate and Medical Education”, Prostate Cancer Foundation 29th Annual Scientific Retreat, San Diego, CA
- 11/15/22 “Tackling Prostate Cancer Health Disparities at the Bench, Clinic, and Community”, Emerging 100 of Atlanta General Body Meeting, Atlanta, GA

VIII. TEACHING/EDUCATION/EDUCATIONAL ACTIVITIES

Other Research Mentoring Activities

Arthur Love	High School Student	Martin Luther King High School	2013 2016
Greisha Ortiz-Hernandez	BS Student	Universidad Metropolitana	2013
Evelyn Sanchez	BS Student	California State, Northridge	2015
Isaiah Sailors	High School Student	Chaffey High School	2019 – 2022
Serene Dowiri	BS Student	UCLA	2021 – 2022
Johnny Gitau	BS Student	Oakwood University	2022
Nicole Mavingire, Ph.D.	Postdoctoral Fellow	Morehouse School of Medicine	2022 – Present
Janelle Moore	Graduate Student	Morehouse School of Medicine	2023 – Present

IX. SERVICE TO INSTITUTIONS

Loma Linda University School of Medicine

- Member, ABC/UTP Summer Research Admissions Committee, 2013 – 2017
- Member, Project C.H.A.N.G.E. Planning Committee, 2013 – 2018
- Student Representative, Curriculum Committee, 2014 – 2015
- Academic Vice-President, Student Council, 2016 – 2018
- Member, Admissions Committee, 2017 – 2018

City of Hope

- Member, Genitourinary Disease Team, 2018 – Present
- Co-Director, Division of Health Equities Community-Based Prostate Cancer Screening Program, 2018 – 2022
- 09/22/18 Antelope Valley Prostate Cancer Screening, Grace Church, Lancaster, CA
- 01/26/19 KKLA Radio Family Health Fair, First Church of the Nazarene, Altadena, CA
- 04/06/19 Set for Life, 10th Annual MAP Neighborhood Conference, Monrovia, CA
- 06/08/19 Health Pasadena, Men Educating Men About Health, Pasadena, CA
- 06/01/19 Omega Psi Phi and Charles R. Drew Blood Drive and Health Fair, Compton, CA
- 09/21/19 32nd Long Beach Jazz Festival Health and Wellness Pavilion, Long Beach, CA
- 10/13/19 Crenshaw Christian Center, First Ladies Los Angeles Chapter Annual Health Day, Los Angeles, CA
- 10/13/19 First A.M.E. Los Angeles, First Ladies Los Angeles Chapter Annual Health Day, Los Angeles, CA
- 10/19/19 Taste of Soul Los Angeles, Los Angeles (Crenshaw), CA
- 11/16/19 Mejor Salud Pasadena, Pasadena, CA
- 01/25/20 KKLA Radio Family Health Fair, First Church of the Nazarene, Altadena, CA
- 02/01/20 39th Annual Orange County Black History Parade and Cultural Fair, Anaheim, CA
- 02/15/20 Annual Pasadena Black History Parade and Festival, Pasadena, CA
- 05/21/22 Azusa Pacific University/Foothill Unity Center Annual Health Fair, Azusa, CA
- 06/18/22 Crenshaw Church of Christ, Los Angeles, CA

06/25/22 Omega Psi Phi and Charles R. Drew Blood Drive and Health Fair, Compton, CA
Participant, Graduate Medical Education and Health Disparities Collaboration, 2020 – 2022
Site Point-of-Contact, Advancing Inclusive Research (AIR) Site Alliance Roche/Genentech-City of Hope,
2021 – 2022
Admissions Committee, Roberts Summer Academy, 2021

X. SERVICE TO PROFESSION

Professional Memberships

Participating Member

2022 – Present Associate Scientist, O'Neal Comprehensive Cancer Center, Cancer Control and
Population Sciences

2022 – Present Member, Genitourinary Disease Team, Grady Health

Societies

2012 – Present Member, American Association for Cancer Research, Philadelphia, PA

2018 – Present Member, Endocrine Society, Washington D.C.

Committees

2019 – 2020 Member, Endocrine Society Research Affairs Core Committee, Washington D.C.

2022 – Present Member, Prostate Cancer Foundation, Young Investigator Prostate Cancer Disparities
Working Group

2022 Member, Prostate Cancer Foundation, Scientific Retreat Young Investigator Planning
Committee

Editorial and Review Experience

Review Panels/Study Sections

2022 Ad-hoc Member, NCI Connecting Underrepresented Populations to Clinical Trials
(CUSP2CT) Review Panel

Manuscript Reviews

*The Prostate, American Journal of Men's Health, International Journal of Urology,
Cancer Medicine, American Journal of Preventive Medicine, Frontiers in Pharmacology,
Prostate Cancer Foundation, Med by Cell Press, Prostate Cancer and Prostatic Diseases*

XI. PATENTS, INVENTIONS & COPYRIGHTS

None

Major Task 3, Subtask 1: Extract genomic DNA.
TMA: 456 Case Race Disparity TMA frozen cell pellets [DOD]

We extracted and quantified genomic DNA from frozen cell pellets which were patient-matched with the 456 Case Race Disparity TMA from DOD PCBN.

Sample ID	5ng/uL in 20uL	Final Volume 20uL
15770	7.462686567	12.53731343
31283	1.892147588	18.10785241
31707	0.379434642	19.62056536
31737	2.178649237	17.82135076
31834	2.570694087	17.42930591
31915	0.747943156	19.25205684
31992	0.668002672	19.33199733
32145	2.290950745	17.70904926
32213	1.144819691	18.85518031
32372	4.395604396	15.6043956
32628	3.231017771	16.76898223
32650	5.208333333	14.79166667
32789	0.532623169	19.46737683
32798	1.771479185	18.22852081
32824	3.401360544	16.59863946
33045	3.412969283	16.58703072
33047	1.432664756	18.56733524
33080	5.361930295	14.63806971
33122	4.366812227	15.63318777
33124	2.656042497	17.3439575
33131	2.980625931	17.01937407
33139	1.366120219	18.63387978
33150	1.005025126	18.99497487
33292	3.868471954	16.13152805

33466	2.980625931	17.01937407
33505	1.901140684	18.09885932
33631	0.659630607	19.34036939
33936	2.447980416	17.55201958
34593	0.930232558	19.06976744
34716	2.242152466	17.75784753
34718	1.571091909	18.42890809
34849	2.777777778	17.22222222
34998	1.23228589	18.76771411
35163	1.379310345	18.62068966
35228	1.482579689	18.51742031
35232	2.002002002	17.997998
35298	1.100715465	18.89928453
35308	1.430615165	18.56938484
35445	1.068947087	18.93105291
35706	0.294811321	19.70518868
35766	0.352485019	19.64751498
35778	0.338294993	19.66170501
36250	0.461467467	19.53853253
36366	0.276014353	19.72398565
36494	0.562113547	19.43788645
36609	2.083333333	17.91666667
36999	0.729927007	19.27007299
37115	0.956022945	19.04397706
37651	2.237136465	17.76286353
37770	1.347708895	18.65229111
37814	1.013171226	18.98682877
37918	0.22426553	19.77573447
38180	0.967117988	19.03288201
38400	0.180603215	19.81939679
38468	1.068376068	18.93162393
38469	0.319386777	19.68061322
38692	2.506265664	17.49373434
38823	0.903342367	19.09665763
39239	0.159033079	19.84096692
39394	8.771929825	11.22807018
39559	5.291005291	14.70899471
39773	9.302325581	10.69767442
39792	6.079027356	13.92097264
39816	17.24137931	2.75862069

39958	8.771929825	11.22807018
40069	12.5	7.5
40449	14.81481481	5.185185185
40654	3.490401396	16.5095986
40694	10.92896175	9.071038251
40892	2.059732235	17.94026777
41715	10.30927835	9.690721649
41831	10.3626943	9.637305699
41927	7.547169811	12.45283019
42716	2.157497303	17.8425027
43626	14.18439716	5.815602837
43815	7.751937984	12.24806202
45072	10.92896175	9.071038251
45371	11.11111111	8.888888889
45389	17.69911504	2.300884956
45810	6.097560976	13.90243902
46438	12.5	7.5
48634	1.191895113	18.80810489
48912	2.43902439	17.56097561
49537	2.583979328	17.41602067
49726	4.739336493	15.26066351
49728	4.347826087	15.65217391
49952	9.803921569	10.19607843
50271	2.941176471	17.05882353
50400	4.291845494	15.70815451
50459	4.201680672	15.79831933
50636	3.086419753	16.91358025
52113	8.26446281	11.73553719
52708	7.633587786	12.36641221
52971	4	16
53628	5	15
54956	2.849002849	17.15099715
55354	5.319148936	14.68085106
55722	4.081632653	15.91836735
57138	5.649717514	14.35028249
57319	3.378378378	16.62162162
57321	1.851851852	18.14814815
57394	3.6900369	16.3099631
57433	3.125	16.875
57825	3.134796238	16.86520376

57827	2.5	17.5
58174	3.236245955	16.76375405
59169	0.611246944	19.38875306
59241	1.016260163	18.98373984
59276	1.510574018	18.48942598
59302	0.972762646	19.02723735
59500	3.278688525	16.72131148
59512	3.610108303	16.3898917
59645	7.575757576	12.42424242
60445	5.434782609	14.56521739
60603	4.237288136	15.76271186
60624	0.631313131	19.36868687
60882	1.319261214	18.68073879
61051	2.624671916	17.37532808
61970	2.88184438	17.11815562
62076	3.378378378	16.62162162
62125	1.680672269	18.31932773
62280	2.057613169	17.94238683
62315	3.787878788	16.21212121
64881	0.658761528	19.34123847
65920	8.771929825	11.22807018
66009	0.686341798	19.3136582
66495	3.289473684	16.71052632
66555	6.024096386	13.97590361
66674	3.154574132	16.84542587
66764	7.142857143	12.85714286
66844	5.025125628	14.97487437
67557	3.25732899	16.74267101
67665	2.777777778	17.22222222
67893	4.975124378	15.02487562
67925	3.424657534	16.57534247
68031	1.919385797	18.0806142
68194	6.493506494	13.50649351
68342	0.933706816	19.06629318
68404	3.184713376	16.81528662
68406	3.215434084	16.78456592
68416	0.481463649	19.51853635
68986	1.336898396	18.6631016
69279	1.371742112	18.62825789
69303	0.881834215	19.11816578

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69608	1.03950104	18.96049896
70743	1.219512195	18.7804878
71327	0.64808814	19.35191186
71454	2.341920375	17.65807963
71865	1.572327044	18.42767296
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101004	10.41666667	9.583333333
101150	6.024096386	13.97590361
101755	2.816901408	17.18309859
101911	0.867302689	19.13269731
102021	2.444987775	17.55501222
102744	4.545454545	15.45454545
102788	2.257336343	17.74266366
103250	5.102040816	14.89795918
105654	2.141327623	17.85867238
108756	2.304147465	17.69585253
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109169	2.43902439	17.56097561
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111240	5.319148936	14.68085106
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112220		
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115082	4.484304933	15.51569507
115425	1.721170396	18.2788296
115741	40	-20
116150	4.739336493	15.26066351
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800424	7.874015748	12.12598425
800515	15.87301587	4.126984127
800517	1.84501845	18.15498155
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801623	6.369426752	13.63057325
801869	1.612903226	18.38709677

802073	3.164556962	16.83544304
802092	1.584786054	18.41521395
802301	5.952380952	14.04761905
803538	1.477104874	18.52289513
803575	1.152073733	18.84792627
803960	2.732240437	17.26775956
803962	1.841620626	18.15837937
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804336	3.039513678	16.96048632
804356	1.34589502	18.65410498
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804574	4.115226337	15.88477366
804687	4.237288136	15.76271186
805033	0.267022697	19.7329773
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808529	1.006036217	18.99396378
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809098	0.304136253	19.69586375
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809454	5.235602094	14.76439791

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813641	3.802281369	16.19771863
813874	12.34567901	7.654320988
814185	0.644329897	19.3556701
814254	5.681818182	14.31818182
814429	11.62790698	8.372093023
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901688	1.689189189	18.31081081
901712	2.89017341	17.10982659
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903367	1.285347044	18.71465296

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911782	2.164502165	17.83549784
912017	1.312335958	18.68766404
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913490	3.115264798	16.8847352
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925928	1.426533524	18.57346648
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1006160	1.314060447	18.68593955
1006302	1.054852321	18.94514768
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1011163	0.729394602	19.2706054
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1020835	0.900090009	19.09990999
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1025676	0.67294751	19.32705249
1026744	5.18134715	14.81865285
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1030214	1.055966209	18.94403379
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1030727	0.379362671	19.62063733
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1033830	6.329113924	13.67088608
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1104852	0.61462815	19.38537185
1105501	10.86956522	9.130434783
1105503	29.41176471	-9.411764706
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1108665	4.366812227	15.63318777
1109089	8.928571429	11.07142857
1109785	4.149377593	15.85062241
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1137736	0.381533766	19.61846623
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1200773	0.678886626	19.32111337
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1202621	3.95256917	16.04743083
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1208840	16.39344262	3.606557377
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