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TITLE: Vitamin D and Related Genes, Race and Prostate Cancer Aggressiveness

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The overall goal of the study is to examine whether altered vitamin D status (as measured by serum metabolites and by functional polymorphisms within genes related to vitamin D transport, metabolism and activity) is associated with increased risk of aggressiveness prostate cancer, and may explain some of the racial disparity seen in aggressive prostate cancer. The project activities, as outlined in the statement of Work Tasks and Milestones, are running on schedule. The study team has been organized and participates in monthly conference calls (or more frequent as needed) to discuss study progress and data collection and analyses. IRB approval was obtained from all local institutions and the DOD HSRRB in year 1. All biospecimens have been shipped the institutions and measurements of circulating concentrations of vitamin D metabolites, calcium, phosphorus and PTH are complete. Data has been requested and obtained from the parent study, PCaP. Data merging and cleaning has been accomplished and preliminary results have been presented or submitted to national scientific meetings with the large representation of African Americans in this investigation, the proposed research has tremendous potential to provide insights into a chronically underserved population carrying an unequal burden of disease.
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INTRODUCTION:

Experimental and ecologic studies support a role of vitamin D in prostate cancer prevention and prognosis; however, epidemiologic study results are inconsistent. Altered vitamin D status (as measured by plasma metabolites and by functional polymorphisms within genes related to vitamin D transport, metabolism and activity) is hypothesized to be associated with increased risk of aggressive prostate cancer, and may explain some of the racial disparity seen in aggressive prostate cancer. It is also hypothesized that plasma parathyroid hormone (PTH), serum calcium and serum phosphorus levels are inversely and directly correlated with plasma 25(OH)D and 1,25(OH)2D levels, respectively, and are positively associated with disease aggressiveness. Polymorphisms within ten genes involved in vitamin D transport, metabolism and activity will be examined to determine whether 1) allele and genotype frequencies differ by race, 2) plasma vitamin D metabolite concentrations are related to polymorphisms in these genes, 3) allele and genotype/haplotypes frequencies are different in more aggressive disease as compared to less aggressive disease, and 4) vitamin D and genetic polymorphisms act synergistically to affect prostate cancer aggressiveness. We will examine these associations among vitamin D status, PTH, calcium, phosphorus, polymorphisms in vitamin D-related genes, and prostate cancer aggressiveness in the North Carolina-Louisiana Prostate Cancer Project (PCaP), a previously-conducted case-only study of prostate cancer among equal numbers of African Americans and European Americans. New laboratory data will be generated using previously-collected biospecimens from PCaP, and data will be analyzed using epidemiologic techniques for estimating odds of highly aggressive prostate cancer according to vitamin D, PTH, calcium, phosphorus and genetic polymorphisms.

BODY:

The project activities, as outlined in the Statement of Work (SOW) Tasks and Milestones, are running on schedule. Activities in Task #1, the run-in phase of months 1-6, have been accomplished. The majority of activities related to Task #2 (planned to occur in months 7 to 24 of the grant award period) have been accomplished as outlined below. Below please find the original SOW activity listed in the numbered bullet, and the progress and status of those activities listed in the indented lettered bullet underneath each activity.

Task 1: Run-in Phase, Months 1-6:

1. Organize the investigative team and schedule regular conference calls between investigators
   a. Conference calls have been occurring once per month or more often as needed
2. Obtain IRB approval for the study from all institutions and DoD HSRRB
   a. IRB approval was granted by each of the institutions (USC, Roswell Park Cancer Institute, UCLA, and UNC-CH) and by DoD HSRRB
3. Complete the data acquisition form from the parent PCaP Study
   a. Data was requested and obtained from PCaP
4. Develop a Manual of Operations (MOP), a detailed document describing data transfer, data merging, and data management systems. The MOP content is based on our successful experience with other large-scale epidemiologic studies.
   a. A system of data transfer has been developed, and the MOP has been assembled.
5. Arrange for shipment of 1,200 serum samples to Roswell Park for vitamin D analyses, 1,200 plasma and 1,200 DNA samples to USC for PTH analyses and genotyping, and serum samples to UCLA for calcium (1,200 samples) and phosphorus (1,200 samples) analyses
   a. It was decided that plasma samples were more appropriate for vitamin D analyses, instead of serum samples, because the plasma samples were collected and transported under light-protected conditions.
   b. It was decided that genotyping would be conducted by Roswell Park Cancer Institute Shared Genomic Resources facility due to their having the appropriate technology and experience for the Illumina Goldengate and Sequenom genotyping methodology being used.
   c. Plasma samples and DNA samples were shipped from UNC-CH to Roswell Park, serum samples were shipped to UCLA, and plasma samples were shipped to USC.

6. Drs. Steck and Johnson attend PCRP IMPaCT Annual Meeting or other scientific meeting.
   a. There was not an IMPaCT meeting in Year 1. Dr. Steck attended the American Society of Preventive Oncology meeting in March 2012.

All milestones for Task #1 were met (IRB and HSRRB approval, samples aliquotted and shipped to labs, data systems in place for capture of all data from different sources).

Task 2: Laboratory Analyses, Interim Data Analyses, Months 7-24:

1. Conduct plasma 25(OH)D and 1,25(OH)2D lab measurements at Roswell Park Cancer Institute
   a. Plasma vitamin D metabolite measurements are complete.

2. Conduct genotyping at Roswell Park Cancer Institute Shared Genomics Resource facility
   a. This will occur in Year 3. We are developing the list of tagSNPs of multiple genes related to vitamin D transport, metabolism and activity. SNPs will be analyzed using Illumina Goldengate and Sequenom methodology.

3. Conduct plasma PTH measurements at Psychoneuroimmunology Lab at USC
   a. Plasma PTH measurements are complete

4. Conduct serum calcium and phosphorus measurements at UCLA
   a. Serum calcium and phosphorus measurements are complete.

5. Hire graduate assistant at USC
   a. A senior-level doctoral student, Daria McMahon, has been hired as a GA.

6. Have all raw data sent to USC and to PCaP parent study
   a. Raw data from Roswell Park and UCLA have been distributed to USC. Data generated from the study will be sent to PCaP at the end of the study.

7. Manage data, begin cleaning data as it becomes available
   a. Data from PCaP, Roswell Park, UCLA, and USC have been merged and cleaned.

8. Drs. Steck and Johnson attend PCRP IMPaCT Annual Meeting or other scientific meeting.
   a. There was no IMPaCT meeting in Year 2. Dr. Steck attended the American Association for Cancer Research Frontiers in Cancer Prevention meeting in November 2012.

All of the milestones for Task #2 (successful completion of lab work and raw data deposit at centralized location) have been completed, with the exception of the genotyping which will occur in Year 3. One abstract was presented at the American Association for Cancer Research (AACR) annual meeting in April 2013, and another will be presented at the AACR Science of Cancer Health Disparities meeting in December 2013 (these are tasks associated with Task 3 to
occur in months 25-36, indicating we are ahead of schedule for some of the lab assays and preliminary data analyses).

**KEY RESEARCH ACCOMPLISHMENTS, YEAR 2:**

- Arranged for shipment of plasma samples to USC for PTH analyses and DNA samples to Roswell Park for genotyping.
- Conducted plasma PTH measurements (by USC).
- Hired graduate assistant at USC.
- Merged and cleaned data from PCaP, Roswell Park, UCLA and USC.
- Conducted preliminary data analyses of vitamin D metabolites and prostate cancer aggressiveness by race.
- Presented abstract at AACR Annual Meeting in April 2013.
- Submitted abstract to the AACR Science of Cancer Health Disparities 2013 meeting (to be presented in December 2013).

**REPORTABLE OUTCOMES:**


**CONCLUSION:**

The project is proceeding on schedule. Data are currently being collected and analyzed, and we are determining the optimal strategy for selecting SNPs and genotyping to provide the most robust genetic data related to vitamin D activity and metabolism. Presentation of findings at scientific meetings and submission of manuscripts for publication are planned for Year 3, with one abstract being presented at the AACR Science of Cancer Health Disparities meeting in December 2013, and other abstracts and manuscripts in preparation. With the large representation of African Americans in this investigation, the proposed research has tremendous potential to provide insights into a chronically underserved population carrying an unequal burden of disease. By examining modifiable biomarkers of risk of aggressive disease and genetic susceptibility by race, this study will impact the identification of subjects at high risk for advanced disease and aid in the design of interventions to target those individuals who will receive the most benefit.