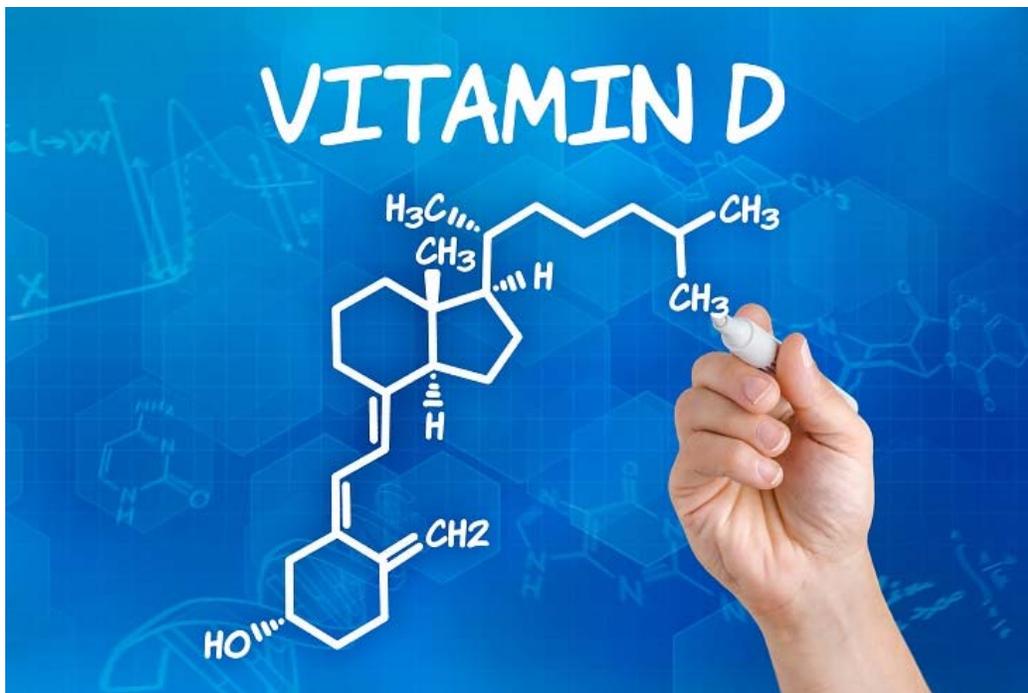




A blog for Progressnotes

Transcriptome Differences in Prostate Cancer Highlight Racial Disparities and Vitamin D

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*Summary: Investigators at the Medical University of South Carolina and Ralph H. Johnson VA Medical Center report clinical research showing that African-American and European-American men with prostate cancer exhibit significantly different expression of genes associated with immune response and inflammation, in the July 2016 issue of *Pharmacogenomics*. Systems-level, RNA analyses support the concept that inflammatory processes may contribute to racial disparities in disease progression and that vitamin D₃ supplementation can modulate pro-inflammatory transcripts.*

The results of clinical studies by investigators at the Medical University of South Carolina (MUSC) and the Ralph H. Johnson VA Medical Center (VAMC), reported in the July 2016 issue of *Pharmacogenomics* ([doi:10.2217/pgs-2016-0025](https://doi.org/10.2217/pgs-2016-0025)), demonstrate transcriptome-level linkages between racial disparities in circulating levels of vitamin D and expression of pro-inflammatory genes in African American (AA) patients with prostate cancer compared to European American (EA) patients.

Racial disparities in prostate cancer are well documented with AA men having significantly higher risk of developing prostate cancer and significantly higher mortality rates than EA men. In addition, among patients presenting at the same disease stage, AA men often have higher prostate-specific antigen (PSA) levels and higher-grade tumors than EA men. However, the biological mechanisms underlying these substantial and persistent disparities are unclear.

Researchers at MUSC and VAMC noticed that racial disparities in prostate cancer mirror differences in circulating levels of vitamin D between AA and EA patients. Vitamin D₃ is known to have multiple anti-cancer actions including suppression of cyclo-oxygenase-II (an independent predictor of cancer recurrence) and inhibition of IL-8 (an angiogenic, pro-inflammatory cytokine).

have multiple anti-cancer actions including suppression of cyclo-oxygenase-II (an independent predictor of cancer recurrence) and inhibition of IL-8 (an angiogenic, pro-inflammatory cytokine). Prostate cells express the vitamin D receptor, vitamin D-25-hydroxylase, 25 hydroxyvitamin D-1-alpha-hydroxylase, and 25-hydroxyvitamin D-24-hydroxylase. Thus, normal prostate cells can synthesize 25(OH)D₃ (calcidiol) from vitamin D (cholecalciferol), and 1,25(OH)₂D₃ (calcitriol) from 25(OH)D₃. 1,25(OH)₂D₃ (calcitriol) is the bioactive, hormonal, and most potent form of vitamin D and facilitates cell-to-cell communication via paracrine/autocrine pathways.

Sebastiano Gattoni-Celli, M.D., Professor of Radiation Oncology at MUSC, and senior author on the article, explains how his team came to explore the therapeutic potential of vitamin D supplementation in prostate cancer, "A lot of previous work shows that D₃ levels are much lower in African Americans than in European Americans and it's well established that prostate cells are very sensitive to vitamin D levels. So this raised the possibility that long-term vitamin D deficiency may contribute to the progression of prostate cancer, especially among African American men. We began to wonder whether eliminating racial disparities in circulating levels of vitamin D, through supplementation, could help reduce the disparities we see in prostate cancer outcomes."

The team designed a prospective, placebo-controlled, clinical study to investigate the effects of a daily 4,000 IU vitamin D₃ supplementation over a two-month period among 27 men (10=AA, 17=EA) who had elected to treat their prostate cancer via prostatectomy. A trial length of two months was chosen to leverage the recommended, standard-of-care recovery period between their biopsy and surgery procedures. Using high-throughput RNA sequencing, they performed a series of genome-wide expression profiling experiments to generate transcriptional profiles of patients' prostate tissue samples and assessed them using systems-level analyses. Their primary aims were to: (1) illuminate any molecular differences in gene expression that may be related to prostate cancer disparities between AA and EA men; and (2) investigate any effects vitamin D supplementation may have on the prostate transcriptome.

Not only did the team find significant differences in gene expression between AA and EA men but also between AA men receiving vitamin D supplements and AA men receiving placebo. Due to the size of the RNA sequencing dataset, results are reported as adjusted p-values (or q-values) which represent the smallest 'false discovery rate' at which a result can be called significant. A total of 3,107 prostate genes were differentially expressed between the AA and EA groups (q<0.1) with 8,238 differentially expressed transcripts between AA and EA subjects (q<0.4). Analyses of these found that AA study patients had substantially elevated expression of transcripts related to immune response and inflammation.

"The number of genes expressed differently in AA and EA was a really big surprise—we found differences in over 8,000 genes," said Gattoni-Celli. "I expected something but not this massive difference and it was not a fluke. When we compared our results with previous studies using a less advanced technology, we saw that they, too, found these differences, but not as many."

"Our findings captured all of the differences observed in previous studies but also many more because newer RNAseq technology and Big Data analytical approaches allowed us to see the transcriptome in greater detail," noted **Gary Hardiman, Ph.D.**, Professor of Medicine and Public Health Sciences and Bioinformatics Director for the **Center for Genomic Medicine at MUSC**, and co-senior author on the article. "This analysis was performed using the OnRamp Bioinformatics Genomics Research Platform we deployed at MUSC a little over a year ago. Our approach converged advanced genomics analysis, comprehensive data management, big data analytics and hyperscale servers. A 'Big Data' analytical pipeline that utilized Hadoop software was

Genomics Research Platform we deployed at MUSC a little over a year ago. Our approach converged advanced genomics analysis, comprehensive data management, big data analytics and hyperscale servers. A 'Big Data' analytical pipeline that utilized Hadoop software was implemented. This enabled an automated RNAseq workflow to process the patient data and explore differential prostate gene expression analysis between AA and EA men and sensitively interrogate the effects of vitamin D supplementation with robust statistical power.”

When comparing AA men receiving vitamin D supplementation to AA men receiving placebo, the team found 817 differentially expressed genes ($q < 0.4$). However, no similar difference in gene expression was observed between EA men receiving vitamin D supplementation versus placebo. Comparing the 8,238 differentially expressed transcripts between AA and EA subjects with the 817 genes that were differentially expressed among AA men receiving vitamin D supplementation and AA men receiving placebo, the team found an overlap of 346 genes. This finding suggests that a substantial number of genes that are differentially expressed across racial groups can be affected by a brief (2-month) course of vitamin D₃ supplementation in AA patients.

This research is an important step in understanding the molecular underpinnings of health disparities in prostate cancer. Further clarification of race-based transcriptome differences and the role of vitamin D in prostate tissue may lead to use of vitamin D₃ supplementation as an early-stage therapeutic option in prostate cancer. Furthermore, results from studies among AA and EA women with breast cancer could extend these findings because breast cancer, like prostate cancer, is an endocrine cancer, with many similarities including sensitivity to vitamin D.

An accompanying editorial by Batai K and Kittles RA, "Can vitamin D supplementation reduce prostate cancer disparities?" was published in the same issue of *Pharmacogenomics* (volume 17, number 11, 2016, pages 1117-1120).