NEXT GENERATION SEQUENCING OF MICROBIOMES IN POST-DRE URINE SAMPLES OF PROSTATE CANCER PATIENTS

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Background: Pathogenic microorganisms could be responsible for inflammatory processes in the prostate (1). Bacterial populations were observed according to different inflammatory and tumor conditions in prostate cancer (PCa) samples, which may promote the development of cancer (2-4). Most of these studies use polymerase chain reaction (PCR) techniques to identify specific microorganisms. We investigated microorganisms in post-digital rectal exam (DRE) urine samples from PCa patients and a screening population with a PSA <1.5 ng/mL using PCR and NGS techniques.

Methods: This study was conducted under COMIRB #00-812. We identified 100 post-DRE urine samples collected from patients with PCa. 100 samples from our annual Prostate Cancer Awareness Week participants with PSA <1.5 ng/mL served as the control group without PCa. The 5-year risk of developing high-grade PCa in men with PSA <1.5 is low (5). Microbial PCR and NGS was performed by MicroGen DX. Universal primers for 16S and ITS barcode loci were used for PCR amplification followed by NGS to characterize bacterial and fungal communities. Differential abundance of species between cancer and control groups was assessed using ANCOM (Analysis of Composition of Microbiomes) (6).

Results: The mean age and PSA of cancer group were 60.1±7.79 (range: 41 - 82) years and 8.81±2.65 (range 1.49 - 41.82) ng/mL. For the control group 64.6±10.14 (range: 27 - 85) years and 0.67±0.35 (range: 0.04 – 1.42) ng/mL. NGS informative rates were 40% and 56% for PCa and control groups, respectively. In the cancer group, 5, 19, 9, and 7 had Grade Group (GG) 1, GG2, GG3, and GG5 PCa, respectively. Figure shows a stacked bar plot of relative abundances of the 19 most common bacterial species found in PCa and control samples. ANCOM identified *Cutibacterium acnes* (P < 0. 05) and *Finegoldia magna* (P < 0.05) as significantly more abundant in cancer samples compared to controls, especially for high grade cancers. PCR informative rates were 3% and 8% for cancer and control groups, respectively. One patient with GG2 PCa had *Klebsiella pneumoniae*, a second patient with GG3 PCa had *Gardnerella vaginalis*, *Prevotella bivia*, and *Ureaplasma parvum*, and a third patient with GG5 PCa had *Gardnerella vaginalis* and *Ureaplasma parvum*. PCR detected *Escherichia coli*, *Prevotella bivia*, *Mycoplasma hominis*, *Gardnerella vaginalis*, and *Ureaplasma parvum* in the control samples.

Conclusions: NGS has shown higher sensitivity for identifying microbes in post-DRE urines samples than the PCR technique. *Finegoldia magna* is responsible for the production of equol, a soy metabolite associated with lower risks of breast and prostate cancer (7). *Cutibacterium acnes* strains can be divided into the major types IA, IB, II, and III. Type II is reported as the most prevalent type in prostate tissue samples from patients with PCa (8). Further investigation is needed to elucidate mechanisms of cancer induction.

References:
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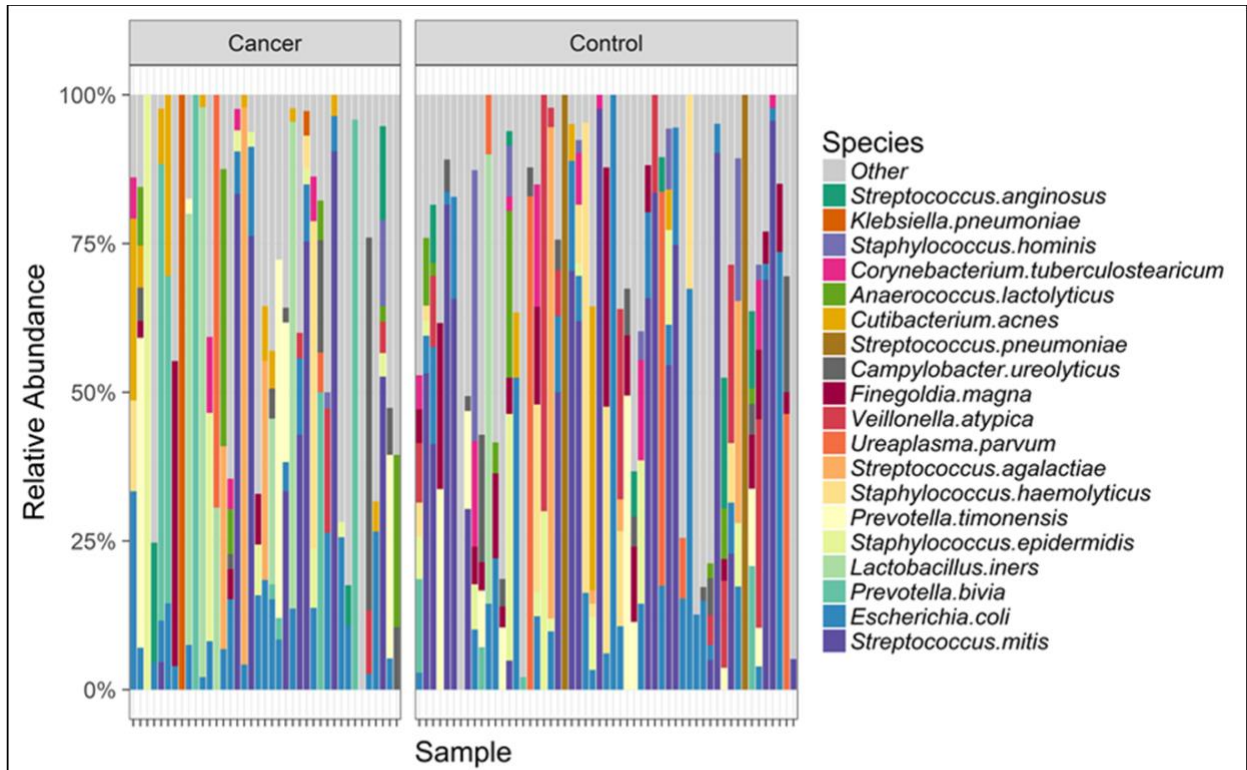   (8) Davidsson, Infect Agents Cancer. 11:26, 2016

Figure: Stacked bar plot. Each sample is represented by a unique location on the x-axis. The y-axis sums to 100% and coloring is proportional to species abundance in each sample. Samples are faceted by group (i.e. Cancer/Control)
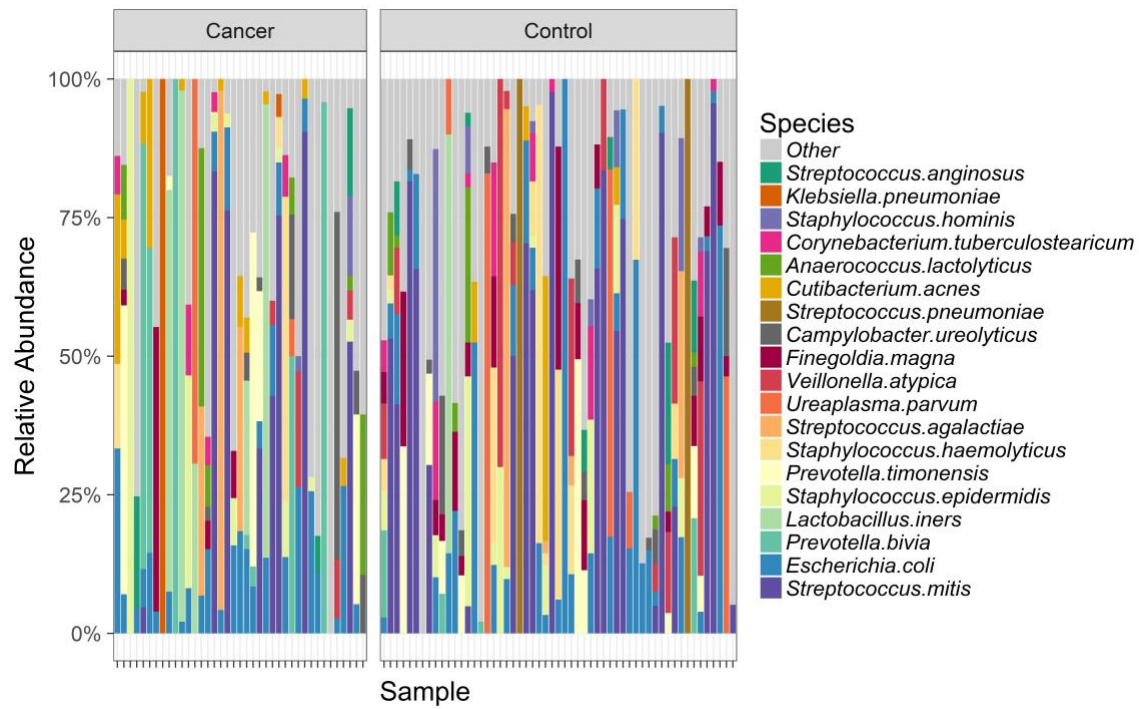
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