Impact of Chemokine Single Nucleotide Polymorphism (SNP) Pair Jointly Modified Prostate Cancer Risk among men of African Descent.

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INTRODUCTION

Prostate Cancer as a Public Health Issue

- African American men are more likely to receive a PCa diagnosis than European American men and twice more likely to die from the disease.

Chemokines Role in Prostate Cancer

- Chemokines belong to a family of small chemoattractant cytokines that mediate their effects by binding to protein-coupled receptors.
- Chemokines play role in cancer development and disease progression by promoting blood vessel formation, tumor growth and tumor spreading.
- Genetic alterations detected in coding and regulatory regions of chemokine associated genes may alter macromolecules (mRNA/protein express, chemokine-chemokine receptor production/function, protein-protein interactions), cellular behavior and ultimately modify PCa susceptibility.

Prior Lab Discoveries

- Previously, our lab demonstrated the sequence variants in chemokine ligand 5 and its receptor were individually related to prostate cancer development among men of African descent.

Table 1. Chemokines Studied in Literature

Gene	Cases/Controls	Disease	Reference
CCR7 CCL5 CCR9	279/535	Prostate Cancer	Jones, 2011
CCL4	861/1192	Oral Cancer	Lien, 2017
CCL4	346/1200	Hepatocellular Carcinoma	Wang, 2017
IL-8	300/300	Oral Cancer	Genet, 2016

RESEARCH GAP

The impact of two genetic alterations detected in chemokine and chemokine receptor genes in relation to prostate cancer (PCa) susceptibility remains understudied.

RESEARCH OBJECTIVES

Evaluated 1176 pairwise joint modifying effects of chemokine associated sequence variants in relation to PCa risk among men of African Descent.

MATERIALS AND METHODS

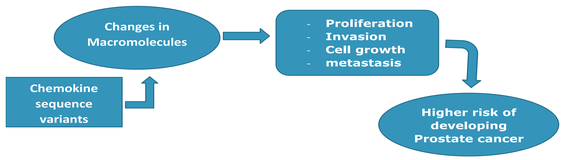
Study Design

- Using a case-control study design, we evaluated joint modifying effects of 49 sequence variants detected in chemokine associated genes in relation to PCA risk.
- 814 Men of African Descent (279 cases, 535 controls) were recruited from cancer screening programs, hospitals, or cancer centers located in the Washington D.C., South Carolina, and Kingston, Jamaica.

Statistical Design

- Plink was used to analyze 1176 pairwise interactions (http://zzz.bwh.harvard.edu/plink/).
- Compared the frequency distribution of genotypes between cases and controls using the chi-square and Fisher's Exact test.
- Risk estimates associated with inheritance of at least one minor chemokine-associated sequence variant allele were expressed as odds ratios (ORs) and corresponding 95% Confidence Intervals (95%CI) using unconditional multivariate logistic regression models, adjusted for age.
- Entropy-based Multiple Dimensionality Reduction with permutation testing was used to validate the top SNP pair.

HYPOTHESIS



RESULTS

Table 2. Top 20 Joint modifying effects of Chemokine SNP pairs on PCa using a multiplicative scale

Gene_dbSNP ID#	Adj. Overall Model (P-value)	Adj. Dominant Model (P-value)
CCL21_rs2812378 CCL5_rs3817655	0.0001	0.0005
CCL21_rs11574916 CCL5_rs2107538	0.0017	0.0008
CCL5_rs2107538 CCL21_rs2812378	0.0003	0.0017
CCR7_rs3136685 CXCL12_rs1801157	0.0027	0.0028
CCR6_rs3093024 CCL17_rs11076191	0.0210	0.0044
IL8RB_rs4674257 CCL25_rs7259568	0.0139	0.0050
IL8RB_rs4674259 CCL25_rs7259568	0.0173	0.0057
CCL17_rs223895 CCR9_rs41289608	0.0131	0.0065
CCR6_rs2023305 CCR4_rs6550178	0.0018	0.0067
CCR4_rs7632357 IL8RB_rs11574752	0.0073	0.0143
CCL1_rs2282691 CCL25_rs7259568	0.0202	0.0177
CCR9_rs41289608 CCL17_rs11076191	0.0285	0.0182
CCR6_rs3093023 CCL25_rs2032887	0.0337	0.0190
CCL2_rs1024611 CCL25_rs7259568	0.0354	0.0231
CXCL12_rs2839695 CCR9_rs1488371	0.0266	0.0231
CCR5_rs2227010 CCR6_rs1556413	0.1603	0.0344
CXCL12_rs17880777 CCR9_rs1488371	0.0218	0.0344
CCL5_rs2280789 CCL21_rs11574916	0.0126	0.0392
CCR5_rs1799988 CCR6_rs2023305	0.2412	0.0402
CXCR7_rs1045879 CCL7_rs17809012	0.0334	0.0413

Table 3. Joint modifying effects of Chemokine SNP pairs on PCa using an additive scale

Gene_dbSNPID#	# Variant Alleles	OR_{adj} (95% CI)	p-value	P-trend
CCL21_rs2812378 CCL5_rs3817655	0	1.00		0.049
	1	1.62 (0.87, 3.01)	0.1280	
	2	1.40 (0.76, 2.59)	0.2790	
	3–4	0.77 (0.39, 1.52)	0.4500	
	1 SNP w/ ≥1 var allele(s)	1.85 (1.02, 3.36)	0.0001	
	2 SNPs w/ ≥1 var allele(s)	0.86 (0.47, 1.58)	0.0001	
	2 SNPs w/ ≥1 var allele(s)	1.14 (1.12, 1.16)	0.0001	
IL8RB_rs4674257 CCL25_rs7259568	0	1.00		0.666
	1	1.28 (0.88, 1.85)	0.0078	
	2	0.52 (0.27, 1.01)	0.0439	
	3–4	0.54 (0.06, 5.05)	0.1922	
	1 SNP w/ ≥1 var allele(s)	1.21 (0.85, 1.74)	0.0190	
	2 SNPs w/ ≥1 var allele(s)	0.38 (1.12, 1.16)	0.0123	
	2 SNPs w/ ≥1 var allele(s)	0.35 (0.15, 0.82)	0.0123	
IL8RB_rs4674259 CCL25_rs7259568	0	1.00		0.626
	1	1.28 (0.89, 1.87)	0.0094	
	2	0.49 (0.25, 0.95)	0.0430	
	3–4	0.54 (0.06, 5.04)	0.1914	
	1 SNP w/ ≥1 var allele(s)	1.20 (0.84, 1.72)	0.0246	
	2 SNPs w/ ≥1 var allele(s)	0.38 (0.16, 0.91)	0.0155	
	2 SNPs w/ ≥1 var allele(s)	0.35 (0.15, 0.83)	0.0152	
CCR5_rs1799988 CCR6_rs2023305	0	1.00		0.001
	1	1.11 (0.70, 1.79)	0.0008	
	2	1.74 (0.92, 3.30)	0.0585	
	3–4	1.14 (1.12, 1.16)	0.0006	
	1 SNP w/ ≥1 var allele(s)	0.77 (0.50, 1.18)	0.0213	
	2 SNPs w/ ≥1 var allele(s)	1.38 (0.83, 2.30)	0.0004	
	2 SNPs w/ ≥1 var allele(s)	1.65 (1.09, 2.51)	0.0005	
CCL5_rs2107538 CCL21_rs2812378	0	1.00		0.050
	1	1.71 (0.92, 3.21)	0.0262	
	2	1.50 (0.81, 2.81)	0.5675	
	3–4	0.90 (0.45, 1.79)	0.0061	
	1 SNP w/ ≥1 var allele(s)	1.89 (1.03, 3.46)	0.0001	
	2 SNPs w/ ≥1 var allele(s)	1.01 (0.55, 1.87)	0.0005	
	2 SNPs w/ ≥1 var allele(s)	1.14 (1.12, 1.16)	0.0005	
IL8_rs2227545 CCR6_rs1012656	0	1.00		0.570
	1	1.34 (0.93, 1.95)	0.0092	
	2	1.04 (0.55, 1.97)	0.2778	
	3–4	0.68 (0.06, 8.25)	0.3665	
	1 SNP w/ ≥1 var allele(s)	1.33 (0.93, 1.90)	0.0091	
	2 SNPs w/ ≥1 var allele(s)	0.88 (0.38, 2.03)	0.0431	
	2 SNPs w/ ≥1 var allele(s)	1.14 (1.12, 1.16)	0.0459	
CCR6_rs3093023 CCL25_rs2032887	0	1.00		0.742
	1	1.36 (0.93, 2.00)	0.0394	
	2	0.95 (0.55, 1.66)	0.2168	
	3–4	0.88 (0.26, 2.90)	0.4193	
	1 SNP w/ ≥1 var allele(s)	1.34 (0.92, 1.94)	0.0287	
	2 SNPs w/ ≥1 var allele(s)	0.80 (0.42, 1.53)	0.0417	
	2 SNPs w/ ≥1 var allele(s)	0.68 (0.36, 1.25)	0.0408	
CXCL12_rs2839695 CCL25_rs2032887	0	1.00		0.017
	1	1.07 (0.74, 1.56)	0.676	
	2	0.55 (0.29, 1.05)	0.0184	
	3–4	0.21 (0.02, 1.83)	0.1403	
	1 SNP w/ ≥1 var allele(s)	1.07 (0.74, 1.53)	0.5831	
	2 SNPs w/ ≥1 var allele(s)	0.38 (0.18, 0.81)	0.0010	
	2 SNPs w/ ≥1 var allele(s)	0.37 (0.17, 0.77)	0.0009	

Table 4. Interactions of Inflammatory associated SNPs as predictors of PCa using MDR among men of African Descent

Best Model (dbSNPID#)	# of Interactions	Bal Accuracy	CVC	Pt P-value
One Factor CCL5_rs3817655	49	0.5687	10	0.0045
Two Factor CCL5_rs3817655 CCL21_rs2812378	1176	0.5756	10	0.0155
Three Factor CCL5_rs3817655 CCL21_rs2812378 CCR7_rs3136685	18424	0.5917	9	0.0012

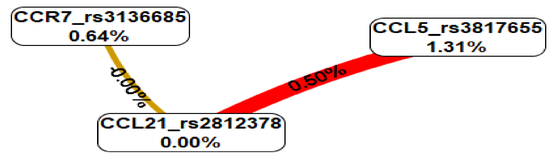


Figure 1. Entropy graph of Chemokine gene-gene interactions among men of African Descent. MDR analysis revealed a significant two-way interaction model for (CCL5 rs3817655 and CCL21 rs2812378).

CLINICAL IMPACT

Table 5. Clinical Relevance of Chemokines in Study Trials

Target	Drug	Conditon	Status	Study Description	Company
CCL2	Carlumab	Prostate Cancer Cardiovascular disease	completed	safety and effectiveness of the study drug carlumab in participants with metastatic castrate-resistant prostate cancer.	Orange City, Florida, United States
CCR5	Maraviroc	Colorectal Cancer Neoplasm Metastasis Liver Metastases	completed	Inhibition of CCR5 molecule leads to a reduction in growth signals for tumor cells and subsequent slowed or halted tumor growth	National Center for Tumor Diseases
CCL21	Biological: autologous dendritic cell-adenovirus CCL21 vaccine	Lung Cancer	Phase I	to determine the effects a tumor vaccine used in combination with GM.CD40L and CCL21 have on the patient and their cancer.	H. Lee Moffitt Cancer Center and Research Institute
CXCR2		Pancreatic Cancer	completed	The investigators hypothesize that the CXCR2 ligands/CXCR2 biological axis plays an important role in promoting angiogenesis in PC.	Mayo Clinic Jacksonville, Florida, United States

SUMMARY

- Out of 1176 SNP pairs, 38 chemokine SNP pairs significantly modified PCa risk.
- A complex interaction along the CCL5-CCL21 axis (rs3817655, rs2812378) was selected as best two-factor predictor of PCa using Multifactor Dimensionality Reduction (MDR) based on a 100% CVC, 58% balance accuracy and permutation p-value of 0.0160.
- The synergistic interaction between CCL5-CC21 is primarily driven by CCL5 rs3817655.

FUTURE DIRECTIONS

- Evaluate whether CCL21 and CCL5 and their corresponding receptors are dysregulated in biospecimen collected from PCa patients.
- Identify and validate chemokine SNP pairs and other complex gene-gene interactions using advanced bioinformatics techniques [e.g., statistical epistasis networks (SEN), Computational Evolutionary Systems, and Random Forest].

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