An Immunogenetic Approach to Dissecting the Biology of Prostate Cancer

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Abstract
Prostate cancer disproportionately affects African American men over the age of 50. This project was done under the hypothesis that there will be common genetic variations in individuals that have prostate cancer. Given that race is not a biological construct, this research project sought to stratify the prostate cancer population based on the oldest polymorphism known, the ABO blood groups. A polymorphism is a genetic variation that occurs in greater than 1% of the population. There were 96 prostate cancer DNA samples collected from a case and control study. RNA primes were designed to form the DNA sequence that, after being cut with certain restriction enzymes, could determine the different blood groups within the genome. In order to perform ABO genotyping, the polymerase chain reaction process, restriction enzyme digestion, and gel electrophoresis were utilized.

Introduction
Prostate cancer is a disease in which the cells of the prostate gland, a part of the male reproductive system, multiply and multiply uncontrollably. Aside from lung cancer, prostate cancer is responsible for the majority of cancer-related deaths. Most frequently this disease affects African American men over the age of fifty, but there is no true cause. Many men, who develop prostate cancer, show no symptoms, never diagnosed, and eventually die of complications. The disease is diagnosed due to elevated an amount of Prostate Specific Antigen (PSA). There are numerous speculations that the disease is linked to genetics, diet, or lifestyle, but there is no answer to why it is a serious threat to only African American men. Patients that succumb to diseases are normally conditioned according to racial background. Because race is not a biological construct, there must be some other way to divide the affected population.

The ABO blood group system presents an easy and effective way of partitioning the affected population. Discovered in 1900 by Karl Landsteiner, this system divides humans into four principal groups, A, B, AB, and O. Those persons with type A blood have the A antigen on the surface of their red blood cells, those with type B blood have the B antigen. AB persons have both antigens, and those with type O have no antigens. In addition, persons with type A blood have anti-B antibodies, type B blood have anti-A antibodies, type AB blood have anti-A and anti-B antibodies, AB has no antibodies, and type O has anti-A and anti-B antibodies. Blood types are genetically inherited, but the environment can determine which blood types in a population will be passed on more frequently to the next generation. Specific environmental factors might be linked with increased or decreased susceptibility to certain diseases.

The overall goal is to eventually locate candidate genes within the newly partitioned prostate cancer population, and from there, link the disease to a specific genetic variation. This current project was a precursor that involved genotyping the prostate cancer samples, and partitioning them for further investigation.

Materials and Methods

Results

The following data was collected from the project: Blood Types of Prostate Cancer Patient Samples

<table>
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<th>Primer</th>
<th>Description</th>
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| 5’ TGT GCC TCT GGT TGG TTT CCC G 3’ | Primer 261 amplifies a 275 bp DNA fragment containing a section of DNA that is O specific within the ABO locus. If there is a deletion of nucleotide 261 the PCR product will have a cleavable site for the restriction enzyme Kpn I. Deletion of this nucleotide results in a frameshift mutation that is specific to the O allele. If nucleotide 261 exists, there will be no cleavable site for Kpn I. Therefore, if the fragment was unable to be digested by Kpn I (fragment at 275), the genotypes for the subject will be homozygous for O.
| 5’ CAA TGG GGG TCC TCT CTT GAG T 3’ (703) | Primer 261 amplifies a 275 bp DNA fragment containing an section of DNA that is O specific within the ABO locus. If there is a deletion of nucleotide 261 the PCR product will have a cleavable site for the restriction enzyme Kpn I. Deletion of this nucleotide results in a frameshift mutation that is specific to the O allele. If nucleotide 261 exists, there will be no cleavable site for Kpn I. Therefore, if the fragment was unable to be digested by Kpn I (fragment at 275), the genotypes for the subject will be homozygous for O.
| 5’ TGC TTC TTG GAG ATG TAG GCC 3’ | Primer 703 amplifies a 171 bp DNA fragment containing the nucleotide 703. If this nucleotide is a adenine residue, an A allele specificity cleavage site will be present with B and AB specificity if nucleotide 703 is an guanine residue the PCR product will have a cleavable site for the restriction enzyme Alu I. Therefore, if the fragment was not digested (fragment at 707) the genotypes can be AA, AB, or BB. Therefore, if the fragment was not digested (fragments at 171, 96, 75) the genotypes can include AB, or BB. Full digestion (fragments at 96, 75) will be called as a BB.

The genotypes of the isolated DNA samples were determined from patients of the Howard University Hospital.

Conclusions

The National Human Genome Center’s main focus is to find the causes of common variant chronic diseases that disproportionately affect African Americans and the African Diaspora. This case controlled experiment attempts to utilize one of the oldest polymorphisms known (the ABO blood group) and partition a targeted population. The ABO blood group in itself is a very unique system. It contains four principle types: A, B, AB, and O. The difference between these four groups depends on the order of two antigens and two antibodies responsible for the ABO blood group. For this study, 96 samples of isolated DNA were genotyped. The results of the genotyping established a concrete system that will be instrumental in dividing the targeted population.

Once genotyped, the National Human Genome Center will be able to group the samples by blood type and search for candidate genes for the disease. Candidate genes are genes located in a chromosome region and are suspected of being involved in the expression of a trait such as a disease. In this experiment, the disease being studied is prostate cancer. In recent studies, it has been hypothesized that prostate cancer disproportionately affects African American men over the age of 50. Discovery of a candidate genes for prostate cancer are already contributing to the growing knowledge of this disease. Using a biological marker to partition these candidate gene studies may allow researchers to gain valuable knowledge that will bring them closer to the causes of prostate cancer. At the same time, a profound revision of drugs for prevention and treatment of prostate cancer may be discovered.

References

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