ISID Scientific Exchange Fellowship Program ~ Final Report

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Analysis of Ethnic Differences in the Distribution of TNF-SNPs and Human Papillomavirus (HPV) Infection

Overview of Fellowship Project:

Infection with Human Papillomavirus (HPV) is known to play a central role in the development of cervical cancer [1,2]. Polymorphisms in the promoter region of the TNF- gene have been associated with high (SNPs -307) and low (SNPs -237) cytokine production, and these functional differences may modulate the magnitude of immunological response following HPV infection but results are contradictory [3-11]. The province of Misiones (Argentina) is considered a region with a high prevalence of cervical carcinoma (12/100,000) compared to the urban areas of the country (Buenos Aires 3/100,000) [12-14]. Within Misiones, different ethnic groups inhabit specific regions of the province. The Guarani Indian populations are concentrated in small communities in the rain forest, while the white populations (with a wide range of parental genetic contributions from Europe) live in the urban and rural areas [15-17].

The goal of this project was to analyze genetic variation in the TNF- promoter (SNPs -237, -243, -307, -375) and examine its potential association with HPV infection and cervical cancer among different ethnic groups from Misiones. To this end, we have analyzed a sample of 123 urban and rural women (admixed populations of European descent) and compared the data with previous work involving American Indians from the region [18]. This approach has allowed us to undertake risk analysis in different ethnic groups. Polymorphisms in this gene were determined through PCR amplification and direct sequencing [18]. Our results showed no association between the presence of the -307 and -237 SNPs and HPV infection. However, the SNP distribution was statistically significantly different between study populations. In particular, we observed low genetic diversity in Amerindians that may be a result of small population size and random genetic drift associated with their particular history, cultural and geographical isolation. This study of human genetic variation in the TNF- γενε ηασ προϖιδεδ new information about the genetic differences among Misiones populations. These differences may help us to better understand the role of genetic factors in the development of disease.

All work completed during the Fellowship period is being prepared for submission to a peer-reviewed journal.

Work Currently in Progress

During the Fellowship, we have had an opportunity to discuss new projects and implement pilot studies that have provided initial data from which to expand the collaboration in the future.

Perspectives on HLA-Linkage disequilibrium analysis: Variation in human major histocompatibility genes may influence the risk of cervical cancer development by altering the efficiency of the T-cell–mediated immune response to HPV antigens [19, 20]. The human TNF-a gene is located on chromosome 6 between the class I HLA-B and the class II HLA-DR loci. Multiple TNF- SNPs have been shown to be in non-random association with neighboring HLA genes. In particular, the presence of the -856 SNP has been associated with an Amerindian haplotype defined by HLA-Cw*0102, -B*1522, -DR*0407. [21]

In order to explore linkage disequilibrium with HLA alleles in Guarani and Admixed population from Misiones, we have developed a Real Time PCR assay for the identification of the SNP-856. The rationale behind this approach was the possibility of directing HLA extended analysis to only specific subset of individual who are carrying this SNP (as previously described by Baena et al., 2002). During the Fellowship, we successfully screened 90% of our samples for the -856 SNP. In contrast with previously targeted polymorphisms, the high frequency of the -856 SNP (30% of the population were carriers) has revealed it to be a more appropriate marker for linkage analysis.

Perspectives on cases and controls study design: The frequencies of the -307 SNP found in Admixed population (13% of women were carriers of the A allele) led us to explore its presence in a Case Group of women diagnosed with cervical cancer from Misiones. This work is currently being carried out at Misiones University.

Transfer of Technology

All methods and technologies employed during this study (Real Time PCR, Sequencing and Human Population Genetics analysis) are being transferred to the Lab. Biología Molecular Aplicada, Universidad Nacional de Misiones (Argentina).

Beyond Lab Duties

My training experience in the laboratory of Dr. Theodore Schurr has been enjoyable and challenging. The University of Pennsylvania (UPenn)

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Ines Badano, PhD Student

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Ms. Badano has a degree in Genetics (Licenciatura en Genetica) and is currently finishing her PhD program at the University of Buenos Aires, Argentina. Her research focuses on the detection and characterization of Human Papilloma virus infection (the etiological agent of cervical cancer) in aboriginal and non-aboriginal women inhabiting Misiones. Along with epidemiology she is interested in the relationship between human genetic markers and viral infection, in particular the genetic history of human populations and viral co-evolution. Her work is performed in the Laboratorio de Biologia Molecular Aplicada, National University of Misiones.

ranks among the top 10 universities in the USA and, with more than 150 research centers and institutes on campus, offers a great opportunity for interdisciplinary study. During my fellowship, I had the opportunity to attend several seminars, workshops and lab meetings, as well as informal brainstorming sessions with members of the laboratory. Moreover, since the Annual Meeting of the American Society of Human Genetics was held in Philadelphia this year, I had the unique opportunity to attend to this important event. Besides the academic opportunities, the beautiful urban campus of UPenn, rich in green open spaces, museums, public art and architecture, made my stay a pleasant experience. I was privileged to meet the lab staff of Dr. Schurr, and, with their help, developed the skills to perform the experiments and enhance my understanding of human population genetics analysis. It was, indeed a very productive experience. The Fellowship has enriched me in several ways, and I would like to thank the ISID for supporting this project. ❖

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