



The Local Genome Diversity Studies: Frequency of endometriosis related mutation in *TLR4* gene in the San Germán municipality of Puerto Rico



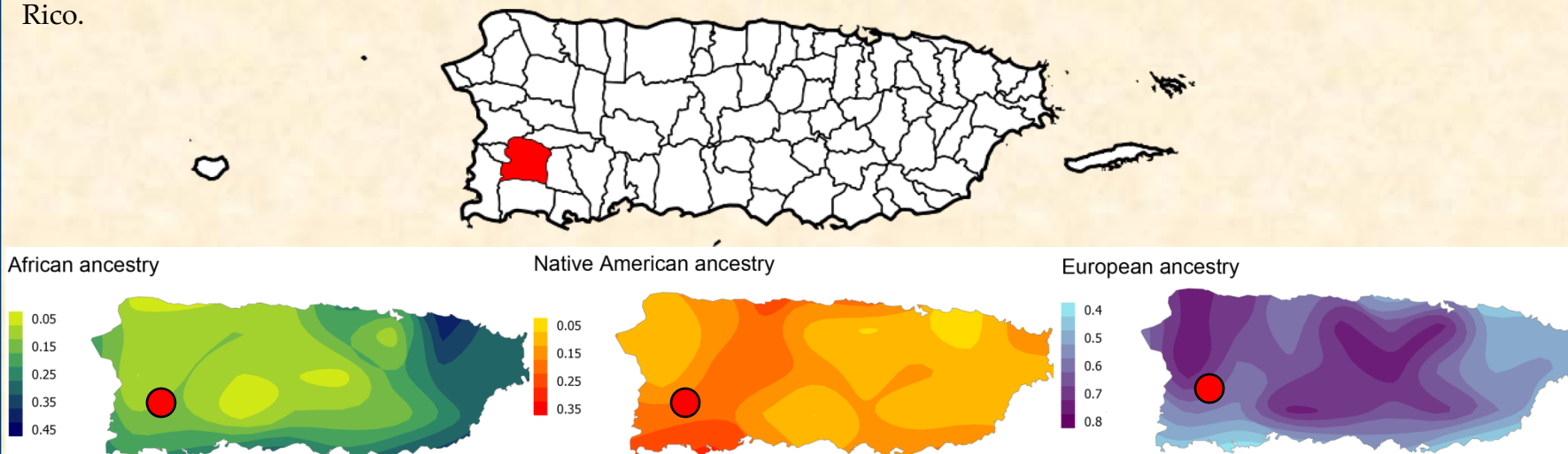
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Introduction

The Local Genome Diversity Studies is a novel instructional course cycle that will transition students from in-class learning to a mentored research experience through the description of the local genome diversity of the island. Due to the increase of availability of data through the next-generation sequencing technologies, the aim of the educational component is to expose students to the different aspects of genomics through a series of undergraduate courses that will encourage independent research, scientific reading, literature research, publication and responsible conduct in research. In the research component, the major goal is to map the local genome diversity of the island by:

- 1) Assembling a geographical representative sample of the Puerto Rican population in a form of a repository.
- 2) Identifying patterns of European, African and Amerindian admixture throughout the island using Ancestry Informative Markers (AIM's) and relate this distribution to the history of Puerto Rico.
- 3) Identifying locally polymorphic genetic variants, especially those related to disease, and describe their distribution and relevance to the history and epidemiology of Puerto Rico.
- 4) Creating an online resource with detailed interactive maps and data that features the genetic variation across Puerto Rico available for the academic community.

One of the projects within the LGDS is called **Adopt a Gene**, in which we will identify a gene related to a particular phenotype. We use genome browsers to identify beginnings and the ends of the gene, map regulatory elements, and identify population-specific polymorphisms to classify them according to the region and functional impact in the Puerto Rico.



Around the world, human genetic variation has been shaped by a long history of events like migrations. Geography can be accounted for the distribution of the human genetic variation and is supported by the substantial archeological evidence and historical sources. Puerto Rico is an ideal population to recognize and understand the impact of social and historical factors that may have shaped the distribution of genetic variation in a population. To understand and examine the interaction between genetic ancestry and social factors with health and disease, it is imperative to study the interaction of admixed populations. San Germán is a municipality located in the southwestern region of the island. It is the second oldest city in Puerto Rico, after the capital San Juan. According to some researchers, the historical beginnings of the municipality can be traced back to 1506, like Historian Aurelio Tió, claims that the municipality had its early origins in the surroundings next to the Guaorabo River, now Añasco River, in 1506.³

Population of San Germán	37,105 (U.S. Census Bureau, Census 2000)
Samples Collected	96 samples (Duplicate 192 samples)
Samples with DNA extraction	96 (Duplicate 192 samples)
Samples sequenced	4 (8 sequences)

Adopt a Gene: Toll-Like Receptor 4 (TLR4)

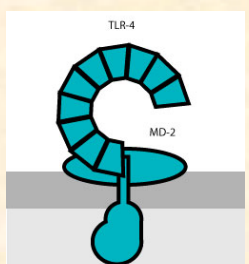


Figure 1. Scheme of the Toll-Like Receptor 4¹

Toll-Like receptors (TLR's) are a recognition and signaling system that was discovered in *Drosophila melanogaster* for playing a key-role in embryonic development. It was later found to have an important role in the defense against bacterial infections. There are 10 expressed TLR genes in humans and each protein recognizes a different microbe-associated molecular patterns and have limited specificity compared with the antigen receptors in the adaptive immune system. When in macrophages, TLR4 signals the presence of bacterial lipopolysaccharide (LPS) by associating with CD14 and MD-2 proteins¹. Activation of the TLR4 by LPS, results in an inflammatory response that alerts the host to the presence of a microbial infection and initiates an immune response.

Molecular Pathogenesis of Endometriosis; Toll-Like Receptor-4 A896G (D299G) Polymorphism: A Novel Explanation

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Figure 2. Paper that associates A896G polymorphism with endometriosis

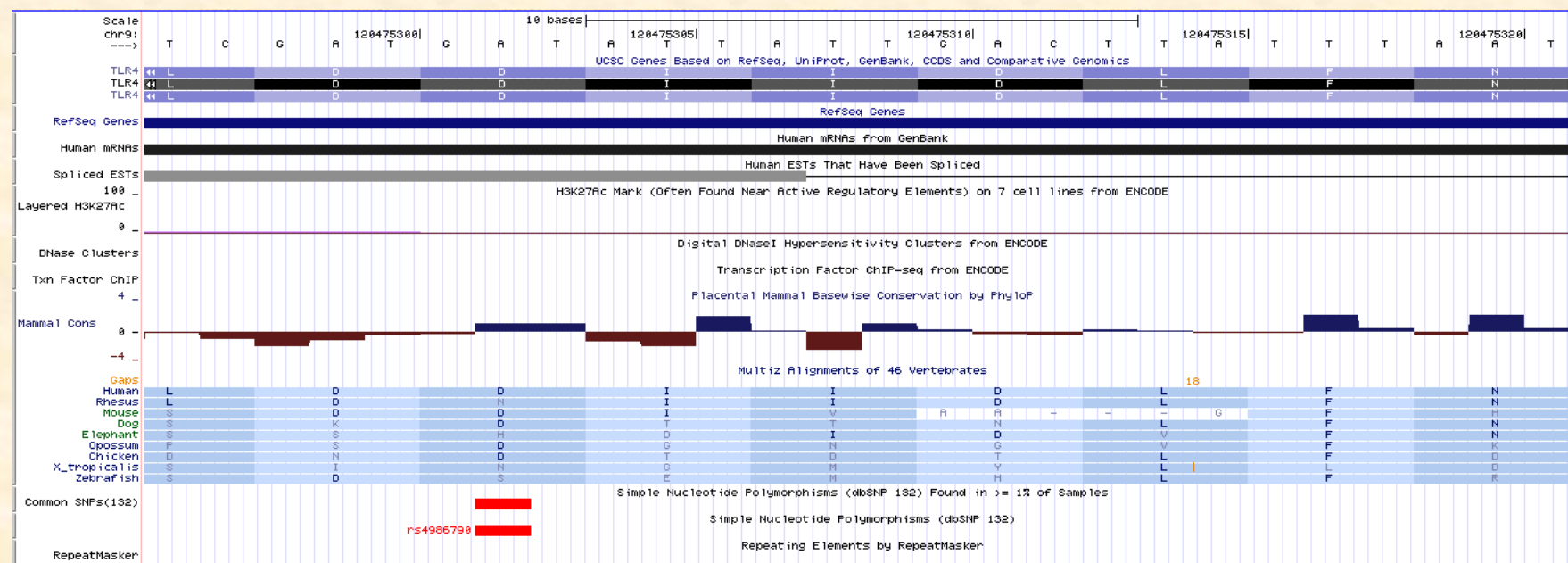


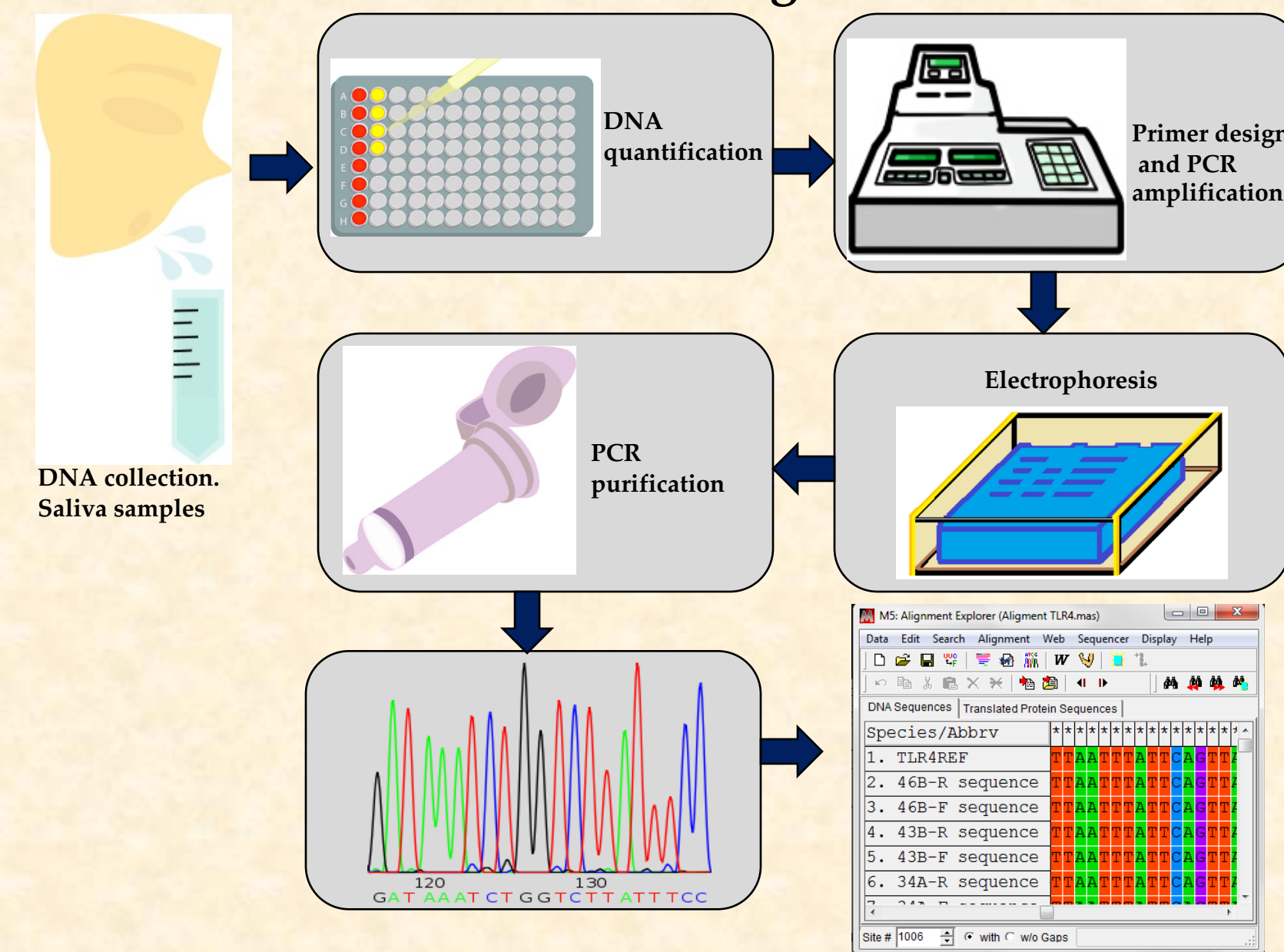
Figure 3. UCSC Genome browser view of the rs4986790

Endometriosis is a gynecological disorder in which the lining of the uterus, the endometrium, grows outside the uterine cavity causing several symptoms such as abdominal pain. This condition may lead to infertility. Researchers in Hyderabad, India have studied the polymorphism A896G (rs4986790) as a factor involved in endometriosis. The A896G polymorphism has been associated to hyporesponsiveness of the gene, allowing the persistence of inflammation. They analyzed the association of TLR4 A896G polymorphism in India individuals suffering from endometriosis. They found that the polymorphism was present in a higher percentage (3.5%) in patients suffering from endometriosis compared to the healthy controls.²

Hypothesis

By taking a representative sample of the municipality of San Germán, we will uncover the genetic variation of the *TLR4* gene and the frequency of the polymorphism in the municipality

Methodologies



Results

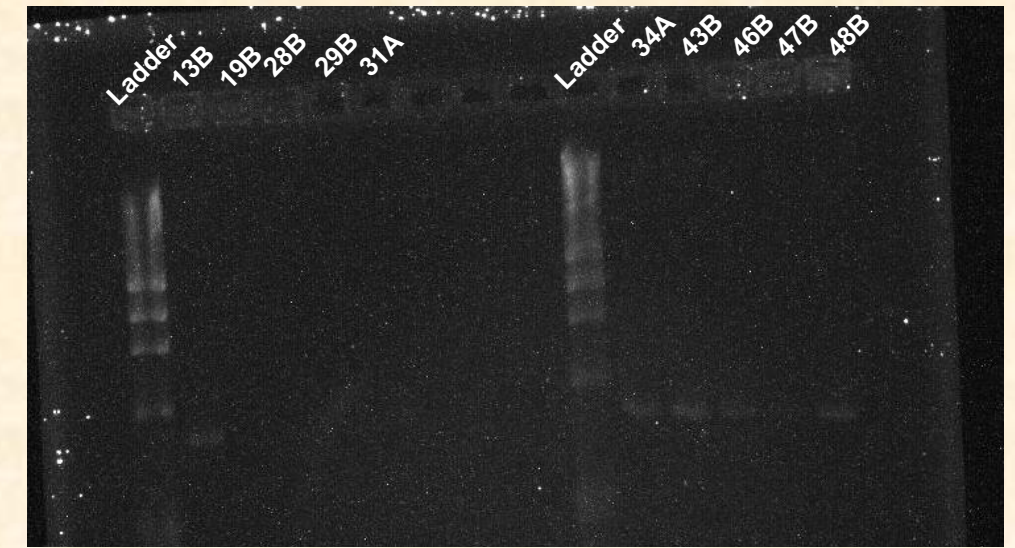


Figure 4. Gel of PCR purified samples.

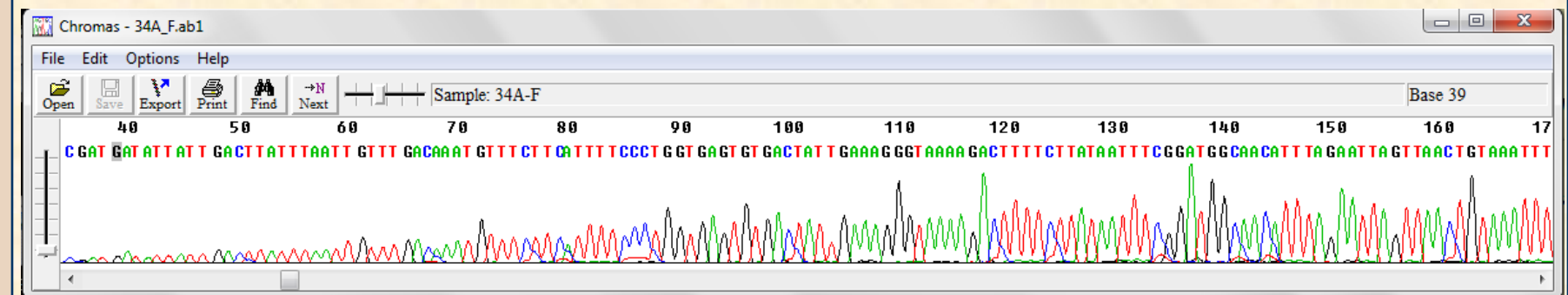


Figure 5. DNA chromatogram of sample 34A.

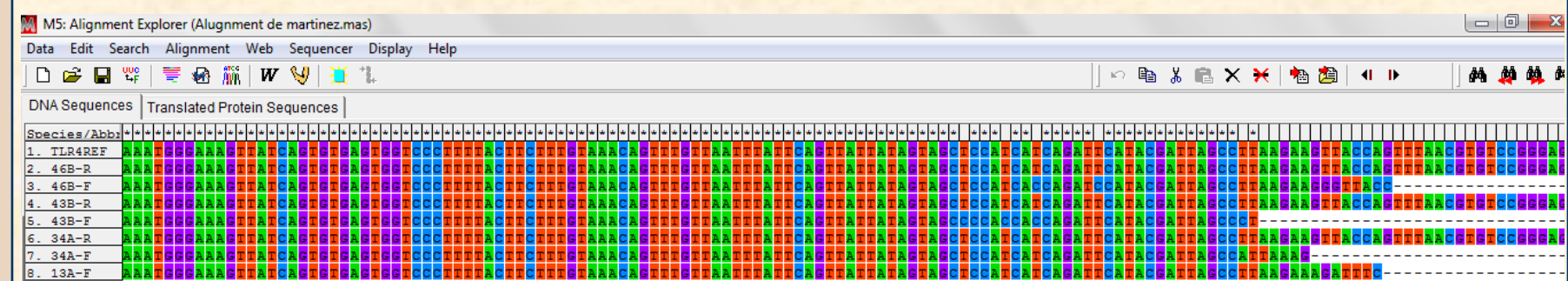


Figure 6. Alignment of the sequences using a TLR4 reference sequence.

Conclusions

- 1) The A896G (rs4986790) polymorphism was not found in the four sequenced samples.
- 2) No other polymorphism was found in the fragment sequenced.
- 3) We will amplify the other samples and sequence them to see if the polymorphism is present in the representative sample

References

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- 2) Latha M., Vaidya S., et al. (2011) Molecular Pathogenesis of Endometriosis; Toll-Like Receptor-4 A896G (D299G) Polymorphism: A Novel Explanation. Genetic Testing and Biomarkers 15:181-184
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Acknowledgements

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