CHARACTERIZATION OF NEW MICROSATELLITE MARKERS OF HUMAN X-CHROMOSOME.

Simas, M.C.C.1; Mello, I.C.T.2; Moura-Neto, R.S.1; Silva, R.2
1Instituto de Biologia and 2Instituto de Biofísica Carlos Chagas Filho, Universidade Federal do Rio de Janeiro, Rio de Janeiro, Brazil

INTRODUCTION: The X-chromosome markers, such as STR (short tandem repeats), are widely used in population genetics, anthropological studies, kinship and paternity tests. Next Generation Sequencing technology improves identification of polymorphisms because of its capacity of analyzing several different loci sequences from the same individual. These sequences can be compared and, consequently, produce reliable results. OBJECTIVES: To find polymorphisms to better characterize STR in the X-chromosome markers, predicted by in silico mining, in a sample population of Rio de Janeiro, Brazil. MATERIALS AND METHODS: Samples were chosen in order to cover all the alleles found in previous study from our group. We quantified those samples using NanoDrop Spectrophotometer (Thermo Scientific). We designed primers using OligoPerfect™ Designer (Invitrogen) and Geneious Software and sorted by physicochemical properties. We prepared the library using QIAcube (Qiagen) with GeneRead Library Prep L kit (Qiagen) and quantified the library by qPCR. Then, we amplified by emulsion PCR and sequenced with Ion PGM (Life Technologies). We analyzed the data using CLC Genomics Workbench V.7.5.6, mapping the reads against the reference genomic sequence hg19. DISCUSSION AND RESULTS: For all the 20 different markers, 08 markers were sequenced in a pool of 60 samples. We obtained a total of 20.000 reads. After mapping, 77% of the reads were aligned with the reference sequence with 95% of accuracy. To increase the number and size of reads and consequently the coverage, a new sequencing step will be performed. CONCLUSIONS: Genetic characterization of human X-chromosome from different populations can contribute to population studies, kinship analysis and determination of risk for certain diseases.

Keywords: Microsatellites, X-chromosome, NGS
Supported by: FAPERJ, CNPq, CAPES