PARKINSON'S PROGRESSION MARKERS INITIATIVE

Play a Part in Parkinson's Research

Exome-sequencing of PPMI samples

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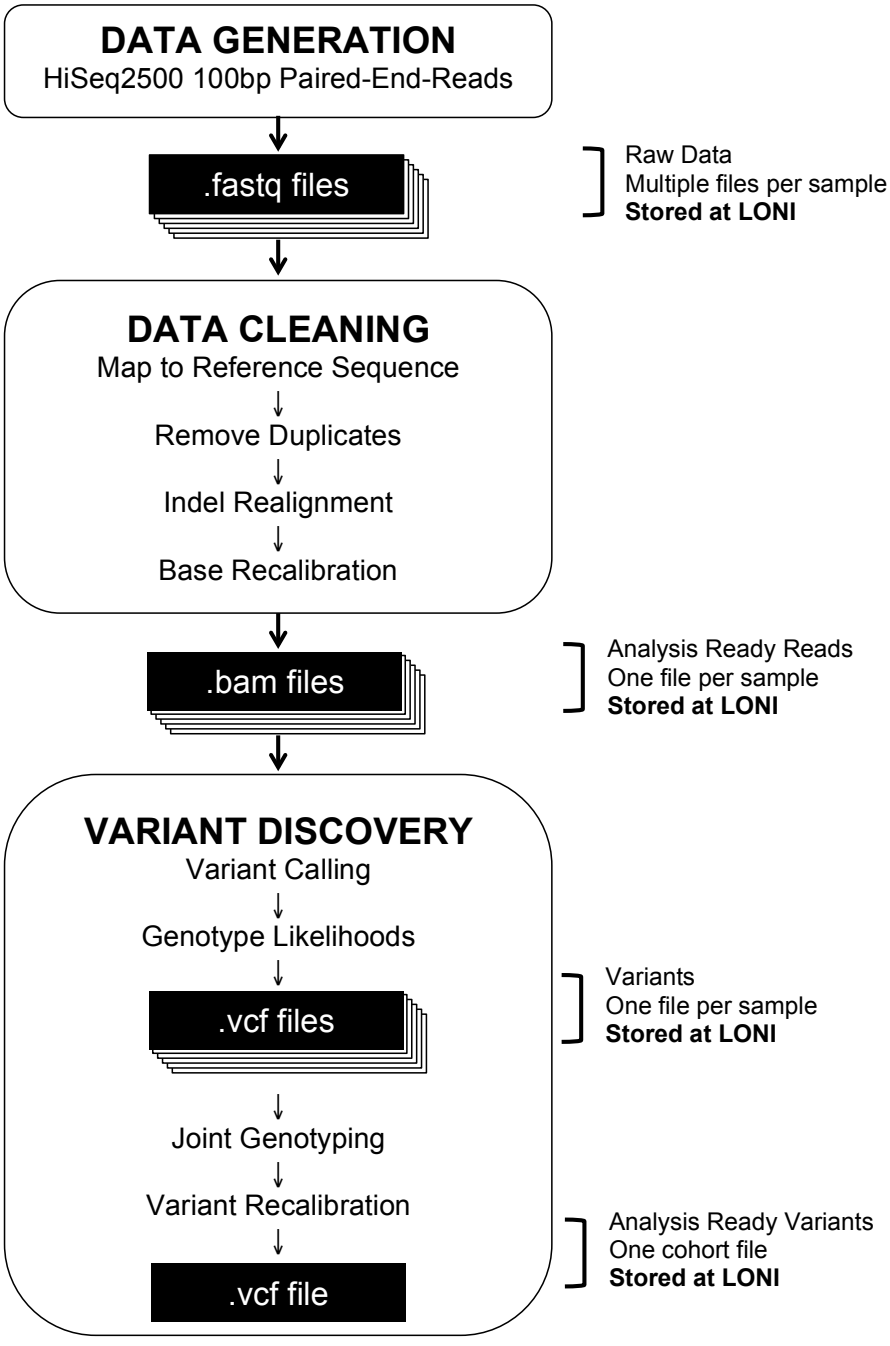
Summary

Exome sequencing was performed on whole-blood extracted DNA samples collected according to the PPMI Research Biomarkers Laboratory Manual using Illumina Nextera Rapid Capture Expanded Exome Kit. Nextera Expanded Exome targets 201,121 Exons, UTRs and miRNA and covers 95.3% of Refseq exome. >340,000 probes are constructed against the human NCBI37/hg19 reference genome. Targeted genomic footprint is 62Mb.

Methods

Library preparation for next-generation sequencing using Nextera Rapid Capture Expanded Exome Kit was performed per manufacturer's protocol (Illumina, Inc. San Diego). Exome-enriched libraries (multiplexed sets of 12 samples) were sequenced on the Illumina HiSeq 2500 sequencing platform using 2 x 100 bp paired-end read cycles. An overview of the data processing and available files is shown in the figure below. Briefly, paired-end sequence reads (fastq files) were aligned using BWA [1] against the reference human genome (UCSC hg19). Duplicate read removal; format conversion and indexing were performed with Picard (picard.sourceforge.net/index.shtml). The Genome Analysis Toolkit (GATK) [2–4] was used to recalibrate base quality scores and perform local re-alignments around indels for the aligned sequencing reads (per subject bam file). Variant calling and genotype likelihoods were generated per subject using the GATK HaplotypeCaller (per subject genomic vcf file). GATK CombineGVCFs and GenotypeGVCFs were used to perform joint genotyping for the cohort from the set of per subject genomic vcf files. Variant filtering was then applied using the GATK Variant Quality Score Recalibration tools (cohort vcf file). Subject quality control was performed based on variant call-rate, heterzygosity rate, gender check, relatedness/duplicates and population outliers using PLINK [5].



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