Targeted Next-Generation Sequencing revealed rare variations in two siblings affected by late onset Parkinson's disease.

Cavalcanti F.[1], La Cognata V.[2], Bellofatto M.[3], Criscuolo C.[3], De Rosa A.[3], De Michele G.[3], Citrigno L.[1]

[1]Institute for the Biomedical Research and Innovation IRIB, National Research Council CNR, Mangone (CS), and [2]Catania, Italy

[3]Department of Neurosciences and Reproductive and Odontostomatological Sciences, Federico II University, Naples, Italy

Background

Genetic variations and aberrations in the form of mutations have been found to be significant contributors to the risk of Parkinson's disease with implications in the pathogenesis by means of Next-Generation Sequencing (NGS) screening. This new approach can be used to genetically define unresolved PD clinical cases.

Objective

The aim of this study was to assess the genetic background of a familial case of two brothers affected by late onset PD and focus on their common genetic variants, using a targeted NGS panel containing 42 known PD-causative genes.

Methods

The two brothers affected by late onset PD were clinically assessed at the Department of Neurosciences, Federico II University, Naples. Both siblings presented l-dopa responsive parkinsonism and developed psychosis, behavioral disorders and dementia. After DNA extraction by standard methods and quantization using the Qubit instrument (ThermoFisher Scientific), high coverage targeted NGS data were generated at the NGS-Core of IRIB, CNR, Mangone (CS) by an amplicon-based approach(Fig.1). We used a custom-made panel comprising 42 PD-related genes. The enriched libraries were sequenced on the Ion Torrent Personal Genome Machine (PGM) system from ThermoFisher Scientific. The primary bioinformatics analysis was carried out using Ion Torrent Suite 5.10. Annotation and filtering/priorization of single-nucleotide variations (SNVs) and copy number variations (CNVs) discovered was made by Annovar.

Results

Multigene panel sequencing generated a median of 5.85 $*10^6$ reads per run, with a mean depth close to 500 reads, a mean read length of 375 called bases and a chip loading of 71% (Fig.2). After the bioinformatics analysis, among the common variants presented by the two brothers (Fig.3), we focused on the heterozygous missense variant c.2710A>G (p.Lys904Glu) in the alpha-synuclein binding domain of *SNCAIP* gene with MAF<0.01, reported as damaging. In addition, we identified 4 common CNVs with high confidence and precision: a deletion in the *ATP13A2* gene (chr1p36.13), a deletion in *DNAJC6* gene (chr1p31.3), a deletion in *ADHC1* gene (chr4q23), and a 12Mb deleted region in 22q11.21-21q12.3. This region expands between *COMT* and *FBXO7* genes, both associated with PD and contains the deletion associated with the 22q11.2 syndrome, a genetic risk factor for early-onset PD.

Conclusions

Our study suggest that multiple genetic variations all together might contribute to PD disease development or progression, since research over the past years revealed the important role of *SNCAIP, FBXO7, DNAJC6*, *ATP13A2* genes respectively in in the major pathways that are central in the pathology of PD/parkinsonism: mitophagy, synaptic vescicle endocytosis, mitochondrial and proteosomal, and lysosomal dysfunction.

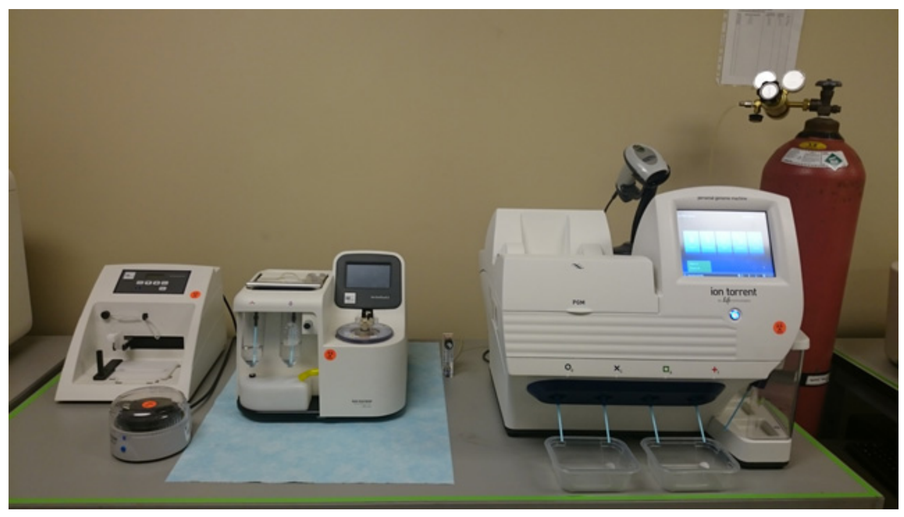

Fig.1: Ion PGM platform

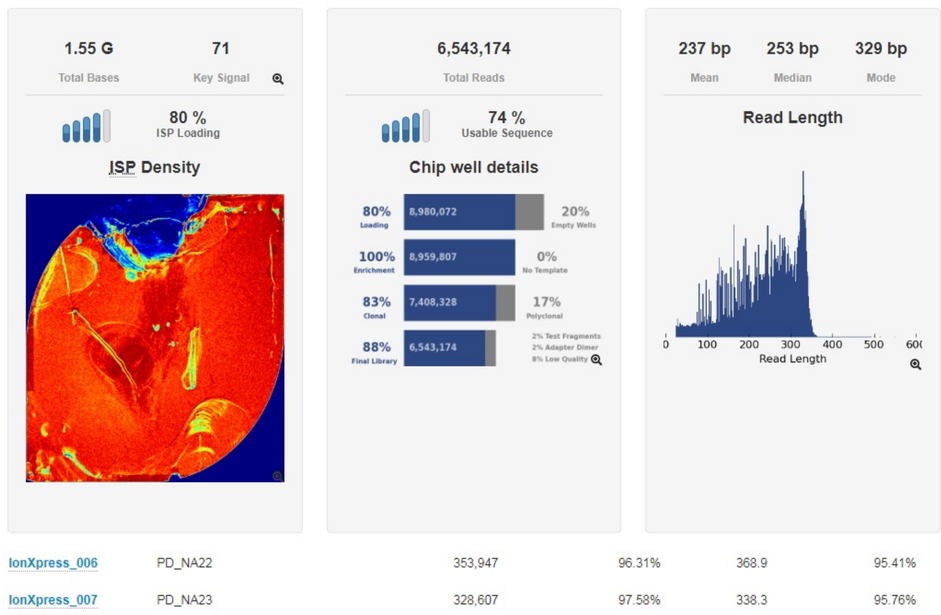

Fig.2: Sequencing run quality metrics

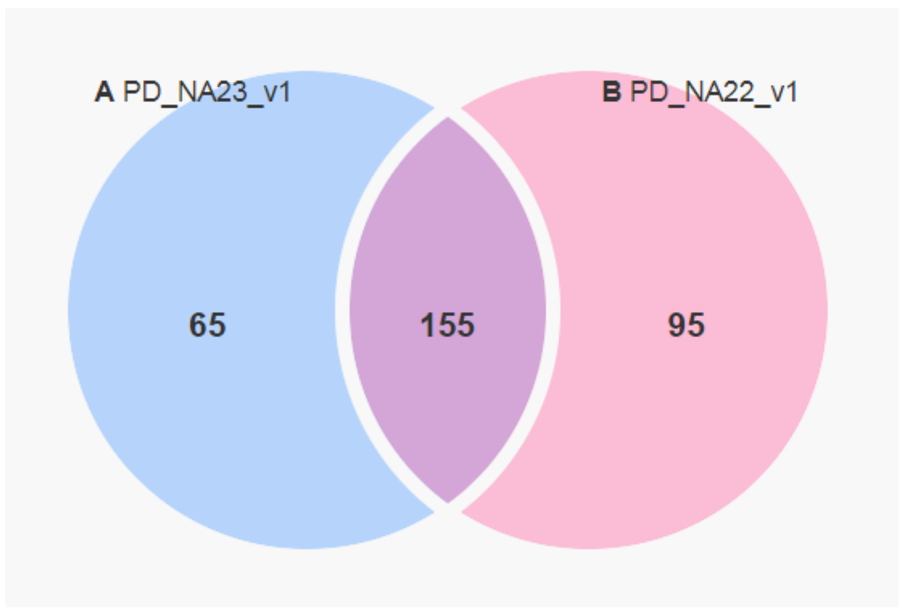

Fig.3: Common variants shared between the two siblings

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