Whole exome sequencing in patients with Primary Generalized Glucocorticoid Resistance, who did not have mutations in the NR3C1 gene

Amalia Sertedaki1,2, Alexandros Polyzois3, Nicolas C. Nicolaides1,2, Dimitris Thanos3, Evangelia Charmandari1,2

1Division of Endocrinology, Metabolism and Diabetes, First Department of Pediatrics, University of Athens Medical School, ‘Aghia Sophia’ Children’s Hospital, Athens, 11527, Greece; 2Division of Endocrinology and Metabolism, & 3Institute of Molecular Biology, Genetics and Biotechnology, Biomedical Research Foundation of the Academy of Athens, 11527, Greece

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Background: Primary Generalized Glucocorticoid Resistance (PGGR) is a rare, familial or sporadic condition, characterized by generalized, partial tissue insensitivity to glucocorticoids. The molecular basis of this condition has been ascribed to mutations in the NR3C1 (human glucocorticoid receptor, hGR) gene, which impair the molecular mechanisms of hGR action and decrease tissue sensitivity to glucocorticoids. However, a considerable number of patients with PGGR do not have mutations in the NR3C1 gene.

Objective and Hypotheses: Using whole exome sequencing, we investigated whether other genes are implicated in the pathogenesis of PGGR.

Patients and Methods: Eleven adult patients (age range: 18-48 years; 6 males, 5 females) with PGGR, who did not have mutations in the NR3C1 following Sanger sequencing, and two patients with PGGR harbouring two mutations (one missense mutation and a 5 bp deletion) of the NR3C1 gene (positive controls) underwent whole exome sequencing on an Ion Proton platform (ThermoFisher Scientific USA).

Results: Each exome sequence revealed the presence of approximately 55000 variants. Using a cut off value of 100 reads/variant, a total number of 507 non synonymous and frameshift mutations were detected in all patients (Fig.1). These mutations corresponded to 390 genes involved in 5 different pathways (Fig.2), one of which was that of steroid hormone biosynthesis (CYP1B1, CYP3A7, AKR1C4, UGT2A3; Fig.3). The mutations detected in these four genes were shown to be known polymorphisms and were present in all 13 samples. One mutation of the UGT2A3 gene was detected in only one patient, the c.1480T>G p.Phe494Val and was reported as damaging, possibly damaging and polymorphism by three in silico tools.

Nineteen of the 390 genes were found to be regulated directly by TP53 possibly indicating the presence of a cascade. One mutation of the GP6 gene present in all patients was not annotated. The presence of mutations in the genes HSPA9A1, NCOA1, SMARCA4, NCOA2, JUN, UBC, CREBBP, NFkB1, RELA and NCOA3 (functional partners of the NR3C1 after searching the STRING database; Fig.4) was examined and no pathogenic variants were detected.

No NR3C1 mutation was detected in any of the patients, whereas the mutations previously identified by Sanger sequencing were also detected by exome sequencing.

Conclusions: Whole exome sequencing may allow us to expand the spectrum of genes associated with PGGR. Further bioinformatic analysis is required to establish pathogenic variants in genes related to this condition.