Functional validation of a mitochondria-specific polygenic risk score in patient-based models for stratification of idiopathic Parkinson's disease

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Fonds National de la Recherche Luxembourg

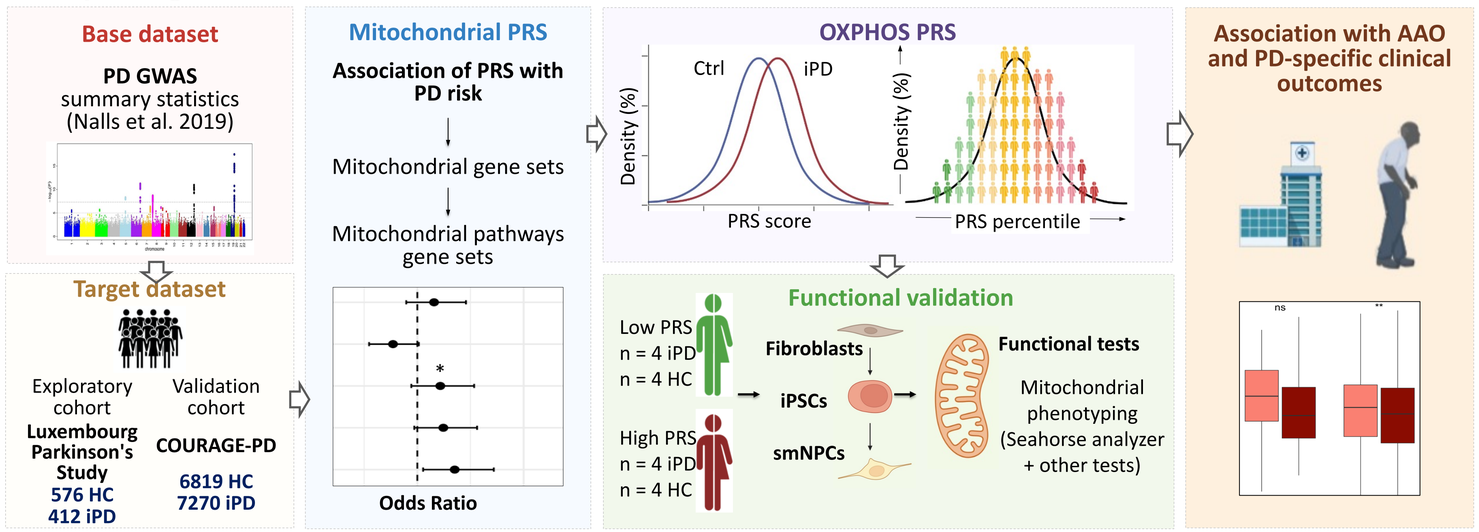
PARKINSON National Centre of Excellence in Research

Project Summary

Background: A large body of evidence specifically points to mitochondrial dysfunction as a major cause of Parkinson's disease (PD) pathogenesis. Given that only ~10% of PD cases can be attributed to monogenic causes, we hypothesize that a fraction of idiopathic PD (iPD) cases may harbour a pathogenic combination of common variants in mitochondrial genes ultimately resulting in mitochondrial dysfunction.

Objectives: To gain essential knowledge on the contribution of genetic variability in nuclear-encoded mitochondrial genes to iPD pathogenesis. Starting from genomic data, we aim to functionally validate mitochondrial polygenic risk profiles in patient-based cellular models, thus defining mitochondrial pathways potentially involved in neurodegeneration in subgroups of iPD patients.

Study design and workflow:



1. Association of common variants in mitochondrial genes with PD risk

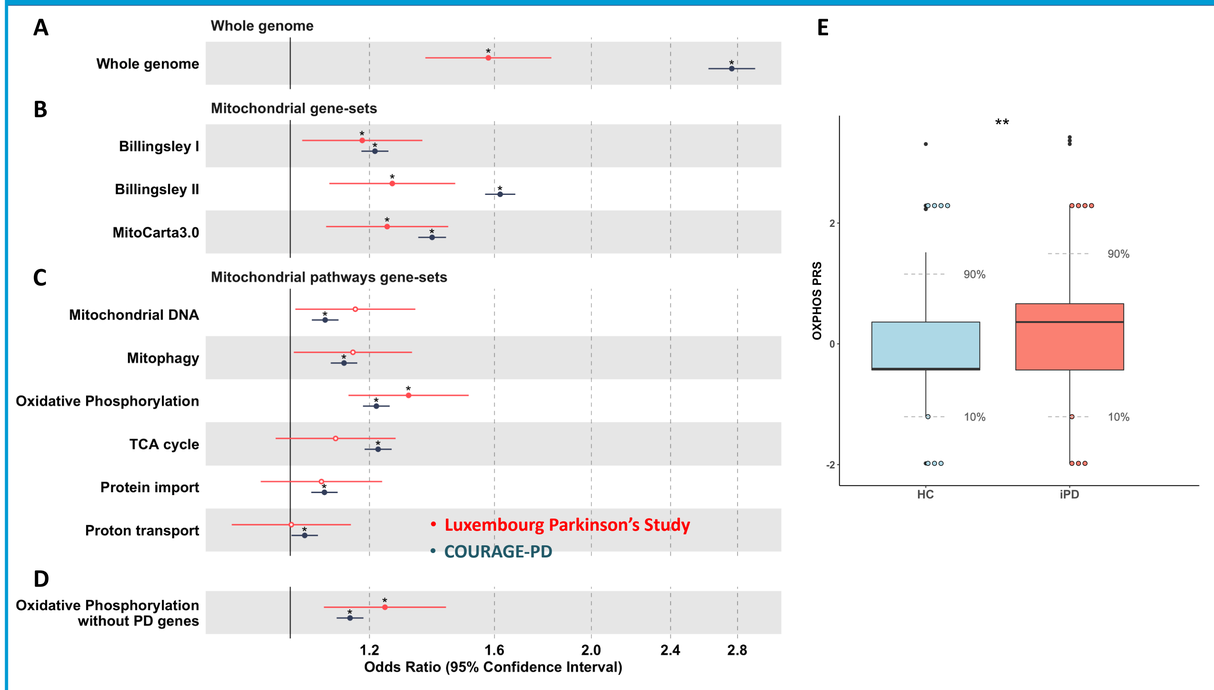


Figure 1. Common variants in mitochondrial genes are associated with higher PD risk in the Luxembourg Parkinson's Study (LuxPark) and COURAGE-PD cohorts. Forest plots of the odds ratio (OR) and 95% confidence interval for the whole genome (A), three different mitochondrial gene-sets (B), and six different mitochondrial pathways (C) polygenic risk scores (PRS) regressed with PD diagnosis for LuxPark and COURAGE-PD. OR was also plotted for the Oxidative Phosphorylation pathway (OXPHOS) without *PINK1*, *SNCA* and *DJ-1* genes (D). * FDR-adjusted p-values <0.05. (E) Distribution of standardized OXPHOS PRS by cases (iPD) and controls (HC) in LuxPark. Colored dots are showing the PRS of HC and PD in the highest and lowest OXPHOS-PRS range (10th and 90th percentile) and whose primary skin fibroblasts were available for functional studies.

2. Mitochondrial oxygen consumption is significantly elevated in primary skin fibroblasts from iPD patients with high *Oxidative Phosphorylation*-PRS

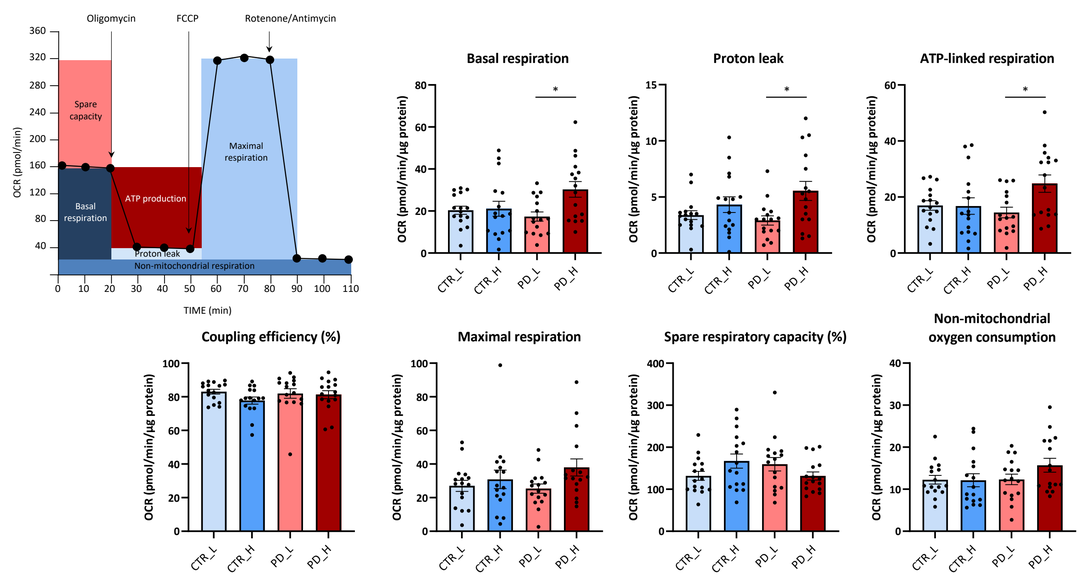


Fig. 2: Analysis of mitochondrial respiration in primary skin fibroblasts from iPD patients and HC stratified based on *OXPHOS*-PRS. Oxygen consumption rates (OCRs) were measured under basal conditions and after targeted inhibition of specific respiratory chain complexes by using a standard Seahorse Mito Stress test. Histobars represent the pooled means of four independent experiments performed in four distinct fibroblast lines established from Luxembourg Parkinson's Study participants (healthy controls vs iPD patients) with high (CTR_H ; PD_H) or low (CTR_L ; PD_L) *OXPHOS*-PRS. * p<0.05.

3. Other mitochondrial readouts are not altered in fibroblasts from iPD patients stratified based on *OXPHOS*-PRS

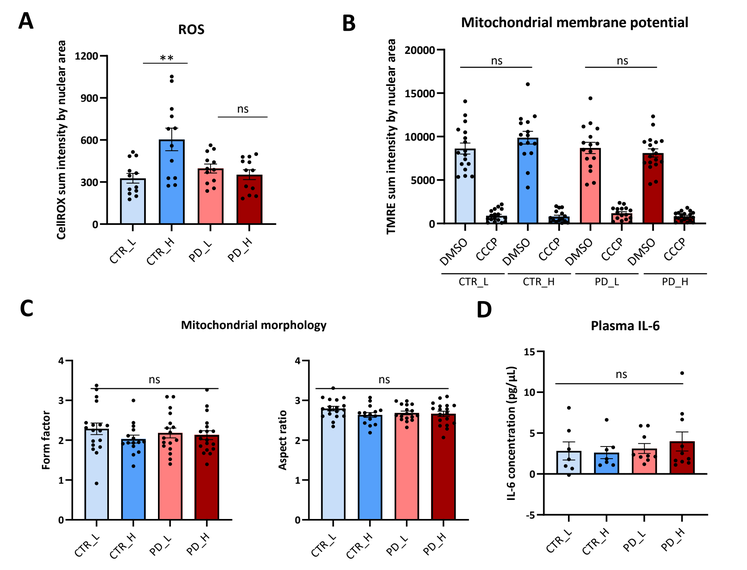


Figure 3: Assessment of additional readouts of mitochondrial activity in primary skin fibroblasts and plasma samples from iPD patients and HC stratified based on *OXPHOS*-PRSs. (**A-C**) High-throughput confocal microscopy analyses of primary skin fibroblasts from HC and iPD patients with high (CTR_H ; PD_H) or low (CTR_L ; PD_L) *OXPHOS*-PRSs. (**A**) Analysis of ROS levels by using the CellRox Deep Red dye. (**B**) Assessment of mitochondrial membrane potential by means of TMRE staining. (**C**) Analysis of mitochondrial morphology after Mitotracker Green staining. (**D**) Measurement of IL-6 levels in plasma samples obtained from HC and iPD *OXPHOS*-PRS. *p<0.05, **p<0.01, ns=nonsignificant.

4. Functional validation of *OXPHOS*-PRS in iPSC-derived neurons

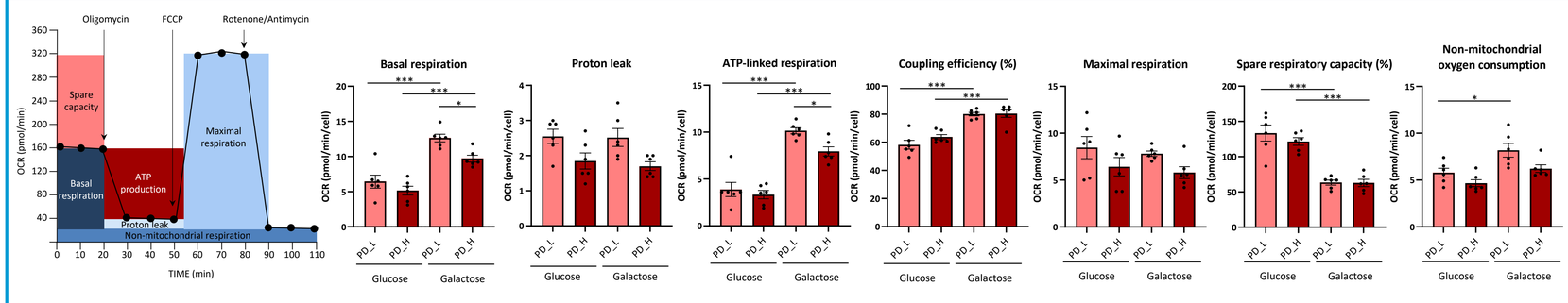


Figure 4: Assessment of mitochondrial respiration in iPSC-derived neuronal progenitor cells (NPCs) from iPD patients stratified based on *OXPHOS-PRSs*. Oxygen consumption rates (OCRs) were measured under basal conditions and after targeted inhibition of specific respiratory chain complexes by using a standard Seahorse mitochondrial stress test. Histobars represent the mean of 3 Independent experiments with 2 distinct iPSC-derived NPCs lines established from iPD patients with high (PD_H) or low (PD_L) *OXPHOS*-PRSs, cultivated in glucose or galactose medium. * p<0.05, *** p<0.001

5. iPD patients with high *OXPHOS*-PRSs have an earlier age at PD onset

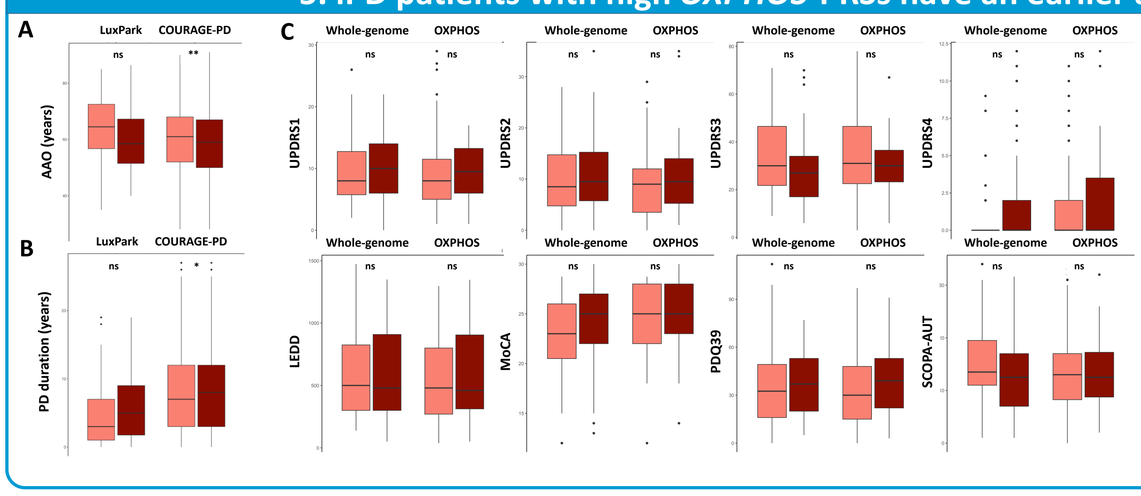


Fig. 5: Association of *OXPHOS*-PRSs with PD-specific clinical outcomes. Comparison of age at onset (AAO) (**A**) and disease duration (**B**) in iPD patients from the LuxPark and COURAGE-PD cohorts with high or low *OXPHOS*-PRS. * p < 0.05, ** p < 0.01, ns = not significant. (**C**) The same analysis was extended to eight PD-specific motor and non-motor clinical scores available only in LuxPark. The whole-genome PRS group was used here as reference.

Conclusion: We developed and functionally validated novel mitochondria-specific PRSs that could be used as a genetic tool to stratify the heterogeneous group of iPD patients. Using patient-based models relying on specific mitochondrial signatures for drug screening approaches may pave the way for future more tailored therapeutic strategies.