PARKINSON'S PROGRESSION MARKERS INITIATIVE

Play a Part in Parkinson's Research

IGF-1 and DAT imaging as biomarkers of early cognitive impairment in Parkinson's disease

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Summary

Please describe the analysis performed (Should not exceed 200 words)

Baseline serum samples from 405 PD patients and 191 healthy controls enrolled in the PPMI study were analysed for IGF-1 levels in order to determine if variations of such biomarker affect cognitive performances at baseline.
Baseline DAT imaging data from the same subjects have been also analysed to determine if variations of DAT uptakes are related to cognitive performances. In particular, DAT uptake values for the right and left caudate and putamen, for mean caudate and mean putamen, and for average striatum have been included in the analysis.
The following cognitive scores were evaluated: MoCA, Hopkins Verbal Learning Test-Revised (HVLT-R), Benton Judgment of Line Orientation (JOLO); Symbol-Digit Modalities Test (SDM); Letter-Number Sequencing (LNS) and semantic fluency (SF). With regard to HVLT-R, Immediate Recall (IR), Delayed Recall (DLRY), Retention and Discrimination recognition (DR) were analysed.

Methods

Describe the methods used. Sufficient information should be provided to enable investigators to understand the methodology and interpret the data

We performed the IGF1 measurement using the Quantikine Human IGF1 Immunoassay that is a solid-phase ELISA designed to measure human IGF1 in serum and plasma. Serum has been pretreated to release IGF1 from IGF-binding proteins with acid-ethanol extraction. This assay employs the quantitative sandwich enzyme immunoassay technique. A monoclonal antibody specific for IGF1 has been pre-coated onto a microplate. Standards and pretreated samples have been pipetted into the wells and any IGF1 present was bound by the immobilized antibody. After washing away any unbound substances, an enzyme-linked polyclonal antibody specific for IGF1 was added to the wells. Following a wash to remove any unbound antibody enzyme reagent, a substrate solution was added to the wells and color developed in proportion to the amount of IGF1 bound in the initial step. The color development was stopped and the intensity of the color was measured determining the optical density with a microplate reader (Victor X4- Perkin Elmer) set to 450 nm.

A standard curve has been generated for each set of samples assayed. Three samples of known concentration were tested twenty times on one plate to assess intra-assay precision (CV %: 3.5, 4.3, 4.3) and forty separate assays to assess inter-assay precision (CV%: 8.1, 8.3, 7.5).
This assay recognizes natural and recombinant human IGF1. Indeed, this immunoassay is calibrated against a highly purified *E. coli*-expressed recombinant human IGF1 produced at R&D Systems and has been shown to accurately quantitate the recombinant factor. Results obtained using natural human IGF1 showed linear curves that were parallel to the standard curves obtained using the Quantikine kit standards. These results indicate that this kit can be used to determine relative mass values for naturally occurring IGF1. The NIBSC/WHO IGF1 International Reference Reagent 02/254 was evaluated in this kit. The dose response curve of the International Reference Reagent parallels the Quantikine standard curve. To convert sample values obtained with the Quantikine Human IGF1 kit to NIBSC/WHO 02/254 values, we used the following equation: NIBSC/WHO (02/254) value (ng/mL) = 1.54 x Quantikine IGF1 value (ng/mL).
After Kolmogorov-Smirnov testing for normal distribution, parametric testing was performed for group comparisons (independent sample t-test). Descriptive statistics are given as means and standard deviation (SD) with range. Differences in the distribution of categorical variables among groups were assessed by the chi-square test. Linear regression analyses evaluating the association between serum IGF-1 levels and cognitive tasks scores were performed both in HC and PD. The lowest quartile compared to the other quartiles was added as a covariate. The effects of DATScan uptakes (right and left caudate and putamen and mean striatal uptake) on cognitive scores were examined by means of stepwise multiple linear regression analyses both in HC and PD. All the analyses included age, education and gender as covariates with no interaction terms. In the PD cohort, MCI was defined at two levels: (1) with MoCA score <26 ; (2) using psychometric tests, MCI categorization was reached through a cognitive test-based classification, requiring impairment (>1.5 standard deviations below the standardized mean score) on any *two* cognitive test scores (using immediate recall and recognition recall from the HVLT-R and single scores from each of the other tests). Logistic regression analyses was used to evaluate if IGF 1 lowest quartile was able to predict the diagnosis of MCI1 or MCI2 in PD cohort, considering age, gender and education as covariates. Logistic regression analyses (forward conditional method) was used to evaluate if DATScan uptakes were able to predict the diagnosis of MCI1 or MCI2 in PD cohort, considering age, gender and education as covariates. Multiple regression analyses were run to assess relationship between cognitive performances and DAT binding in MCI1 versus non MCI1 PD patients.
Results were considered statistically significant at $P<.05$.

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