Clinical Analysis

P-1: This patient showed no family history of neurological or psychiatric disease. The first signs of the disease appeared with bradykinesia in the left arm. Subsequently, the subject developed a classic phenotype of PD (resting tremor, rigidity, bradykinesia and good response to-L-Dopa treatment). The clinical diagnosis of PD was made by movement disorder specialists who used UK brain bank criteria for PD.

P-2: This subject previously of good health, was included in the study as a control. The subject had no family history for Parkinson's disease, (PD). A few years later the subject was diagnosed with atrial fibrillation and suffered a large cerebral hematoma from which this patient died a week later. Prior to stroke the patient had no signs of a movement disorder and mental functions were intact

P-3: This patient was diagnosed with PD. The subject had no family history for PD but tested positive for the LRRK2 G2019S mutation. The subject responded fairly well to levodopa. Later- on the subject gradually developed dementia of the PDD type. In addition to that the subject had mild visual hallucinations, but no severe psychiatric symptoms.

P- 4. The first symptoms of this subject were right sided bradykinesia and tremor. The patient had no family history for PD, and responded well to levodopa. Later-on, the subject showed gradual cognitive decline and developed severe psychiatric symptoms. The situation deteriorated and eventually requiring nursing home placement with akinetic mutism and with feeding through a percutaneous endoscopic gastroscopy, (PEG), tube.

P-5: This patient has no family history for PD. The patient developed axial bradykinesia and left-sided rest tremor, and responded well to levodopa. The patient developed severe psychiatric symptoms and was admitted to a psychiatric hospital. Since that time PD has been fluctuating with on and off periods. The patient needed support from home healthcare services and mental status has been unstable with periods of aggression. In parallel the patient has also developed renal failure needing dialysis 3 times a week.

P-6: This patient was diagnosed with Parkinson disease. The clinical symptoms included gait dysfunction in the absence of tremor (typical postural instability gait disorder phenotype). The patient responded well to levodopa. The patient reported multiple affected members with PD.

P-7: This patient was diagnosed with PD after approximately one year of muscular skeletal symptoms (rigidity) and tremor. The patient responded well to levodopa. The patient reported family history of PD.

P-8: This patient developed left hand kinetic tremor. The initial symptoms were mild without impairment of daily living functions. At later stages the patient exhibited bilateral rest tremor in both lower extremities symmetrically. Five years later the patient was diagnosed with PD. At that time patient had postural tremor in both upper extremities. However, the rest tremor in both hands followed later. Other stigmata of parkinsonism including rigidity and bradykinesia were also present. The patient responded positively to carbidopa/levodopa therapy. With progression of illness and disability, the bilateral subthalamic nucleus deep brain stimulation surgery was performed. The patient died at age 67 years. Autopsy was not performed. There was a positive family history of PD.

P-9: This patient was evaluated and diagnosed with Parkinson's disease. Onset had been 1.5 years before, initially manifesting as left lower limb rest tremor. The patient reported positive family history. The neurologic exam was remarkable only for left hemiparkinsonism and the MRI brain scan was essentially normal.

P-10: Parkinsonism onset was first noted with tremor in the left lower limb and "dragging" of that leg. Symptoms progressive and patient were diagnosed with Parkinson's disease the same year; a brain MRI scan was unremarkable. She proved levodopa responsive, but complicated by a short-duration response (wearing-off), plus levodopa-dyskinesia. The patient reported positive family history.

P-11: PD symptoms first developed insidiously consisting of slowness and clumsiness of the left upper limb. The patient was diagnosed with Parkinson's disease by a movement specialist and treated with carbidopa/levodopa with control of symptoms. Depression was first noted around that same time. Subsequently patient developed motor fluctuations and dyskinesias, further treated with adjunctive drugs. Later-on patient developed dream enactment behavior. About 15 years after parkinsonism onset, mild cognitive impairment was noted. The patient reported positive family history.

P-12: This patient developed the classical signs of PD and was followed at the Movement Disorders clinic when the subject was in early 70s. At that time in addition to PD, patient also exhibited signs of cerebellar ataxia, disturbance of gait and balance. The contact with the patient was lost and recent attempt to re-establish it failed.

P-13: This subject was enrolled in ongoing genetic study of PD as a control subject. The subject had no family history of PD. The neurological examination including motor and cognitive functions was entirely normal. The blood specimen was collected approximately 5 years ago and re-examination of this subject to confirm unaffected status for this study has not been possible.

P-14: This patient developed right hand tremor and was followed by asymmetrical bradykinesia and rigidity. The patient benefited from therapy with levodopa/carbidopa and after 12 years of treatment developed motor fluctuations and dyskinesias. He retired 16 years after the onset of his symptoms. One year after his retirement, the patient demonstrated classical features of PD with H&Y stage in “ON” of 3, and in “OFF” of 4, and UPDRS total score in “ON” of 61 and in “OFF” of 70. His MMSE is 30/30 but on this treatment patient reports some autonomic dysfunction (orthostatic hypotension).

P-15: This patient presented with truncal rigidity, shuffling gait and resting tremor affecting the left leg. She has no family history of Parkinson’s disease (PD). Physical examination showed a typical mask face, generalized bradykinesia and mild rigidity, with left side more severe than right side, and a mild resting tremor of the left leg. The subject had no postural instability. This patient presented with features compatible with idiopathic PD and symptoms were responsive to anti-PD medications.

P-16: The neurological examination is normal. The subject did not have a family history of parkinsonism or dementia.

P-17: This patient developed progressive motor disturbances. The patient developed progressive decrease in speech output, dysarthria, bradykinesia, bradyphrenia and

progressive gait disturbance. Initiation difficulty was prominent with shuffling and festinant gait. The patient subsequently developed slurred monotonous speech with drooling. Autonomic dysfunction with urinary frequency, retention sense and orthostatic hypotension was present. The MMSE score was 21/30. Resting tremor was absent but postural tremor was present in both hands worse on the left side. Cogwheel rigidity was worse on the left side. Posture was stooped. The patient walked in short steps and festination. Postural instability was severe. Arm swing was decreased more on the left. Symptoms were responsive to levodopa treatment. The patient was lost on follow-up and died at age 84. The patient did not have a family history of parkinsonism or dementia.

P-18: This patient developed the classical signs of PD, including mild cognitive impairment. The patient had a positive family history for PD.

Supplementary Table 1: Assessment of non-synonymous variants of VPS35 gene in PD. Analysis is performed irrespective of ethnicity. Two variants, p.Arg524Trp and p.Ile241Met are monomorhic and hence not shown in the table.

Gene	Non-synonymous variant	Minor/Major allele	MAF in carriers	MAF in controls	Odds ratio (95%CI)	p-value
VPS35	p.Gly51Ser	T/C	0.0002	0.0001	1.30 (0.21-7.84)	0.629
VPS35	p.Leu774Met	T/G	0.0004	0.00008	5.35(0.65-43.56)	0.116

MAF: minor allele frequency, CI: confidence interval,

Supplementary Table 2: Overall analysis irrespective of ethnicity and influence of between-study heterogeneity

Gene	SNP	Q test p-value	I2(95%CI)	Odds ratio (95%CI) by random effects	Fixed effects p-value	Random effects p value
VPS35	rs4966616	0.26	17(0-53)	0.96 (0.88-1.05)	0.629	0.466
VPS35	rs33994299	0.65	0(0-46)	0.99(0.93-1.05)	0.878	0.878

CI: confidence interval