Supplementary Online Content


eAppendix 1. Full details of the antibodies used in the study including dilution and manufacturer

eAppendix 2. Clinical summaries

eReferences

This supplementary material has been provided by the authors to give readers additional information about their work.
eAppendix 1: Full details of the antibodies used in the study including dilution and manufacturer

Glial fibrillary acidic protein (GFAP 1:1000, Dako, Ely, UK), α-synuclein (1:50, Vector, Peterborough, UK), tau (1:600, AT8 clone, Autogen Bioclear, Wiltshire, UK), Aβ (1:100, Dako, Ely, UK), ubiquitin (1:200, Dako, Ely, UK), p62 (1:200 BD Transduction Labs, Oxford, UK), neurofilaments (NFC 1:20, MP Biochemicals, Ohio, USA), IC2 (IC2 1:1000, Merck Millipore, Billerica, MA, USA), fused in sarcoma (FUS 1:200 NB100-565, Novus biological, Cambridge, UK) and TDP-43 (1:800, Abnova, Taipei city, Taiwan).

Diffuse and mature cortical Aβ plaques were graded using a method based on the CERAD (Consortium to Establish a Registry for Alzheimer's disease) criteria1 as described previously2. Cerebral amyloid angiopathy was assessed using a published four point scale3. Alzheimer tau pathology was analysed to provide the Braak & Braak stage4. Small vessel disease was graded as previously described5.

eReferences


eAppendix 2: Clinical Summaries

Parkin case 2

At age 25 years, patient 2 developed a shake and dystonic posturing in her left hand. She was diagnosed with young-onset Parkinson’s disease (YOPD) aged 28 and commenced on levodopa therapy to which she was very responsive. Dyskinesias appeared shortly after initiation of levodopa. Aged 42 years she developed freezing of gait and refractory motor fluctuations, and started subcutaneous, waking day, apomorphine pump therapy. This led to a marked reduction in the severity of her dyskinesias and a smoother motor response; although an ‘off’ period foot dystonia persisted and required regular treatment with botulinum toxin therapy. Aged 60 years she had a flexed posture, an unsteady short stepping gait, moderate bradykinesia, mild action and rest tremor and dystonia of her right ankle. Severe abdominal wall panniculitis after 18 years of apomorphine led to a switch to enteral levodopa therapy (Duodopa). Formal neuropsychology showed evidence of mild executive dysfunction and psychomotor slowing but no dementia. She scored 62 in the UPDRS III when ‘off’ improving to 33 one hour following a levodopa challenge (250mg co-beneldopa dispersible); her PDQ-39 was scored at 80/156. Enteral Levodopa therapy improved her symptoms but she died 3 months later of unknown cause. She was the eldest of 6 siblings and had one brother diagnosed with PD aged 58 years.

Parkin case 3

This woman had onset of left arm and leg tremor aged 33 years and was initially treated with a beta blocker for suspected essential tremor. Aged 36 years, she reported dragging of her left leg and increased shaking of her left hand and the diagnosis was revised to PD and she was commenced on treatment with levodopa (co-careldopa 100mg 6-8/day) to good effect. Aged 42 years her tremor re-emerged and she was weaned off levodopa due to some uncertainty relating to the diagnosis, but became ‘a complete cripple, unable to move or do anything’ and was wheelchair bound for 12 months until the levodopa was restarted. Aged 54 years she described freezing of gait, restless legs and an excellent response to levodopa, despite prominent chorea of her legs. Examination when ‘off’ revealed a masked face, global akinesia, axial and limb rigidity (left>right), impaired rapid alternating movements of the left hand, difficulty arising from a chair, stooped posture, retropulsion and freezing of gait. At age 59 years her parkinsonism was complicated by painful dystonic spasms in her thighs which were controlled by an apomorphine
infusion. She denied hallucinations and had no cognitive impairment. She died aged 61 years of pneumonia. Her brother presented with abnormal posturing of his foot at age 15 years and was subsequently diagnosed with young onset parkinsonism.

**Parkin case 4**

Patient 4 was diagnosed with parkinsonism at the age of 32, but reported symptoms of resting tremor and difficulty walking from his 20’s. He managed to work as a carpenter until retiring at age 60 years. He responded well to low dose co-careldopa and his main complaints in his later years were of anxiety, motor fluctuations and dyskinesias. Aged 68 years he was admitted to hospital for management of his severe motor fluctuations, dyskinesias and off-period dystonia. There was no cognitive impairment and he was fully independent for all activities including driving. Unfortunately he developed fulminant aspiration pneumonitis and died during the admission. Two of his seven siblings had parkinsonism: a brother (died in his 60’s) and a sister (now aged 84 years) whose parkinsonian symptoms began in her 20’s, who recently tested positive for mutations of *parkin* (G430D; Pro113fs).

**Parkin case 5**

Patient 5 has been reported in a study of patients with benign tremulous parkinsonism (Selikhova et al, under review). He was in his mid-40’s when he developed a resting tremor of the left foot, which quickly spread to involve both his legs, becoming so severe that he had to have his car converted to an automatic gear box. At age 55 years he developed tremor of his left hand, although the bilateral leg tremor remained more prominent and troublesome (video 1). He was diagnosed with benign tremulous parkinsonism and treated with co-careldopa 100mg, 6/day which made him ‘100% better’. After 1 year he experienced ‘wearing off’ in between doses. On examination it was noted when ‘on’ he was normal, but when ‘off’ he had a marked rest tremor in his legs and decreased arm swing. At age 60 years he was experiencing abrupt motor fluctuations, with ‘off’ pain in both his legs and in his chest. Examination when ‘on’ revealed no tremor or rigidity, good arm swing, mild bradykinesia, and chorea of both feet (video 2). He remained on the same dose of levodopa (600mg/day) for 25 years with a small dose of cabergoline (4mg) added in his 70’s. In his last decade he experienced falls, freezing of gait and emotional lability and was treated with antidepressants for low mood. At age 81 years he was described as ‘cognitively extremely good’. He died aged 82 years of a myocardial infarction one day following knee replacement surgery.