|  |  |  |  |  |  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- | --- |
| **No** | **Study** | **Country** | **Ethnicity** | **Disease group** | **No of cases** | **Repeat size in cases** | **No of controls** | **Repeat size in controls** | **Repeat sizing method** | **Cut-off for expansion** | **Remarks** |
| 1 | Akinomoto et al, 2013 | Sweden | Swedish | PD | 135 | Mean (sd) 5.3 ± 4.3(2–23) | 645  | Mean (sd) 5.9 ± 4.7 (2-31) | rp-PCR with fragment length analysis | Detection of saw-tooth pattern; intermediate unit defined as >20  | Intermediate alleles in 2.2% of PD, 0% of atypical PD, 2.9% in control group. Mean repeat length ± sd: MSA 4.0 ± 2.7 (range 2–10); PSP 7.1 ± 4.8 (range 2–17); unclassified 3.3 ± 2.3 (range 2–6). 3 PSP cases with >10 repeats. |
| 2 | Daoud et al, 2013 | Canada(Montreal, Quebec) | French-Canadian | PD | 285 | No range mentioned | 190  | No range mentioned | rp-PCR; Southern blot for 1 case with 24 repeats | Detection of saw-tooth pattern used | 1 PD patient with 24 alleles on Southern blotting aged 66 with no family history of PD or other neurodegenerative disease |
| 3 | DeJesus-Hernandez et al, 2013 | USA | Caucasian | PD | 676 | 2-23 | 1356 | 2-23 | Fluorescent-labelled PCR with capillary electrophoresis | Not specified, presumed >30 | No association between repeat length and risk of PD, ET or RLS after correction for multiple testing (P<0.0028). No significant association with repeat length and age-at-onset. |
| 4 | Harms et al, 2013 | USA | Caucasian | PD | 478 | Mean (sd) 7.2 ± 4.5;all alleles had ≤27 repeats | 662 | Mean (sd) 7.6 ± 4.5;3 controls had ≥50 units | rp-PCR | >30 | 3 control subjects with ≥50 repeats were normal and non-demented at last contact (ages 50, 67, and 71 years), but 2 had a first- or second-degree relative with ALS. Length of longest repeat allele did not correlate with age at onset (p=0.11) in PD. |
| 5 | Lesage et al, 2013 | France | French | PD | 1225 | <26 | 445 | Mean (sd) 4.5 ± 3.3 (2–22) | rp-PCR and fragment length analysis;  | ≥60 | Southern blot performed in carriers and intermediate alleles. |
| 6 | Lin et al, 2014 | Taiwan | Han Chinese  | PD, parkinsonism syndromes; AD and FTD | 482 (including 17 with PDD) | 2-25  | 485 | 2-25 | 2-step rp-PCR  | Pathogenic >30, intermediate 20-29 repeats | Parkinsonism syndrome (n=95), familial PD (n=109), young-onset PD (n=201), FTD (n=9), sporadic AD (n=61), and early-onset AD (n=7).One young-onset typical PD case had intermediate range of 25 repeats, and 1 control with 21 repeats.  |
| 7 | Majounie et al, 2012 | USA | Caucasian, non-Hispanic | PD | 781  | Average 3, (0-38) | None | - | rp-PCR | >30 | 4 classical PD cases had intermediate alleles (21, 23, 24, and 38 repeats); no correlation between repeat length and family history or age of disease onset. |
| 8 | Nuytemans et al, 2013  | USA | Non-Hispanic Caucasians | PD | 396 + 481 (replication dataset) | 13 PD cases had >20 repeats | 1144 | <23 | rp-PCR and fragment length analysis | Intermediate: >20–30+ repeats; large >30 repeats | Overall, 14 cases (13 PD, 1 ETP) and 3 controls had >20 repeats (p=0.002). 7 cases and no controls had >23 repeats (p=0.003). C9ORF72 intermediate repeats may contribute to risk for PD and essential tremor with parkinsonism (ETP). |
| 9 | Nuytemans et al, 2014 | USA | Non-Hispanic Caucasians | Path-confirmed PD | 488  | ≤4-19; No intermediate carriers  | Samples with >20 repeats from previous dataset used as controls | ≥20  | rp-PCR with fragment length analysis | ‘larger repeats’ defined as >20 | Cut-off of >20 chosen due to most commonly reported lower limit of “intermediate” C9ORF72 repeats. No larger (intermediate or expanded) repeats found in autopsy-confirmed PD samples.  |
| 10 | Theuns 2014 et al, (GEO-PD Consortium) | Multicentre | Europe, Asia, North America, Australia | PD | 7494 | 0-17;Only 0.7% of PD cases had ≥17 repeats | 5886 | 0-17; only 0.5% of controls had ≥17 repeats | 2-step procedure: STR fragment length assay with flanking PCR primers followed by forward and reverse rp-PCR | >60 | Size of this global cohort used to estimate a PD-related threshold – suggesting role for 10 repeats and for pooled alleles of ≥17 repeats in PD susceptibility; but did not reach significance after correction for multiple comparisons.Repeat range for controls: Caucasian 0-32; Asian 7-14  |
| 11 | Xi et al, 2012 | UK, Italy, Spain, North America | European and North American | PD(Including 29 LRRK2 G2019S carriers  | 289 | 2-30 | 602 | 2-30 | 2-step PCR – fragment length analysis followed by rp-PCR | >30 | No intermediate or pathological number of repeats for the second allele (2-11 repeats) detected in expansion carriers. Intermediate alleles seen in 4 PD cases (2 with severe dementia). None of the 29 LRRK2 mutation carriers had an expanded or intermediate allele (<11 repeats).  |
| 12 | Yeh 2013 | Taiwan | Han Chinese | PDD | 71 | Mean (sd) 4.2 ± 3.15 (1-19) | 100 | Mean (sd) 4.23 ± 3.08 (1-17) | Fluorescent rp-PCR | Not specified |  |

Supplementary Table 3. PD studies included. PD = Parkinson’s disease; rp=PCR = repeat-primed polymerase chain reaction; MSA = multiple system atrophy; PSP = progressive supranuclear palsy; ET = essential tremor; RLS = restless legs syndrome; ALS = amyotrophic lateral sclerosis; PDD = Parkinson’s disease with dementia; AD = Alzheimer’s disease; ETP = essential tremor with parkinsonism; STR = short tandem repeat; LRRK2 = Leucine-rich repeat kinase 2.