




BRAIN COMMUNICATIONS

Intellectual enrichment and genetic modifiers of cognition and brain volume in Huntington's disease

 Marina Papoutsis,^{1,2}  Michael Flower,¹ Davina J. Hensman Moss,¹ Peter Holmans,³ Carlos Estevez-Fraga,¹ Eileanoir B. Johnson,¹ Rachael I. Scahill,¹  Geraint Rees,^{4,5} Douglas Langbehn,⁶ and Sarah J. Tabrizi^{1,7} the Track-HD Investigators

See Hannan (<https://doi.org/10.1093/braincomms/fcac308>) for a scientific commentary on this article.

An important step towards the development of treatments for cognitive impairment in ageing and neurodegenerative diseases is to identify genetic and environmental modifiers of cognitive function and understand the mechanism by which they exert an effect. In Huntington's disease, the most common autosomal dominant dementia, a small number of studies have identified intellectual enrichment, i.e. a cognitively stimulating lifestyle and genetic polymorphisms as potential modifiers of cognitive function. The aim of our study was to further investigate the relationship and interaction between genetic factors and intellectual enrichment on cognitive function and brain atrophy in Huntington's disease. For this purpose, we analysed data from Track-HD, a multi-centre longitudinal study in Huntington's disease gene carriers and focused on the role of intellectual enrichment (estimated at baseline) and the genes *FAN1*, *MSH3*, *BDNF*, *COMT* and *MAPT* in predicting cognitive decline and brain atrophy. We found that carrying the 3a allele in the *MSH3* gene had a positive effect on global cognitive function and brain atrophy in multiple cortical regions, such that 3a allele carriers had a slower rate of cognitive decline and atrophy compared with non-carriers, in agreement with its role in somatic instability. No other genetic predictor had a significant effect on cognitive function and the effect of *MSH3* was independent of intellectual enrichment. Intellectual enrichment also had a positive effect on cognitive function; participants with higher intellectual enrichment, i.e. those who were better educated, had higher verbal intelligence and performed an occupation that was intellectually engaging, had better cognitive function overall, in agreement with previous studies in Huntington's disease and other dementias. We also found that intellectual enrichment interacted with the *BDNF* gene, such that the positive effect of intellectual enrichment was greater in Met66 allele carriers than non-carriers. A similar relationship was also identified for changes in whole brain and caudate volume; the positive effect of intellectual enrichment was greater for Met66 allele carriers, rather than for non-carriers. In summary, our study provides additional evidence for the beneficial role of intellectual enrichment and carrying the 3a allele in *MSH3* in cognitive function in Huntington's disease and their effect on brain structure.

- 1 UCL Huntington's Disease Centre, Queen Square Institute of Neurology, University College London, London, UK
- 2 Ixico plc, London, UK
- 3 MRC Centre for Neuropsychiatric Genetics and Genomics, Cardiff University, Cardiff, UK
- 4 Wellcome Centre for Human Neuroimaging, Queen Square Institute of Neurology, University College London, London, UK
- 5 Institute of Cognitive Neuroscience, University College London, London, UK
- 6 Carver College of Medicine, University of Iowa, Iowa City, IA, USA
- 7 UK Dementia Research Institute at University College London, London, UK

Correspondence to: Dr Marina Papoutsis
UCL Huntington's Disease Centre, Russell Square House
10-12 Russell Square, London, WC1B 5EH, UK
E-mails: m.papoutsis@ucl.ac.uk; marina.cogsci@gmail.com

Received April 04, 2022. Revised August 22, 2022. Accepted October 27, 2022. Advance access publication October 31, 2022

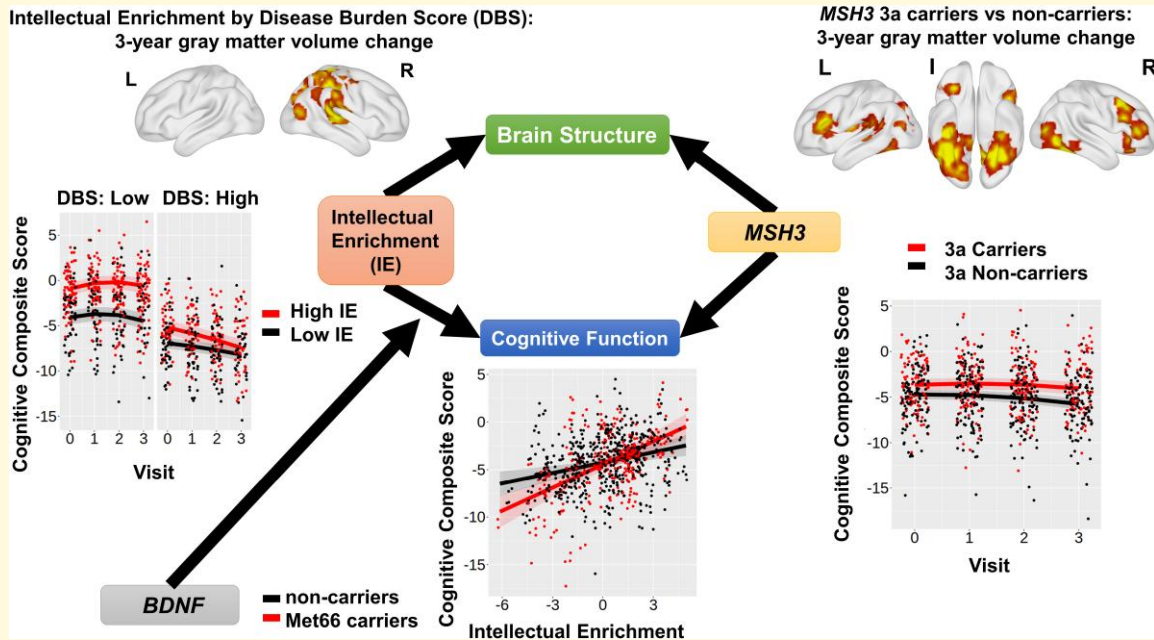
© The Author(s) 2022. Published by Oxford University Press on behalf of the Guarantors of Brain.

This is an Open Access article distributed under the terms of the Creative Commons Attribution License (<https://creativecommons.org/licenses/by/4.0/>), which permits unrestricted reuse, distribution, and reproduction in any medium, provided the original work is properly cited.

Keywords: Huntington's disease; brain-derived neurotrophic factor; intellectual enrichment; MSH3; cognitive modifiers

Abbreviations: BDNF = brain-derived neurotrophic factor; CAG = coronary artery angiography; CI = confidence interval; COMT = catechol-O-methyl transferase; DBS = disease burden score; DF = degrees of freedom; FAN1 = fancd2- and fanci-associated nuclease 1; GM = grey matter; HTT = Huntingtin; IQ = intelligence quotient; MAPT = microtubule-associated protein tau; MIDAS = medical image display and analysis software; MSH3 = MutS homologue 3; SD = standard deviation; SE = standard error; SEM = structural equation modelling; SNP = single-nucleotide polymorphism; SPM = statistical parametric mapping; TIV = total intracranial volume; UHDRS TMS = unified Huntington's disease rating scale total motor score; VBM = voxel-based morphometry

Graphical Abstract



Introduction

Huntington's disease is a genetic, neurodegenerative disorder caused by an abnormal coronary artery angiography (CAG) repeat expansion in the Huntingtin (*HTT*) gene. It is characterized by a triad of symptoms, motor, psychiatric and cognitive. All Huntington's disease gene carriers will eventually develop dementia,¹ but there is substantial variability in its onset and severity, which cannot be explained fully by CAG repeat length and age. Cognitive impairment is present in Huntington's disease gene carriers many years before predicted disease onset and in the absence of motor symptoms.² However, research on the genetic and environmental factors that contribute to this variability in cognitive impairment in Huntington's disease is still limited.

Individual differences in cognitive function and rate of decline have been extensively studied in ageing and Alzheimer's disease. One prominent hypothesis is that of brain maintenance,³ according to which the primary determinant of preserved cognitive function is lower levels of pathology and a slower rate of neurodegeneration. However, it has also been observed that individual differences in cognitive impairment exist despite similar levels of neurodegeneration, which led to the

theory of cognitive reserve.⁴ Although the genetic and environmental factors that support brain maintenance and cognitive reserve in ageing and dementia are not all known, lifelong participation in intellectual activities, also known as intellectual enrichment,⁵ as well as genetic polymorphisms,⁶ have been associated with preserved cognitive function and mechanisms of brain maintenance and cognitive reserve. Genetic factors and intellectual enrichment have also been shown to interact and enhance their effects on brain structure and cognition.⁷⁻⁹

In Huntington's disease, a small number of studies have so far examined the role of genetic polymorphisms and lifestyle factors on individual differences in cognitive function. More specifically, environmental enrichment,¹⁰ education and participation in lifelong intellectual activities,¹¹⁻¹³ as well as a number of genes, including fancd2- and fanci-associated nuclease 1 (*FAN1*),¹⁴ catechol-O-methyl transferase (*COMT*),¹⁵ MutS homologue 3 (*MSH3*)¹⁶ and microtubule-associated protein tau (*MAPT*),¹⁷ predict cognitive function. Two of these studies have also provided preliminary evidence that intellectual enrichment is associated with less striatal atrophy in humans,^{12,13} suggesting that it supports greater brain maintenance.

The aim of our work was to provide evidence regarding the effects of intellectual enrichment and genetic factors on cognitive function and brain structure in Huntington's disease. For this purpose, we retrospectively analysed data from a multi-centre, longitudinal study, Track-HD,^{18–21} that measured changes in behaviour and brain structure over 3 years in individuals with the Huntington's disease gene mutation in pre-manifest (maximum 15 years from predicted onset) and early stages of the disease. We quantified lifetime intellectual enrichment using level of education, pre-morbid intelligence quotient (IQ) and occupational cognitive demands¹² measured at baseline. In terms of genetic polymorphisms, we selected common polymorphisms that have been previously associated with cognitive function in Huntington's disease [*COMT*, brain-derived neurotrophic factor (*BDNF*), *FAN1*, *MSH3* and *MAPT*].

Materials and methods

Participants

Track-HD is a multi-centre, 4-year observational study in Huntington's disease gene carriers and matched controls. A full description of the Track-HD study has been previously reported.^{18–21} In summary, 243 Huntington's disease gene carriers (both manifest and pre-manifest) and 123 matched controls were recruited across four sites (London, UK; Paris, France; Leiden, The Netherlands and Vancouver, Canada). The participants were predominantly Caucasian (97.5%), which limits our ability to test differential effects across different populations. However, it is important to note that Huntington's disease is predominantly found in individuals with European ancestry.²² Local ethics committees approved the study at each site and all participants provided written informed consent according to the Declaration of Helsinki.

The Track-HD study included detailed measures of brain structure, cognitive and motor function, in addition to information regarding education, pre-morbid IQ and profession. Blood for DNA analysis was also collected. **Table 1** shows details of the measures that were used in this study and the number of participants included (split by visit for longitudinal measures). Data from all Huntington's disease gene carriers with at least one follow-up visit ($n = 229$), irrespective of disease diagnosis, were used for the analyses. Data from the matched control group were only used to create standardized scores of cognitive performance in the gene-carrier group (demographic information on the control group is provided in **Supplementary Table 1**).

Global cognitive function

To quantify cognitive function and change over 3 years, we created a composite score from the available cognitive measures. This composite score represents global cognitive function and is composed of the following measures from the Track-HD cognitive battery¹⁸: number correct in 90 s from

the symbol digit modality test (a measure of processing speed), number correct in 45 s from the stroop word reading test (a measure of psychomotor speed), number correct adjusted for guessing (>0 means better than chance) for the five items condition from the spot the change task (a measure of working memory), number correct for negative emotions from the emotion recognition task (a measure of facial emotion recognition) and variability in the inter-tap interval in a paced tapping task at 3 Hz (a measure of temporal precision). These tasks were included in all four visits, in addition to the circle tracing task. However, that task was excluded from the composite score because of large practice effects that persisted across all visits.²³ The remaining five measures were then used to create a composite score of global cognitive function.

To calculate the composite score, raw values were transformed to Z-scores using the mean and standard deviation (SD) of the control group at baseline and then summed. In the paced tapping task, the reciprocal of the variance in the inter-tap interval was used, such that a higher value indicated better performance (i.e. lower variability), consistent with all other measures. Therefore, higher values in the composite score indicate better performance. When computing the composite score, if one or two measures were missing, they were replaced by the mean Z-score of the existing measures. This was the case for 21 participants in Visit 1, 7 participants in Visit 2, 3 participants in Visit 3 and 16 participants in Visit 4, respectively. If a participant had less than three out of five measures available for a visit, then we did not compute a composite score for that visit and therefore, this participant visit was not included in the analyses with cognitive function as a variable.

Genetic polymorphisms

Track-HD subjects were genotyped using Illumina Omni2.5v1.1 arrays²⁴ and genotypes were extracted using PLINK software.²⁵ Because the main outcome measure was cognitive function, we focused on single-nucleotide polymorphisms (SNPs) that had been previously associated with cognitive and psychomotor function in Huntington's disease. Based on previous literature, we, therefore, selected the following SNPs: rs4680 on Chromosome 22¹⁵ in *COMT*, which relates to dopamine metabolism, rs9468 on Chromosome 17¹⁷ in *MAPT*, which relates to tau protein production, rs2140734 on Chromosome 15 near *FAN1*,^{14,26} which is involved in DNA repair and a polymorphic repeat expansion in Exon 1 on Chromosome 5 of *MSH3*,^{16,24,27,28} which is involved in DNA mismatch repair. In addition, we tested another polymorphism, rs6265 (Val66Met) on Chromosome 11 in *BDNF*, which encodes the Val66Met polymorphism and regulates BDNF expression. Although the role of the Val66Met polymorphism in Huntington's disease remains unknown, it has been consistently associated with cognitive function in ageing and dementia^{6,29–31} and *BDNF* expression may be affected by Huntington's disease pathology.^{32,33} It also interacts with

Table 1 Huntington's disease gene-carrier demographics and clinical characteristics

Variable	Missing data No (% all participants)			
	Baseline	Visit 2	Visit 3	Visit 4
Total number of participants				
Male, n (%)	229 104 (45%)			
Age at baseline, mean (SD)	44.9 (10.2)			
CAG, median (min–max)	43 (39–59)			
DBS at baseline, mean (SD)	333.2 (74.2)			
Clinical status at baseline, n (%)	Pre-manifest 115 (50%)	Motor manifest 114 (50%)		
Education Level, median (min–max)	4 (1, 6)			
Occupational cognitive demands, Mean (SD)	3.70 (0.87)		10 (4%)	
Verbal IQ, median (min–max)	UK: 33 (4–47), CA: 34 (11–48) FR: 34 (19–43), NL: 39 (16–48)		0	
Composite cognitive score, mean (SD) per visit	–3.85 (3.89)	–4.10 (4.52)	–4.23 (4.57)	0
Total GM volume Mean (SD) per visit	0.052 (3.9)	–0.17 (3.9)	–0.30 (4.1)	2 (1%)
Caudate Volume Mean (SD) per visit	–0.0012 (0.09)	–0.0070 (0.09)	–0.0231 (0.09)	19 (8%)
UHDRS TMS at baseline median (min–max)	6 (0–52)			14 (6%)
Visit from baseline (years) mean (SD)	0	1.99 (0.08)	3.03 (0.10)	0
MSH3	Number of participants with 0 3a alleles: 126 1 3a alleles: 71 2 3a alleles: 18	3a allele non-carriers: 126 (55%) 3a allele carriers: 89 (39%)		2 (0.9%)
FANI	Number of participants with 0 G alleles: 89 1	G carriers (rs2140734) Non-carriers: 89 (39%)		16 (7%)
BDNF	G alleles: 107 2 G alleles: 17 Number of participants with 0 Met66 alleles: 136 1 Met66 alleles: 73 2 Met66 alleles: 4	Carriers: 124 (54%) Met66 Non-carriers: 136 (59%) Met66 Carriers: 77 (34%)		16 (7%)
COMT	Number of participants with 0 Met158 alleles: 57 1 Met158 alleles: 113 2 Met158 alleles: 43	Val158 carriers: 170 (74%) Met158 homozygotes: 43 (19%)		16 (7%)
MAPT	Number of participants with 0 H2 alleles: 120 1 H2 alleles: 78 2 H2 alleles: 15	H1 homozygotes: 120 (52%) H2 carriers: 93 (41%)		16 (7%)

intellectual enrichment factors to predict cognitive function^{7,34}: it was therefore relevant to our research question.

A previous study showed that the gene *TREM2* (rs75932628) has a role in cognitive function in Huntington's disease.³⁵ However, the minor allele frequency is very low in the population (0.005% in Europeans in 1000 genomes project data Phase 3) and therefore, we did not include it in our analyses.

All genetic predictors were coded as having a binary, dominant effect, similar to the approach of Vuono *et al.*¹⁷ **Supplementary Table 2** shows how each binary predictor was coded. In more detail, the *MSH3* predictor was coded for the presence of the three-repeat allele (3a).²⁷ The rs2140734 (*FAN1*) predictor was coded for the presence of the minor allele G. It is also important to highlight that this SNP is in complete linkage disequilibrium with minor allele C in rs3512. The latter has been more widely examined in other studies and shown to be associated with age of onset and disease progression in Huntington's disease.^{24,26,36} The *MAPT* predictor distinguished between H1 haplotype homozygotes and H2 carriers. Carrying the minor allele C in rs9468 tags for the H2 haplotype, whereas carrying the allele T tags for the H1 haplotype.³⁷ The *COMT* (rs4680) predictor variable distinguished Met158 homozygotes from carriers of the Val158 allele.¹⁵ The *BDNF* (rs6265) predictor variable distinguished carriers from non-carriers of the detrimental allele, Met66.

For completion, we also repeated the analyses coding the variables by the number of minor alleles for all genes except *BDNF*, because there was an insufficient number of cases (see **Table 1** for the number of participants per minor allele).

Intellectual enrichment

Education, bilingualism, leisure and professional intellectual activities are some of the activities associated with a protective effect against cognitive decline.³⁸ Track-HD recorded main profession, education level using the international standard classification of education scale and pre-morbid IQ using vocabulary tests. Because different tests were used in each country, pre-morbid IQ was standardized within the country (national adult reading test -2 in the UK, ANART in Canada, Dutch adult reading test in The Netherlands and Mill Hill in France). Occupational cognitive requirements³⁹ were estimated from the main profession recorded for each participant (see **Supplementary Methods**). These three measures were then standardized and summed up to create a composite score of intellectual enrichment.¹² A higher value in the intellectual enrichment score means a higher level of education, a more cognitively demanding profession and a higher level of estimated pre-morbid IQ.

Measures of disease pathology

Predicted disease severity at the time of recruitment was measured using the CAG by age product [disease burden score (DBS) = age × (CAG − 35.5)].⁴⁰ This is a commonly used

model of predicted exposure to disease pathology describing the well-established relationship between age and the CAG repeat number of the longer allele. The larger the CAG repeat length, the earlier the predicted age of disease onset.⁴¹

Pathology at baseline and rate of change were quantified using structural MRI measures of caudate volume and total grey matter (GM) volume, which are robust and well-defined markers of brain atrophy in Huntington's disease.^{18–21} Measures of white matter integrity using diffusion weighted imaging were only introduced at Visit 4 in Track-HD; therefore, we only focused on GM volume in our study. Whole-brain T₁-weighted 3D magnetization-prepared rapid acquisition with gradient echo images were acquired at 3T at all four visits (for details of the imaging protocol see Tabrizi *et al.*¹⁸). Caudate volume at baseline and longitudinal change was measured using medical image display and analysis software (MIDAS)' semi-automated segmentation and the boundary shift integral, respectively.^{20,42,43} Total GM volume at baseline was measured using statistical parametric mapping 12 (SPM12). Longitudinal change was measured using a non-linear fluid registration method in MIDAS, which produced whole-brain voxel compression maps measuring change from baseline.⁴⁴ Voxel compression maps were then convolved with SPM-derived GM maps to generate a change in total GM over time. The measures of caudate volume and total GM volume used in all the analyses were transformed to per cent of total intracranial volume (TIV), in order to adjust for differences in brain size. TIV was measured at baseline using MIDAS.

In addition to caudate and total GM volume, we also performed exploratory whole-brain analyses using voxel-based morphometry (VBM).⁴⁵ The GM probability maps at baseline and voxel compression maps of change from baseline were normalized to a group template space using diffeomorphic anatomical registration through exponentiated lie algebra. Normalized images were then smoothed using an 8 mm full-width at half maximum Gaussian kernel. Full details of the MRI methods used have been published previously.⁴⁶

Statistical analysis

Statistical analyses were performed using R version 3.6.3 (<http://www.r-project.org/>) and the packages *lmtest* (version 3.1-1) and *lme4* (version 1.1-21).

To examine the relationship between cognitive function and brain volume with our predictors of interest, we used linear mixed models. Our predictors of interest were intellectual enrichment and genetic polymorphism. All models were fitted using maximum likelihood estimation and correlated random intercept and slope. All models included as covariates age, DBS and study site and their interaction with the visit, as well as sex (main effect only, because the sex by visit interaction term did not improve model fit and was therefore dropped). Models with cognitive function as an outcome also included the use of antipsychotic medication as a covariate (see **Supplementary Methods**). Time was modelled in years of follow up as approximated by the

annual visit number. Based on previously published analyses of the Track-HD data,²³ we included quadratic effects of time in models with cognitive function and total GM volume as the outcome. Age and DBS were mean centred (Table 1).

Our hypotheses tested whether our variables of interest significantly predicted cognitive function or brain volume at baseline and changed over time. We used likelihood ratio tests to assess the covariate-adjusted significance of predictors on the outcome variables. We also tested for the significance of the interaction of genetic predictors with intellectual enrichment in the same way. In all the analyses, we visually inspected model residual distributions to assess plausible normality. No outliers were identified. Significance was established using two-sided *P*-values and applying Bonferroni correction to control Type I error rate when multiple measures were used to test a hypothesis.

For VBM analyses, a binary GM mask was created using the mean normalized images from all Huntington's disease gene carriers. This was used in all analyses. Statistical maps were thresholded at two-tailed $P < 0.001$ uncorrected at voxel level and $P < 0.05$ family-wise error corrected at cluster level.

Data availability

Track-HD data are available upon request after appropriate data use agreements are signed by the study funder, the CHDI Foundation. Please direct inquiries to info@chdifoundation.org.

Results

Intellectual enrichment

Previous research showed that participants with early stage Huntington's disease who had higher intellectual enrichment had better cognitive performance than those with lower intellectual enrichment.¹³ Furthermore, among pre-manifest gene carriers who are closer to predicted disease onset, those with high intellectual enrichment have a slower rate of cognitive decline than those with low intellectual enrichment.¹²

In our cohort of participants with pre-manifest and early stage Huntington's disease, intellectual enrichment predicted mean global cognitive function and there was also a significant three-way interaction between intellectual enrichment, DBS and time (i.e. annual visit number) on global cognitive function (both $P < 0.001$; Table 2 and Supplementary Table 3). In agreement with previous studies, the estimate for the main effect of intellectual enrichment was positive, such that participants with high intellectual enrichment had better cognitive function than those with lower intellectual enrichment. More specifically, for average DBS and age, the mean estimates [95% confidence interval (CI)] were -3.14 SD ($-3.66, -2.62$) and -5.57 ($-6.10, -5.04$) for high (1SD above mean) and low (1SD below mean) intellectual enrichment, respectively. The contrast estimate [standard error (SE)] for high versus low = 2.43 SD (0.36), $t(229) = 6.744$, $P < 0.001$.

Table 2 Association between intellectual enrichment with cognitive function

Model	DF	AIC	Chisq	P-value
<i>Intellectual enrichment</i>				
Null model	20	3151.6		
Intellectual enrichment	21	3112.3	41.3	<0.001
Intellectual enrichment by visit	22	3113.7	0.6	0.452
Intellectual enrichment by DBS	23	3111.9	3.8	0.050
Intellectual enrichment by DBS by visit	24	3101.4	12.5	<0.001
<i>Intellectual enrichment correcting for education versus education alone</i>				
Null model	20	3151.6		
Education by visit by DBS	24	3133.0	26.5	<0.001
Intellectual enrichment by visit by DBS	28	3105.3	35.7	<0.001
<i>Intellectual enrichment correcting for education versus intellectual enrichment alone</i>				
Null model	20	3151.6		
Intellectual enrichment by visit by DBS	24	3101.4	58.2	<0.001
Education by visit by DBS	28	3105.3	4.1	0.397

The null model for intellectual enrichment included age at baseline, site (three dummy variables) and DBS at baseline with their interaction with visit and main effects of sex and use of antipsychotic medication. A quadratic term was also included for visit, as well as its interaction with DBS.

The estimate for the three-way interaction between DBS, intellectual enrichment and time were in the opposite direction from the main effect; in individuals with low DBS, those with high intellectual enrichment declined slower than those with low intellectual enrichment; however, in individuals with high DBS, those with high intellectual enrichment declined faster than those with lower intellectual enrichment, despite having better performance at baseline (Fig. 1A and Supplementary Fig. 1). More specifically, for participants with low DBS (1SD below mean) those with high intellectual enrichment declined slower than those with lower intellectual enrichment [for 258.9 DBS slope estimate (95% CI) = 0.141 ($-0.007, 0.289$) and -0.102 ($-0.268, 0.065$)] for high and low intellectual enrichment, respectively; contrast estimate (SE) for high versus low intellectual enrichment was 0.243 (0.119), $t(224) = 2.043$, $P = 0.042$). In contrast, an individual with 407.4 DBS (1SD above mean) and 1SD above mean intellectual enrichment had faster cognitive decline than an individual with the same DBS but 1SD below mean intellectual enrichment [slope estimate (95% CI) = -0.741 ($-0.917, -0.565$) and -0.413 ($-0.556, -0.270$)] for high and low intellectual enrichment, respectively; contrast estimate (SE) high versus low = -0.327 (0.110), $t(247) = -2.971$, $P = 0.003$). To aid with interpretation of this finding, we repeated the same analysis, replacing DBS with group as an ordered factor, coding for manifest and pre-manifest individuals. There was a significant main effect of intellectual enrichment; however, the group by visit by intellectual enrichment interaction was not significant in this case (Supplementary Table 4). Our results, therefore, show that there is a strong positive effect of intellectual enrichment on global cognitive function at baseline. It is unclear what

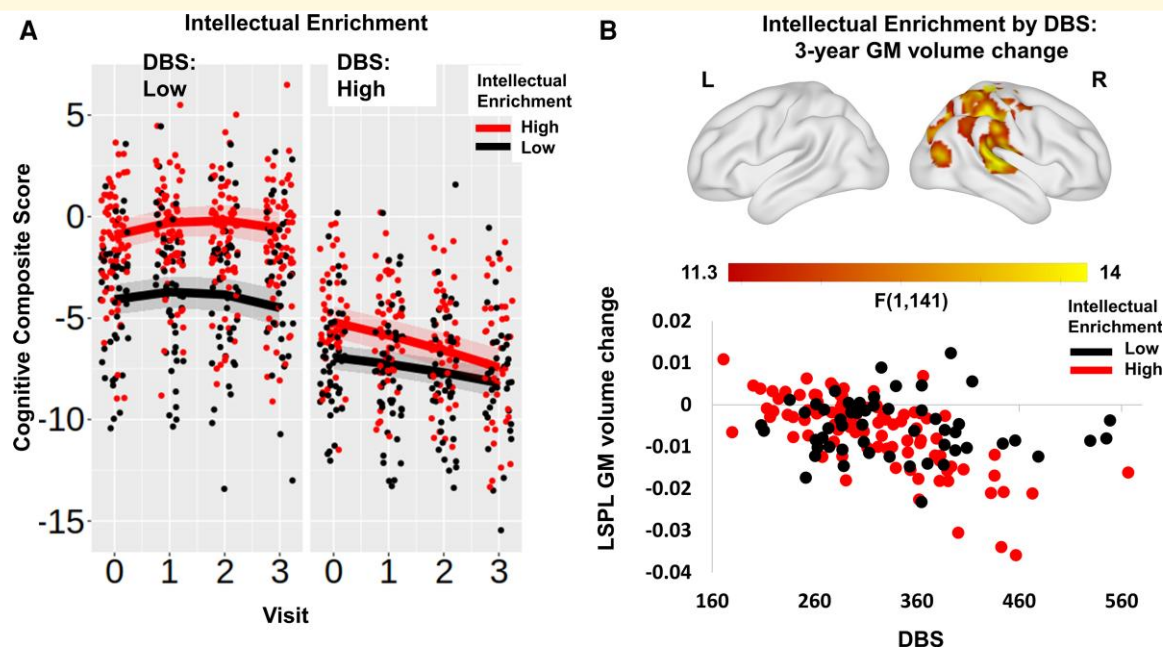


Figure 1 Intellectual enrichment, cognitive function and brain volume. (A) Association between intellectual enrichment and DBS with global cognitive function [$t(225.1) = -3.596, P < 0.001$] and (B) T-map of 3-year GM volume change. In (A) regression lines are generated from a mixed linear model at high (1SD above mean; red) and low (1SD below mean; black) intellectual enrichment. For visualization purposes, results are split into high (above mean) and low (below mean) DBS. The bands around the regression lines are 95% confidence intervals. Data points show the raw data residualized against age, site, sex and use of antipsychotic medication and have been jittered to minimize overlap. In B, significant clusters are overlaid on the ICBM152 template mesh (top). Maps are thresholded at $P < 0.001$ uncorrected at voxel level and $P < 0.05$ family-wise error corrected at cluster level. Shown in a scatter plot (bottom) are the extracted values averaged across the significant cluster. For visualization purposes, data are grouped by high (above mean—red) and low (below mean—black) intellectual enrichment. Data points show the raw data residualized against age, DBS, site, sex and TIV. LSPL, left superior parietal lobe.

is driving the significant three-way interaction between intellectual enrichment, DBS and visits and whether it is a reliable finding. The lack of a significant three-way interaction with the group suggests that it may not be driven by disease stage.

The intellectual enrichment score is a composite measure comprised of education, verbal IQ and occupational cognitive requirements. Education alone is a well-known predictor of cognitive function.⁴⁷ We next wanted to understand whether the effect captured by intellectual enrichment on cognitive function is primarily due to the contribution of education or is incremental to the effect of education. To address this question, we compared a model including all education and intellectual enrichment terms against two different models, one with only the education terms and another with only the intellectual enrichment terms (Table 2; Supplementary Table 5). We found that intellectual enrichment contributed highly significant information to the model above and beyond education alone.

To confirm that the relationship between intellectual enrichment and cognitive function is not driven by motor or neuropsychiatric symptoms, we re-estimated the models, adding unified Huntington's disease rating scale total motor score (TMS) at baseline and the presence of depressive symptoms at baseline with their interactions with visit as confounds. Adding these factors did not alter the results, both

the main effect of intellectual enrichment and the three-way interaction between intellectual enrichment, DBS and visit remained significant (Supplementary Table 6).

To further understand the mechanism by which intellectual enrichment influences global cognitive function, we next examined its relationship with brain volume, measured by caudate and total GM volume over 3 years. There was a significant interaction between intellectual enrichment and DBS on caudate volume at baseline ($p_{\text{bon}} = 0.024$; Supplementary Tables 7 and 8), such that intellectual enrichment had a positive association with caudate volume in Huntington's disease gene carriers far from predicted disease onset (i.e. low DBS), but this effect was attenuated or reversed as the disease progressed (Supplementary Fig. 2A). More specifically, a participant with 258.9 DBS (1SD below mean) and high intellectual enrichment had larger caudate volume at baseline compared with a participant with the same DBS, but low intellectual enrichment [mean estimate (95% CI) = 0.0645 (0.0464, 0.0826) and 0.0291 (0.0097, 0.0486)] for high and low intellectual enrichment, respectively; contrast estimate (SE) high versus low = 0.0354 (0.0139), $t(215) = 2.534, P = 0.012$. However, there was no difference in participants with high DBS (for 407.4 DBS contrast estimate (SE) for high versus low intellectual enrichment was -0.0141 (0.135), $t(215) = -1.044, P = 0.298$). As

Table 3 Association between genetic polymorphisms and cognitive function

Model	DF	AIC	χ^2	P-value	P-value Bonferroni cor.
<i>MSH3</i>					
Null model	20	3139.0			
MSH3	21	3134.2	6.8	0.009	0.045
MSH3 by visit	22	3129.3	6.8	0.009	0.045
MSH3 by DBS	23	3131.1	0.2	0.649	1
MSH3 by DBS by visit	24	3133.1	0.0	0.850	1
<i>FAN1</i>					
Null model	20	3109.3			
FAN1	21	3110.7	0.6	0.4223	1
FAN1 by visit	22	3112.6	0.0	0.895	1
FAN1 by DBS	23	3113.1	1.6	0.213	1
FAN1 by DBS by visit	24	3113.4	1.7	0.193	0.964
<i>MAPT</i>					
Null model	20	3109.3			
MAPT	21	3108.8	2.6	0.110	0.550
MAPT by visit	22	3110.8	0.0	0.988	1
MAPT by DBS	23	3107.2	5.5	0.019	0.094
MAPT by DBS by visit	24	3109.2	0.0	0.947	1
<i>COMT</i>					
Null model	20	3109.3			
COMT	21	3110.8	0.5	0.488	1
COMT by visit	22	3111.2	1.6	0.201	1
COMT by DBS	23	3112.6	0.5	0.462	1
COMT by DBS by visit	24	3112.2	2.4	0.120	0.601
<i>BDNF</i>					
Null model	20	3109.3			
BDNF	21	3111.2	0.1	0.706	1
BDNF by visit	22	3107.1	6.1	0.014	0.068
BDNF by DBS	23	3109.0	0	0.889	1
BDNF by DBS by visit	24	3110.9	0.1	0.741	1

The null model included age at baseline, site (three dummy variables), DBS at baseline with their interaction with visit, sex and use of antipsychotic medication. A quadratic term was also included for visit, as well as its interaction with DBS. P-values were corrected for five independent comparisons using Bonferroni correction.

previously, we repeated the same analysis, replacing DBS with the group as an ordered factor, coding for manifest versus pre-manifest individuals. There was no significant group by intellectual enrichment interaction on caudate volume at baseline (Supplementary Table 9). It is therefore unclear what is driving the intellectual enrichment by DBS interaction and whether it is reliable. There were no significant main effects or interactions with total GM volume (all $p_{\text{bon}} > 0.1$; Supplementary Table 7 and Fig. 2B).

Lastly, we performed exploratory, whole-brain analyses using VBM to identify whether there were specific brain regions that showed an effect of intellectual enrichment or an interaction between intellectual enrichment and DBS. There was no significant main effect of intellectual enrichment on volume or volume change anywhere in the brain, but there was a significant interaction with DBS. Participants with low DBS and high intellectual enrichment had larger GM volume at baseline in the right putamen, the thalamus and the right superior temporal gyrus compared with individuals with similar DBS but low intellectual enrichment (Supplementary Table 10 and Fig. 3). In addition, the rate of GM atrophy over 3 years was faster in

individuals with high intellectual enrichment and high DBS in a cluster extending from the right post-central gyrus to the right superior temporal gyrus ventrally and to the superior parietal lobe and the right precuneus caudally (Fig. 1B and Supplementary Table 10). As previously, we repeated the same analyses, replacing DBS with a group. There were no brain regions that showed a significant interaction between intellectual enrichment and group for baseline volume or 3 year change.

Genetic polymorphisms

We next examined the relationship between cognitive function and five genetic polymorphisms linked to genes known to affect cognitive function in Huntington's disease: *MSH3*, *FAN1*, *MAPT*, *BDNF* and *COMT*. We did not find a significant association between *FAN1*, *MAPT*, *BDNF* and *COMT* variants and global cognitive function at baseline or change over time after Bonferroni correction for five multiple comparisons (all $p_{\text{bon}} > 0.068$; Table 3). In agreement with previous analyses of disease progression in the same cohort²⁷ and recent research,¹⁶ *MSH3* was a significant predictor of global cognitive function at baseline and change (both $p_{\text{bon}} = 0.045$; Table 3 and Supplementary Table 11). More specifically, participants with one or more 3a alleles in *MSH3* had a better cognitive function at baseline and slower cognitive decline compared with non-carriers of 3a alleles [for average age and DBS slope (95% CI) = -0.329 (-0.434 , -0.223) and -0.118 (-0.241 , 0.004)] for non-carriers and carriers, respectively; contrast estimate (SE) non-carriers versus carriers = -0.21 (0.081), $t(213) = -2.587$, $P = 0.010$; Fig. 2A and Supplementary Fig. 4). We repeated the analyses coding for the number of alleles in the genes *MSH3*, *FAN1*, *COMT* and *MAPT*. There was no change in the results (see Supplementary Table 12), i.e. only *MSH3* was a significant predictor of cognitive function at baseline and change over time.

We next examined the relationship between *MSH3* polymorphisms and brain volume to further understand the mechanism by which *MSH3* may influence global cognitive function. *MSH3* had a significant effect on the total GM volume rate of change (i.e. *MSH3* by visit interaction; $p_{\text{bon}} = 0.001$; Supplementary Tables 7, 13 and Fig. 5). Huntington's disease gene carriers with one or more 3a alleles in *MSH3* had a slower rate of total GM atrophy over 3 years compared with non-carriers [for average age and DBS slope (95% CI) = -0.181 (-0.202 , -0.160) and -0.122 (-0.148 , -0.097)] for non-carriers and carriers, respectively; contrast estimate (SE) for non-carriers versus carriers = -0.0586 (0.0168), $t(196) = -3.486$, $P < 0.001$).

Exploratory VBM analyses examined the main effect of *MSH3* on GM volume across the whole brain. There was a significant effect of *MSH3* on GM volume at baseline in the right middle temporal gyrus, the right inferior occipital gyrus and the left post-central gyrus (Supplementary Table 10 and Fig. 6). There was also a significant effect of *MSH3* on GM volume change in the fusiform gyrus

Table 4 Interaction between intellectual enrichment and genetic polymorphisms on cognitive function

Model	DF	AIC	χ^2	P-value	P-value Bonferroni cor.
<i>MSH3</i>					
Null model	24	2931.8			
MSH3 by intellectual enrichment	25	2933.1	0.7	0.404	
MSH3 by intellectual enrichment by visit	26	2933.9	1.1	0.286	
<i>FANI</i>					
Null model	24	2910.6			
FANI by intellectual enrichment	25	2912.4	0.2	0.679	
FANI by intellectual enrichment by visit	26	2914.4	0.0	0.833	
<i>MAPT</i>					
Null model	24	2911.5			
MAPT by intellectual enrichment	25	2913.5	0.0	0.895	
MAPT by intellectual enrichment by visit	26	2915.5	0.0	0.894	
<i>COMT</i>					
Null model	24	2910.2			
COMT by intellectual enrichment	25	2912.2	0.0	0.960	
COMT by intellectual enrichment by visit	26	2914.0	0.2	0.630	
<i>BDNF</i>					
Null model	24	2907.6			
BDNF by intellectual enrichment	25	2902.1	7.5	0.006	0.031
BDNF by intellectual enrichment by visit	26	2903.8	0.2	0.627	

The null model included intellectual enrichment, the genetic polymorphism, age at baseline, site (three dummy variables), DBS at baseline with their interaction with visit, sex and use of antipsychotic medication. A quadratic term was also included for visit, as well as its interaction with DBS. P-values were corrected for five independent comparisons using Bonferroni correction.

BDNF with intellectual enrichment was significant but small on both caudate and total GM volume rate of change (both $p_{\text{bon}} < 0.03$ corrected for two tests; [Supplementary Tables 15–17](#)). The difference in the rate of volume change between individuals with high and low intellectual enrichment was positive in Met66 allele carriers but negative for non-carriers. In more detail, Met66 allele carriers with high (1SD above mean) and low intellectual enrichment (1SD below mean) estimate (95% CI) = -0.0094 (-0.0105 , -0.0082) and -0.0110 (-0.0123 , -0.0097), respectively; contrast estimate (SE) = 0.0017 (0.0009), $t(228) = 1.900$, $P = 0.059$. Met66 allele non-carriers with high (1SD above mean) and low intellectual enrichment (1SD below mean) estimate (95% CI) = -0.0103 (-0.0112 , -0.0094) and -0.0094 (-0.0102 , -0.0085), respectively; contrast estimate (SE) = -0.0010 (0.0007), $t(205) = -1.480$, $P = 0.140$. The difference between high and low intellectual enrichment was positive for Met66 allele carriers, but negative for non-carriers, suggesting that the Met66 allele moderates the effect of intellectual enrichment on caudate atrophy rate [contrast

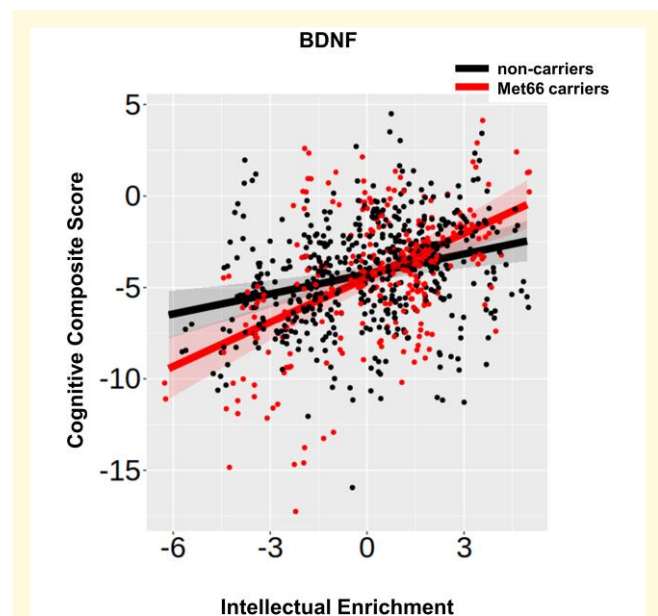


Figure 3 *BDNF*, intellectual enrichment and cognitive function. Association between the *BDNF* predictor and intellectual enrichment with global cognitive function [$t(203.5) = 2.760$, $P = 0.006$]. Regression lines show the predicted effect of carrying (red) and not carrying (black) the Met66 allele. The bands around the regression lines are 95% confidence intervals. Data points show the raw data residualized against age, site, sex and use of antipsychotic medication.

estimate (SE) = -0.0026 (0.0011), $t(221) = -2.406$, $P = 0.017$; [Supplementary Fig. 8](#)].

This was similar to total GM volume. Met66 allele carriers with high (1SD above mean) and low intellectual enrichment (1SD below mean) estimate (95% CI) = -0.129 (-0.167 , -0.090) and -0.206 (-0.248 , -0.1635), respectively; contrast estimate (SE) = 0.0769 (0.0289), $t(214) = 2.658$, $P = 0.008$. Met66 allele non-carriers with high (1SD above mean) and low intellectual enrichment (1SD below mean) estimate (95% CI) = -0.179 (-0.209 , -0.1484) and -0.132 (-0.161 , -0.1020), respectively; contrast estimate (SE) = -0.0469 (0.0218), $t(192) = -2.154$, $P = 0.0324$. The difference between high and low intellectual enrichment was positive for Met66 allele carriers but negative for non-carriers, suggesting that the Met66 allele moderates the effect of intellectual enrichment on total GM atrophy rate [contrast estimate (SE) = -0.124 (0.0362), $t(207) = -3.421$, $P = 0.008$; [Supplementary Fig. 9](#)].

Exploratory whole-brain VBM analyses examined the interaction between *BDNF* and intellectual enrichment on GM volume at baseline and change over time, but we did not identify any significant clusters.

Discussion

The aim of our study was to examine the role of intellectual enrichment and genetic polymorphisms on cognitive function and brain structure in Huntington's disease. Our results

a small effect size ($r = -0.14$). In our study, we did not find strong evidence for an association between *MAPT* and cognitive function; it is therefore currently unclear whether *MAPT* plays a role in cognitive function in Huntington's disease.

Conclusion

In summary, we have shown that cognitive function in Huntington's disease is affected by an interplay between genetic and environmental factors. We have replicated previous findings that cognitive decline is slower in carriers of the 3a allele in the gene *MSH3*, and further showed that slower cognitive decline is supported by the slowing of GM volume atrophy in the cortex, in agreement with the role of *MSH3* in somatic instability. Intellectual enrichment also appears to have a protective effect on cognitive function at pre-manifest stages of the disease, but as the disease progresses, this effect is attenuated and there is faster neurodegeneration and cognitive decline. Importantly, we also observed a significant interaction between intellectual enrichment and the *BDNF* gene, whereby intellectual enrichment counteracted the detrimental effect of carrying the Met66 allele on cognitive function and brain structure, in agreement with the role of intellectual enrichment in enhancing brain trophic support. Future research is now needed to develop and evaluate intellectual enrichment interventions in Huntington's disease and measure their impact on both behaviour and brain structure.

Funding

This work was supported by a Huntington's Disease Society of America (HDSA) Human Biology fellowship award to M.P. S.J.T.'s work is supported by the UK Dementia Research Institute (DRI) which receives its funding from DRI Ltd, funded by the UK Medical Research Council, Alzheimer's Society and Alzheimer's Research UK. The Track-HD study was funded by the CHDI Foundation.

Competing interests

MP is an employ of IXICO plc. All other authors report no competing interests.

Supplementary material

Supplementary material is available at *Brain Communications* online.

Appendix

Track-HD investigators

Peter Kraus, Rainer Hoffman, Alan Tobin, Beth Borowsky, S. Keenan, Kathryn B. Whitlock, Sarah Queller, Colin Campbell, Chiachi Wang, Eric Axelson, Hans Johnson,

Tanka Acharya, Dave M. Cash, Chris Frost, Rebecca Jones, Caroline Jurgens, Ellen P. 't Hart, Jeroen van der Grond, Marie-Noelle N. Witjes-Ane, Raymund A.C. Roos, Eve M. Dumas, Simon J.A. van den Bogaard, Cheryl Stopford, David Craufurd, Jenny Callaghan, Natalie Arran, Diana D. Rosas, S. Lee, W Monaco, Alison O'Regan, Cassie Milchman, Ellen Frajman, Izelle Labuschagne, Julie Stout, Melissa Campbell, Sophie C. Andrews, Natalie Bechtel, Ralf Reilmann, Stefan Bohlen, Chris Kennard, Claire Berna, Stephen Hicks, Alexandra Durr, Christophe Pourchot, Eric Bardinet, Kevin Nigaud, Romain Valabrègue, Stephane Lehericy, Cecilia Marelli, Celine Jauffret, Damian Justo, Blair Leavitt, Joji Decolongon, Aaron Sturrock, Alison Coleman, Rachelle Dar Santos, Aakta Patel, Claire Gibbard, Daisy Whitehead, Ed Wild, Gail Owen, Helen Crawford, Ian Malone, Nayana Lahiri, Nick C. Fox, Nicola Z. Hobbs, Roger Ordidge, Tracey Pepple, Joy Read, Miranda J. Say, Bernhard Landwehrmeyer.

References

1. Snowden JS. The neuropsychology of Huntington's disease. *Arch Clin Neuropsychol*. 2017;32(7):876-887.
2. Duff K, Paulsen J, Mills J, et al. Mild cognitive impairment in pre-diagnosed Huntington disease. *Neurology*. 2010;75(6):500-507.
3. Nyberg L, Lövdén M, Riklund K, Lindenberg U, Bäckman L. Memory aging and brain maintenance. *Trends Cogn Sci*. 2012; 16(5):292-305.
4. Stern Y. Cognitive reserve. *Neuropsychologia*. 2009;47(10): 2015-2028.
5. Vemuri P, Lesnick TG, Przybelski SA, et al. Association of lifetime intellectual enrichment with cognitive decline in the older population. *JAMA Neurol*. 2014;71(8):1017-1024.
6. Harris SE, Deary IJ. The genetics of cognitive ability and cognitive ageing in healthy older people. *Trends Cogn Sci*. 2011;15(9):388-394.
7. Ward DD, Summers MJ, Saunders NL, Ritchie K, Summers JJ, Vickers JC. The BDNF Val66Met polymorphism moderates the relationship between cognitive reserve and executive function. *Transl Psychiatry*. 2015;5(6):e590.
8. Premi E, Grassi M, van Swieten J, et al. Cognitive reserve and TMEM106B genotype modulate brain damage in presymptomatic frontotemporal dementia: A GENFI study. *Brain*. 2017;140(6):1784-1791.
9. Gazzina S, Grassi M, Premi E, et al. Education modulates brain maintenance in presymptomatic frontotemporal dementia. *J Neurol Neurosurg Psychiatry*. 2019;90(10):1124-1130.
10. van Dellen A, Blakemore C, Deacon R, York D, Hannan AJ. Delaying the onset of Huntington's in mice. *Nature*. 2000;404(6779):721-722.
11. Trembath MK, Horton ZA, Tippet L, et al. A retrospective study of the impact of lifestyle on age at onset of Huntington disease. *Mov Disord*. 2010;25(10):1444-1450.
12. Bonner-Jackson A, Long JD, Westervelt H, Tremont G, Aylward E, Paulsen JS. Cognitive reserve and brain reserve in prodromal Huntington's disease. *J Int Neuropsychol Soc*. 2013;19(7): 739-750.
13. Garcia-Gorro C, Garau-Rolandi M, Escrichs A, et al. An active cognitive lifestyle as a potential neuroprotective factor in Huntington's disease. *Neuropsychologia*. 2019;122:116-124.
14. Long JD, Lee JM, Aylward EH, et al. Genetic modification of Huntington disease acts early in the prediagnosis phase. *Am J Hum Genet*. 2018;103(3):349-357.
15. De Diego-Balaguer R, Schramm C, Rebeix I, et al. COMT Val158met polymorphism modulates Huntington's disease progression. *PLoS One*. 2016;11(9):e0161106.

16. Lee JM, Huang Y, Orth M, *et al.* Genetic modifiers of Huntington's disease differentially influence motor and cognitive domains. *Am J Hum Genet.* 2022;109(5):885-899.
17. Vuono R, Winder-Rhodes S, de Silva R, *et al.* The role of tau in the pathological process and clinical expression of Huntington's disease. *Brain.* 2015;138(7):1907-1918.
18. Tabrizi SJ, Langbehn DR, Leavitt BR, *et al.* Biological and clinical manifestations of Huntington's disease in the longitudinal TRACK-HD study: Cross-sectional analysis of baseline data. *Lancet Neurol.* 2009;8(9):791-801.
19. Tabrizi SJ, Scahill RI, Durr A, *et al.* Biological and clinical changes in premanifest and early stage Huntington's disease in the TRACK-HD study: The 12-month longitudinal analysis. *Lancet Neurol.* 2011;10:31-42.
20. Tabrizi SJ, Reilmann R, Roos RA, *et al.* Potential endpoints for clinical trials in premanifest and early Huntington's disease in the TRACK-HD study: Analysis of 24 month observational data. *Lancet Neurol.* 2012;11(1):42-53.
21. Tabrizi SJ, Scahill RI, Owen G, *et al.* Predictors of phenotypic progression and disease onset in premanifest and early-stage Huntington's disease in the TRACK-HD study: Analysis of 36-month observational data. *Lancet Neurol.* 2013;12(7):637-649.
22. Bates GP, Dorsey R, Gusella JF, *et al.* Huntington disease. *Nat Rev Dis Primer.* 2015;1:15005.
23. Langbehn DR, Stout JC, Gregory S, *et al.* Association of CAG repeats with long-term progression in Huntington disease. *JAMA Neurol.* 2019;76:1375-1385.
24. Moss DJH, Pardiñas AF, Langbehn D, *et al.* Identification of genetic variants associated with Huntington's disease progression: A genome-wide association study. *Lancet Neurol.* 2017;16(9):701-711.
25. Purcell S, Neale B, Todd-Brown K, *et al.* PLINK: A tool set for whole-genome association and population-based linkage analyses. *Am J Hum Genet.* 2007;81(3):559-575.
26. Genetic Modifiers of Huntington's Disease (GeM-HD) Consortium. Identification of genetic factors that modify clinical onset of Huntington's disease. *Cell.* 2015;162(3):516-526.
27. Flower M, Lomeikaite V, Ciosi M, *et al.* MSH3 Modifies somatic instability and disease severity in Huntington's and myotonic dystrophy type 1. *Brain.* 2019;142(7):1876-1886.
28. Genetic Modifiers of Huntington's Disease (GeM-HD) Consortium. CAG Repeat not polyglutamine length determines timing of Huntington's disease onset. *Cell.* 2019;178(4):887-900.e14.
29. Buchman AS, Yu L, Boyle PA, Schneider JA, De Jager PL, Bennett DA. Higher brain BDNF gene expression is associated with slower cognitive decline in older adults. *Neurology.* 2016;86(8):735.
30. Lim YY, Hassenstab J, Cruchaga C, *et al.* BDNF Val66met moderates memory impairment, hippocampal function and tau in preclinical autosomal dominant Alzheimer's disease. *Brain.* 2016;139(10):2766-2777.
31. Lim YY, Hassenstab J, Goate A, *et al.* Effect of BDNFVal66Met on disease markers in dominantly inherited Alzheimer's disease. *Ann Neurol.* 2018;84(3):424-435.
32. Zuccato C, Cattaneo E. Role of brain-derived neurotrophic factor in Huntington's disease. *Prog Neurobiol.* 2007;81(5):294-330.
33. Plotkin JL, Day M, Peterson JD, *et al.* Impaired TrkB receptor signaling underlies corticostriatal dysfunction in Huntington's disease. *Neuron.* 2014;83(1):178-188.
34. Ward DD, Andel R, Saunders NL, *et al.* The BDNF Val66Met polymorphism moderates the effect of cognitive reserve on 36-month cognitive change in healthy older adults. *Alzheimers Dement Transl Res Clin Interv.* 2017;3(3):323-331.
35. Vuono R, Kouli A, Legault EM, *et al.* Association between toll-like receptor 4 (TLR4) and triggering receptor expressed on myeloid cells 2 (TREM2) genetic variants and clinical progression of Huntington's disease. *Mov Disord.* 2020;35(3):401-408.
36. Bettencourt C, Hensman-Moss D, Flower M, *et al.* DNA repair pathways underlie a common genetic mechanism modulating onset in polyglutamine diseases. *Ann Neurol.* 2016;79(6):983-990.
37. Stefansson H, Helgason A, Thorleifsson G, *et al.* A common inversion under selection in Europeans. *Nat Genet.* 2005;37(2):129-137.
38. Valenzuela MJ, Sachdev P. Brain reserve and dementia: A systematic review. *Psychol Med.* 2006;36(4):441-454.
39. Pool LR, Weuve J, Wilson RS, Bültmann U, Evans DA, Mendes de Leon CF. Occupational cognitive requirements and late-life cognitive aging. *Neurology.* 2016;86(15):1386-1392.
40. Penney JB, Vonsattel JP, Macdonald ME, Gusella JF, Myers RH. CAG repeat number governs the development rate of pathology in Huntington's disease. *Ann Neurol.* 1997;41(5):689-692.
41. Langbehn D, Brinkman R, Falush D, Paulsen J, Hayden M, on behalf of an International Huntington's Disease Collaborative Group. A new model for prediction of the age of onset and penetrance for Huntington's disease based on CAG length. *Clin Genet.* 2004;65(4):267-277.
42. Freeborough PA, Fox NC, Kitney RI. Interactive algorithms for the segmentation and quantitation of 3-D MRI brain scans. *Comput Methods Programs Biomed.* 1997;53(1):15-25.
43. Hobbs NZ, Henley SMD, Wild EJ, *et al.* Automated quantification of caudate atrophy by local registration of serial MRI: Evaluation and application in Huntington's disease. *NeuroImage.* 2009;47(4):1659-1665.
44. Freeborough PA, Fox NC. Modeling brain deformations in Alzheimer disease by fluid registration of serial 3D MR images. *J Comput Assist Tomogr.* 1998;22(5):838-843.
45. Ashburner J, Friston KJ. Voxel-based morphometry—The methods. *NeuroImage.* 2000;11(6):805-821.
46. Johnson EB, Byrne LM, Gregory S, *et al.* Neurofilament light protein in blood predicts regional atrophy in Huntington disease. *Neurology.* 2018;90(8):e717.
47. Stern Y. Cognitive reserve in ageing and Alzheimer's disease. *Lancet Neurol.* 2012;11(11):1006-1012.
48. Hensman Moss DJ, Flower MD, Lo KK, *et al.* Huntington's disease blood and brain show a common gene expression pattern and share an immune signature with Alzheimer's disease. *Sci Rep.* 2017;7(1):44849.
49. Zuccato C, Cattaneo E. Brain-derived neurotrophic factor in neurodegenerative diseases. *Nat Rev Neurol.* 2009;5(6):311-322.
50. Spires TL, Grote HE, Varshney NK, *et al.* Environmental enrichment rescues protein deficits in a mouse model of Huntington's disease, indicating a possible disease mechanism. *J Neurosci.* 2004;24(9):2270-2276.
51. Swami M, Hendricks AE, Gillis T, *et al.* Somatic expansion of the Huntington's disease CAG repeat in the brain is associated with an earlier age of disease onset. *Hum Mol Genet.* 2009;18(16):3039-3047.
52. Ciosi M, Maxwell A, Cumming SA, *et al.* A genetic association study of glutamine-encoding DNA sequence structures, somatic CAG expansion, and DNA repair gene variants, with Huntington disease clinical outcomes. *EBioMedicine.* 2019;48:568-580.
53. Mouro Pinto RM, Arning L, Giordano JV, *et al.* Patterns of CAG repeat instability in the central nervous system and periphery in Huntington's disease and in spinocerebellar ataxia type 1. *Hum Mol Genet.* 2020;29(15):2551-2567.
54. Goold R, Flower M, Moss DH, *et al.* FAN1 modifies Huntington's disease progression by stabilizing the expanded *HTT* CAG repeat. *Hum Mol Genet.* 2019;28(4):650-661.
55. Williams-Gray CH, Evans JR, Goris A, *et al.* The distinct cognitive syndromes of Parkinson's disease: 5 year follow-up of the CamPaIGN cohort. *Brain.* 2009;132(11):2958-2969.