Reduced amygdala volumes are related to motor and cognitive signs in Huntington's disease: The IMAGE-HD study

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A B S T R A C T

In Huntington's disease (HD), the presence of neurodegeneration in brain regions other than the striatum has been recently gaining attention. The amygdala is one such area, which has been investigated in only eight structural magnetic resonance imaging studies to date, but with inconsistent findings. This is the largest MRI study to date examining manually traced amygdala volumes in HD participants and the relationship of amygdala volumes to clinical measures of HD. Our study included 35 healthy control participants, and groups of 35 pre-symptomatic, and 36 symptomatic HD participants. When comparing the pre-symptomatic and symptomatic HD groups together against the control group, amygdala volumes were significantly lower in HD than controls and in symptomatic HD than pre-symptomatic HD. When examining relationships between amygdala volumes and clinical measures of HD, significantly smaller amygdala volumes were associated with worse motor and cognitive signs. For pre-symptomatic HD participants who were close to disease onset, smaller amygdala volumes were also associated with higher levels of anxiety symptoms. These findings suggest that the amygdala is affected in pre-symptomatic and symptomatic HD, and that the amygdala is related to the clinical profile of HD before onset of motor symptoms.

1. Introduction

Huntington's disease (HD) is an autosomal dominant neurodegenerative disorder that causes motor, cognitive, and psychiatric symptoms (Bates et al., 2015; Papoutsi et al., 2014). Historically, neuropathological and neuroimaging studies in HD focussed on the striatum, the brain area most severely affected across the course of the disease (Walker, 2007; Georgiou-Karistianis et al., 2013). Atrophy in brain regions outside of the striatum, and early in the course of HD, however, is of considerable interest (cortical areas: Tabrizi et al., 2009; Gray et al., 2013; thalamus: Jernigan et al., 1991; hippocampus: Brito et al., 2014). The amygdala, a subcortical almond-shaped structure located deep in the medial temporal lobe, has been gaining interest because of its close functional connectivity with the striatum, the brain region most severely affected across the course of HD (Roy et al., 2009). Further, the amygdala has been ascribed to key cognitive and psychiatric symptoms that occur in the course of HD, including aberrant emotion recognition,

regulation, and social behaviour, depression, and anxiety (Adolphs et al., 1994; Adolphs et al., 1997; Fine and Blair, 2000; Phelps and Ledoux, 2005). Nevertheless, only few studies have investigated the involvement of the amygdala in HD, and so far, results have been inconsistent.

The first study to demonstrate amygdala abnormalities in HD compared voxel-based morphometry and manual segmentation for measuring subcortical volumes (Douaud et al., 2006). Although the research focus was not on the amygdala, the voxel-based morphometry analysis indicated smaller amygdala volumes in symptomatic HD participants when compared to controls (Douaud et al., 2006). A later study showed similar results in pre-symptomatic HD (Kipps et al., 2007). In fMRI studies, pre-symptomatic and symptomatic HD participants have shown less activity in the amygdala and less connectivity between the amygdala and the fusiform face area than controls (Mason et al., 2015; Van Den Stock et al., 2015; Klöppel et al., 2010). In addition, a recent meta-analysis that investigated current evidence on

brain atrophy in HD has demonstrated that smaller amygdala volumes are generally shown in pre-symptomatic HD compared to controls (Dogan et al., 2013). Together, these studies indicate that the amygdala is affected at pre-symptomatic and symptomatic stages of HD. Of note are also findings of amygdala atrophy in Parkinson's disease (De La Monte et al., 1989; Cordato et al., 2000; Harding et al., 2002), often used as a comparison to HD because of the mutual yet selective involvement of the basal ganglia.

In contrast to evidence that the amygdala is affected in HD, other studies have not revealed evidence for amygdala degeneration in HD. For example, using the transgenic rat model of HD, no differences in amygdala volumes were found between wild-type rats and HD rats (Faure et al., 2011). Similarly, in a volumetric human study, the amygdala was the only structure of seven brain areas (including the basal ganglia, hippocampus, and thalamus) that was not smaller in pre-symptomatic and symptomatic HD compared to controls (Van Den Bogaard et al., 2011). These inconsistent findings may be a result of underpowered studies due to small sample sizes, and the use of automated segmentation methods, which are insensitive to subtle and spatially complex grey matter differences between groups (Davatzikos, 2004). Alongside inconsistent findings, no HD study has yet compared amygdala volumes between participants with pre-symptomatic HD and symptomatic HD, or related amygdala volumes to clinical symptoms.

In this paper, we report a cross-sectional analysis of amygdala volumetry in pre-symptomatic and symptomatic HD using data from the Australian-based IMAGE-HD study (Dominguez et al., 2013; Georgiou-Karistianis et al., 2013; Dominguez et al., 2016). In addition, we report on relationships between amygdala volumes and clinical and pathological measures, including motor and psychiatric symptoms, disease burden, and caudate and putamen volumes. Based on previous work suggesting that amygdala atrophy may occur in HD, we hypothesised that amygdala volumes would be smaller in participants with the HD gene overall (both pre-symptomatic and symptomatic), and relatively smaller for symptomatic HD than pre-symptomatic HD participants. In light of previous evidence of associations between atrophy in the striatum and sensorimotor cortex and clinical measures in HD (Dogan et al., 2013; Tabrizi et al., 2011), we hypothesised that smaller amygdala volumes would be associated with higher levels of motor, cognitive, and psychiatric symptoms, and worse disease burden scores and smaller caudate and putamen volumes.

2. Material and methods

2.1. Participants

The sample consisted of 106 participants, of whom 59 were females: 35 pre-symptomatic HD, 36 symptomatic HD, and 35 healthy controls (Table 1). The age of all participants ranged from 23 to 72 years old ($M = 45.44$, $SD = 12.17$), but because of the natural course of HD, the symptomatic HD group was older than the pre-symptomatic HD and control groups ($F(2,103) = 8.94$, $p < .001$). Gender did not differ significantly between groups ($F(2,103) = 2.31$, $p = .10$). The study was approved by Monash University's Human Research Ethics Committee and Melbourne Health Human Research Ethics Committee, and informed written consent was obtained from each participant prior to testing.

2.2. Amygdala volumetry

2.2.1. Image acquisition and pre-processing

High resolution MRI data from the IMAGE-HD study was acquired using a Siemens 3 Telsa scanner with a 32-channel head coil at the Murdoch Children's Research Institute (Royal Children's Hospital, Victoria, Australia). Acquisition parameters for the T1-weighted images consisted of 192 slices, voxel size 0.9, 0.8 mm × 0.8 mm, 9° flip angle. FOV was 320 × 320, inversion time 900 ms, TE 2.59 ms, RT 1900 ms. To allow for comparison of regional grey matter volumes, the raw MRI images were re-oriented, and re-sized using the Freesurfer imaging analysis suite (Martinos Center for Biomedical Imaging, Harvard-MIT, Boston, USA). Participant's images were de-identified prior to volumetric analysis to minimise any biases.

2.2.2. Volumetric analysis

The right and left amygdala was manually delineated on coronal slices using the software ANALYZE 12.0 (Mayo Clinic, Rochester, NY), and following a well validated protocol (Velakoulis et al., 2006). Segmentation began at the caudal end of the amygdala and proceeded, slice by slice, rostrally until the amygdala was no longer visible. To ensure consistency of manual segmentation across each brain, each slice was displayed with the segmentations from the previous slice, which was then edited to reflect the shift in the amygdala boundary for that slice.

The total amygdala volume was calculated by summing the volume for the voxels included across all slices. To account for individual differences in head size, right and left amygdala volumes were normalised by regressing out total intracranial volume, following the residual

Table 1
Participants' demographic information.

	Pre-symptomatic HD	Symptomatic HD	Controls
n	35	36	35
Males (%)	14 (40%)	21 (58%)	12 (34%)
Females (%)	21 (60%)	15 (42%)	23 (66%)
Mood medication (%)	6 (17%)	16 (44%)	0
Age (mean ± SD)	41.59 ± 10.00	51.91 ± 9.36	42.63 ± 14.14
(Range)	(23.93–65.29)	(38.99–70.84)	(24.38–72.98)
HADS Anxiety (mean ± SD)	6.83 ± 3.37	5.47 ± 3.44	4.97 ± 2.81
(Range)	(1–14)	(0–14)	(0−11)
HADS Depression (mean ± SD)	2.74 ± 2.97	2.89 ± 2.38	2.23 ± 2.10
(Range)	(0−12)	(0–8)	(0–8)
UHDRS TMS (mean ± SD)	0.94 ± 1.24	19.47 ± 12.42	–
(Range)	(0–4)	(6–60)	–
CAG repeats (mean ± SD)	42.31 ± 1.97	43.17 ± 2.48	–
(Range)	(39–46)	(40–50)	–
DBS (mean ± SD)	269.70 ± 53.41	379.70 ± 70.02	-
(Range)	(131.64–369.60)	(258.14–556.97)	-

Abbreviations. UHDRS TMS = Unified Huntington's Disease Rating Scale total motor score. HADS = Hospital Anxiety and Depression Scale. DBS = disease burden score.

method described by Free et al. (1995).

For intra-rater and inter-rater reliability for amygdala, tracing was assessed using intra-class correlation coefficient (ICC). Intra-rater reliability was determined for the right (ICC = 0.97) and left amygdala (ICC = 0.96) using ten randomly selected MRIs delineated by the same tracer (LA) on two occasions, at the beginning of the tracing period and at the end, three months later, to map any potential drift in reliability over time (ICCs were 0.89 for the right and 0.84 for the left amygdala). To establish inter-rater reliability, a different rater (YG-J) measured amygdala volumes on the same set of ten scans ICCs, and inter-rater reliabilities were 0.88 and 0.80 for right and left amygdala, respectively.

2.3. Clinical measures and measures of disease pathology

We measured psychiatric symptoms with the Hospital Anxiety and Depression Scale (HADS; Zigmond and Snaith, 1983), and motor symptoms with the Unified Huntington's Disease Rating Scale (UHDRS; Huntington Study Group, 1996) total motor scores (TMS). Cognitive symptoms were measured using the Symbol Digit Modalities Test (SDMT; Smith, 1982), Stroop Word Test (Stroop, 1935), and the University of Pennsylvania Smell Identification Test (UPSIT; Doty et al., 1984).

The HADS is a self-administered questionnaire that screens for levels of depression and anxiety, and consists of 7 items scored on a scale of 0–3. Scores above 7 on either subscale indicate significant levels of depression or anxiety. Our three groups showed similar levels of depressive symptoms, and the pre-symptomatic HD group indicated slightly higher anxiety symptoms than controls (Georgiou-Karistianis et al., 2013).

The SDMT is a brief measure of visuomotor speed and attention, and involves matching symbols to their corresponding numbers in 90 s. The SDMT is scored according to the number of correct answers. Next, the Stroop Word Test is a measure of processing speed, which involves naming the colour of written colour names as quickly as possible. The Stroop Word Test is scored according to number of correct words. Next, the UPSIT is a measure of odour recognition, which involves categorising different odours and is scored according to normative data. In our sample, symptomatic HD participants performed worse on the SDMT, Stroop Word Test, and UPSIT than pre-symptomatic and control participants. In addition, pre-symptomatic HD participants performed worse than controls on the SDMT, although both groups performed similarly on the UPSIT and Stroop Word Test (Georgiou-Karistianis et al., 2013).

The motor section of the UHDRS is an assessment of motor signs of HD by a trained examiner, and consists of 31 items scored on a range from 0 to 4, and summed for the TMS. A TMS over 5 demonstrates substantial motor signs. In our sample, the symptomatic HD group showed greater motor signs than the pre-symptomatic HD group (Georgiou-Karistianis et al., 2013).

We used previously acquired data from the IMAGE-HD study for measures of disease pathology, which included disease burden scores (DBS) and volumetry of the caudate nucleus and putamen (Georgiou-Karistianis et al., 2013) to examine the relationship between disease pathology and amygdala volumes. The DBS is an estimate of the pathological burden of HD, and is calculated based on the participant age and cytosine-guanine-adenine (CAG) repeat length (age × [CAG repeat number − 35.5]) (Dominguez et al., 2013; Penney et al., 1997). As expected, the symptomatic HD group showed greater disease burden than the pre-symptomatic HD group ($F(1,69) = 55.17$, $p < .001$). Estimated years to diagnosis for the pre-symptomatic HD participants was calculated using the following formula: years to onset = (21.54 + EXP (9.556 − 0.146 × CAG repeat number) − age (Langbehn et al., 2004). Volumes of the caudate nucleus and putamen were not available for 3 of our pre-symptomatic and 6 of our symptomatic participants, and therefore these 9 HD participants were removed from this analysis. In the resulting sample, the symptomatic HD group showed smaller

caudate and putamen volumes than the pre-symptomatic HD group (Wilk's Lambda = 0.80, $F(4,57) = 3.59$, $p = .011$), as previously shown by Georgiou-Karistianis et al. (2013).

2.4. Statistical analyses

Total amygdala volumes were compared between the three groups (pre-symptomatic HD, symptomatic HD, and controls) using an analysis of variance (ANOVA) with two planned contrasts. The first contrast examined differences in total amygdala volumes between controls and both HD groups (pre-symptomatic and symptomatic HD), and the second contrast investigated differences between the pre-symptomatic and symptomatic HD groups. The association between amygdala volumes and UHDRS TMS, HADS Anxiety and Depression scores, SDMT, Stroop Word Test, UPSIT, DBS, and caudate and putamen volumes were examined in bivariate linear regression analyses. We used SPSS version 23 (SPSS Inc., Chicago, IL, USA) for all statistical analyses.

3. Results

We found that amygdala volumes were lower in both symptomatic HD and pre-symptomatic HD compared to controls, and in symptomatic HD compared to pre-symptomatic HD. Within all groups, left and right amygdalae were non-significantly different [control group: F $(1,68) = 0.10$, $p = .75$, pre-symptomatic HD: $F(1,68) = 0.033$, $p = .86$, symptomatic HD: $F(1,70) = 0.052$, $p = .82$]. The effect of group membership on total amygdala volumes (as well as right and left separately) was significant ($F(2,103) = 27.64$, $p < .001$, with a large effect size, $\eta_p^2 = 0.40$; Table 2, Fig. 1) with both pre-symptomatic and symptomatic HD participants having smaller total amygdala volumes than controls, $t(103) = 7.34$, $p < .001$ (large effect size, $\eta_p^2 = 0.34$). Further, symptomatic HD participants showed smaller total amygdala volumes than pre-symptomatic HD, $t(103) = 2.56$, $p = .012$, with a small effect size, $\eta_p^2 = 0.060$.

Symptomatic HD participants are naturally older than those with pre-symptomatic HD. Moreover, the controls were matched by age to the pre-symptomatic HD participants, and thus in our sample age and group are confounded. To address this confound, in a separate set of analyses, we excluded the four youngest control participants ($n = 31$ remaining), the five youngest pre-symptomatic HD participants ($n = 30$ remaining), and the five oldest symptomatic HD participants ($n = 32$

Table 2
Amygdala volumetry for pre-symptomatic HD, symptomatic HD, and healthy controls within the total sample and age-matched sample.

	Hemisphere	Mean (SD)	95% CI
Total sample			
Pre-symptomatic HD	R	979.90 (129.59)	[927.36, 1032.43]
	L	974.27 (129.01)	[924.75, 1032.79]
	Total	1954.17 (235.65)	[1860.58, 2047.76]
Symptomatic HD	R	895.83 (144.58)	[844.03, 947.64]
	L	888.41 (132.61)	[839.78, 937.24]
	Total	1784.24 (256.58)	[1691.97, 1876.52]
Controls	R	1128.21 (189.98)	[1075.67, 1180.75]
	L	1142.17 (177.09)	[1092.65, 1191.69]
	Total	2270.38 (335.85)	[2176.80, 2363.97]
Age-matched sample			
Pre-symptomatic HD	R	986.09 (124.73)	[934.06, 1038.12]
	L	970.00 (121.35)	[919.92, 1020.08]
	Total	1956.09 (228.56)	[1863.20, 2048.99]
Symptomatic HD	R	913.78 (139.31)	[863.40, 964.16]
	L	894.61 (131.42)	[846.12, 943.10]
	Total	1808.39 (252.28)	[1718.45, 1898.33]
Controls	R	1110.83 (163.22)	[1059.65, 1162.02]
	L	1138.58 (158.46)	[1089.31, 1187.85]
	Total	2249.41 (283.72)	[2158.03, 2340.79]

Abbreviations. R = right amygdala. L = left amygdala. Total = right + left amygdala. CI = confidence interval.
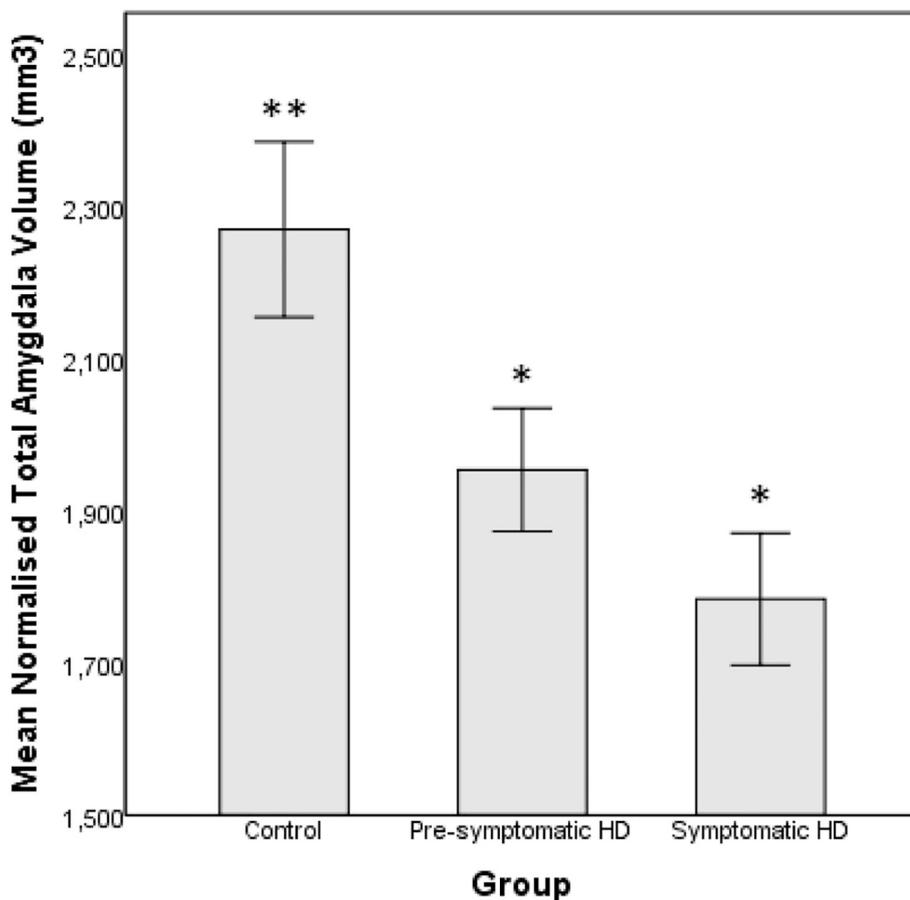
remaining; Table 2). Although this sample yielded relatively less power than the analyses with the full sample, analyses of this age-matched subset indicated that the group effect remained statistically significant for the amygdala volumes ($F(2,90) = 24.09$, $p < .001$, large effect, $\eta^2 = 0.34$). The pattern of differences between the groups in total amygdala volumes was unchanged with smaller volumes in pre-symptomatic HD and in symptomatic HD than controls, ($t(90) = 6.83$, $p < .001$, large effect size, $\eta^2 = 0.34$), and smaller volumes in symptomatic HD than pre-symptomatic HD ($t(90) = 2.27$, $p = .026$, small effect size, $\eta^2 = 0.054$). By combining the unmatched and age-matched approaches, we suggest that the differences in amygdala volumes between the three groups were not attributable to age.

We found that smaller total amygdala volumes were associated with worse motor signs (UHDRS TMS scores) and poorer cognitive performances (SDMT and Stroop Word Test scores) in pre-symptomatic and symptomatic HD participants (Table 3; Fig. 2). Amygdala volumes were not associated with odour recognition (UPSIT scores), or psychiatric symptoms (HADS scores), although both results suggested statistical trends. Finally, disease burden (i.e. DBS), and caudate and putamen volumes were also not significantly associated with amygdala volumes.

Psychiatric symptoms are thought to occur in HD before clinical diagnosis, and in order to asses if the amygdala is related to these symptoms we analysed the pre-symptomatic cohort further. We divided the pre-symptomatic HD group into far and close to estimated disease onset, using a median of 9.66 years to diagnosis. We found that in the close to onset group, lower total amygdala volumes were associated with greater anxiety symptoms and worse motor signs (Table 4, Fig. 3). In contrast, in the far from onset group of pre-symptomatic HD participants, amygdala volumes were not associated with any clinical measures. Amygdala volumes were not associated with odour recognition or cognitive performance in either group.

Table 3

Inferential statistics for the relationship between amygdala volumes and clinical measures and measures of disease pathology in HD participants.

	B	SE	t	p	η^2	95% CI
UHDRS TMS	−7.47	2.26	−3.31	.001**	0.14	[−11.98, −2.97]
SDMT	5.70	2.31	2.47	.016**	0.081	[1.09, 10.32]
Stroop Word Test	3.08	1.36	2.27	.026**	0.070	[0.38, 5.78]
UPSIT	4.75	4.43	1.07	.29	0.016	[−4.09, 13.58]
HADS Anxiety	6.99	11.06	0.63	.53	0.006	[−15.09, 29.06]
HADS Depression	−11.35	14.30	−0.79	.43	0.009	[−39.87, 17.18]
Disease burden score	−0.70	0.37	−1.92	.059	0.051	[−1.43, 0.029]
Right caudate	−19.41	16.72	−1.16	.25	0.023	[−52.89, 14.08]
Left caudate	25.37	16.64	1.53	.13	0.039	[−7.95, 58.70]
Right putamen	−14.35	19.06	−0.75	.46	0.010	[−52.52, 23.83]
Left putamen	3.77	18.21	0.21	.84	0.001	[−32.69, 40.24]

Abbreviations. B = unstandardised regression coefficient. SE = standard error. CI = confidence interval. UHDRS TMS = Unified Huntington's Disease Rating Scale Total Motor Score. SDMT = Symbol Digit Modalities Test. UPSIT = University of Pennsylvania Smell Identification Test. HADS = Hospital Anxiety and Depression Scale.

** $p < .05$.

4. Discussion

In this study, we showed cross-sectional evidence that volume loss in the amygdala begins in HD before clinical diagnosis and occurs to a greater extent after diagnosis, and that this volume loss is related to motor, cognitive, and psychiatric signs. To the best of our knowledge,
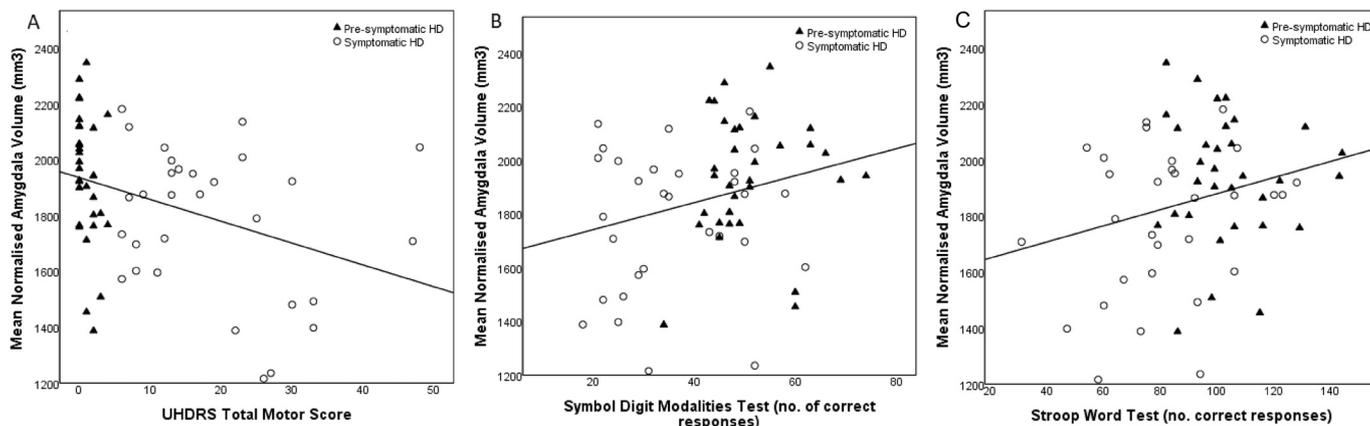
Fig. 2. UHDRS TMS (A), SDMT Scores (B), and Stroop Word Test (C) scores across total amygdala volumes for pre-symptomatic and symptomatic HD participants. Smaller amygdala volumes are associated with higher levels of motor signs ($p < .05$), and worse cognitive performance ($p < .05$).

Table 4
Inferential statistics for the relationship between amygdala volumes and clinical measures in pre-symptomatic HD participants divided into close and far from disease onset.

	B	SE	t	p	η^2	95% CI
Close to onset						
UHDRS TMS	−80.60	35.47	−2.27	.041**	0.28	[−157.22, −3.97]
SDMT	9.39	6.50	1.45	.17	0.13	[−4.55, 23.33]
Stroop Word Test	3.57	3.15	1.13	.28	0.079	[−3.15, 10.92]
UPSIT	−13.51	9.35	−1.45	.17	0.13	[−33.57, 6.55]
HADS Anxiety	−46.79	21.08	−2.22	.045**	0.28	[−92.33, −1.25]
HADS Depression	36.72	24.23	1.52	.15	0.15	[−15.61, 89.06]
Far from onset						
UHDRS TMS	−73.47	69.24	−1.06	.31	0.074	[−221.98, 75.03]
SDMT	−5.63	6.75	−0.83	.42	0.044	[−20.02, 8.76]
Stroop Word Test	−4.90	3.00	−1.63	.12	0.14	[−11.26, 1.47]
UPSIT	9.22	14.65	0.63	.54	0.026	[−22.01, 40.44]
HADS Anxiety	0.58	21.74	0.027	.98	0.00	[−46.04, 47.20]
HADS Depression	−1.78	23.02	−0.077	.94	0.00	[−51.16, 47.60]

Abbreviations. B = unstandardised regression coefficient. SE = standard error. CI = confidence interval. UHDRS TMS = Unified Huntington's Disease Rating Scale Total Motor Score. SDMT = Symbol Digit Modalities Test. UPSIT = University of Pennsylvania Smell Identification Test. HADS = Hospital Anxiety and Depression Scale.
** $p < .05$.

this is the first study in HD to directly compare amygdala volumetry between pre-symptomatic and symptomatic participants and demonstrate an association between amygdala atrophy and clinical symptomatology across the different disease stages. Our results reinforce the importance of extra-striatal structures in the clinical manifestation of HD.

A direct comparison of brain volume loss between pre-symptomatic and symptomatic HD is complex because of the expected differences in age and disease status. Nonetheless, our sub-analysis of an age-matched sample yielded the same pattern of results as the non-age matched groupings, suggesting that the findings of amygdala volume decrements in early HD cannot be explained by the older age of the group. Our findings clearly support the idea that HD exerts a detrimental effect on brain areas outside the striatum (cortical areas: Tabrizi et al., 2009; thalamus: Jernigan et al., 1991; and hippocampus: Brito et al., 2014), including the amygdala (Dogan et al., 2013; Douaud et al., 2006; Kipps et al., 2007), which begins early in the disease.

Our measures of amygdala volumes were not statistically associated with measures of disease reported in the IMAGE-HD study, including disease burden scores, and the size of the caudate nucleus and putamen (Dominguez et al., 2013; Georgiou-Karistianis et al., 2013; Dominguez et al., 2016). A lack of an association between amygdala volumes and measures of disease is somewhat surprising considering that striatal degeneration is thought to predict or strongly associate with many clinical and neurological signs of HD, including with deficits related to amygdala function. As an example is a recent functional MRI study that found an association between reduced anger-selective activation in the amygdala and smaller dorsal striatum volumes in pre-symptomatic HD (Van Den Stock et al., 2015). The lack of association observed in our study may be explained by the different rates of volume loss in amygdala versus striatal structures during the course of HD. The amygdala was approximately 7.6% smaller in symptomatic HD than pre-symptomatic HD, while the caudate and putamen were approximately 17.7% and 10.3% smaller in symptomatic HD than pre-symptomatic HD (see Fig. 4), as shown in the IMAGE-HD study (Dominguez et al., 2013; Georgiou-Karistianis et al., 2013; Dominguez et al., 2016). Furthermore, the different rates of volumes loss in the amygdala and striatal structures may also explain the lack of an association between amygdala volumes and disease burden scores, because disease burden scores are strongly linked to striatal pathology in HD (Penney et al., 1997).

In contrast, we did observe a relationship between the size of the amygdala and the degree of motor signs, in which smaller amygdala volumes were related to greater levels of motor signs. This relationship was dependent on proximity to disease onset: smaller amygdala volumes were related to worse motor signs in pre-symptomatic HD participants who were estimated to be close to disease onset, but not in those who were estimated to be far from disease onset. Atrophy in the striatum (Dominguez et al., 2013) and in primary motor and sensorimotor cortices (Dogan et al., 2013) has previously been shown to be associated with motor signs. IMAGE-HD is the first study to demonstrate an association between atrophy in the amygdala and motor signs.

In addition to the association with motor signs, we found a relationship between amygdala volumes and cognitive signs. In this relationship, smaller amygdala volumes were associated with worse visuomotor skills and slower processing speed, which are the two cognitive outcomes that demonstrate HD related changes most sensitively (Stout et al., 2012). Previously, other cognitive signs, specifically deficits in emotion recognition, were shown to be associated with amygdala volumetry in HD (Kipps et al., 2007). We also found a trend for associations with neuropsychiatric signs, with smaller amygdala volumes being associated with higher levels of anxiety and depressive symptoms. Assessing our pre-symptomatic HD group further, we found an association between smaller amygdala volumes and greater anxiety symptoms in pre-symptomatic HD participants who were nearing estimated disease onset. Previously, research has shown that psychiatric
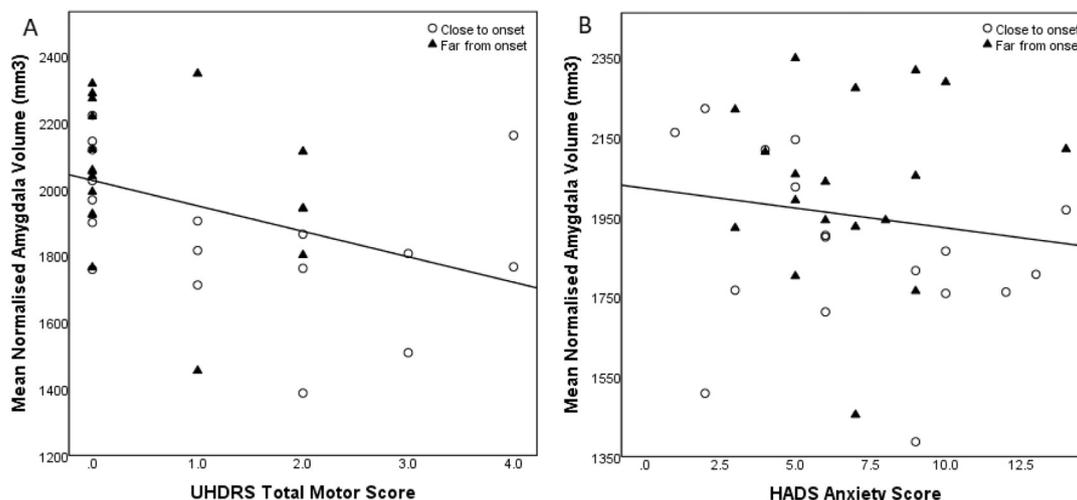
Fig. 3. UHDRS total motor scores (A) and HADS anxiety scores (B) across total amygdala volumes for pre-symptomatic HD participants close to disease onset and far from disease onset. For pre-symptomatic participants who were close to disease onset, smaller amygdala volumes were associated with higher levels of motor signs and increase in anxiety symptoms ($p < .05$).
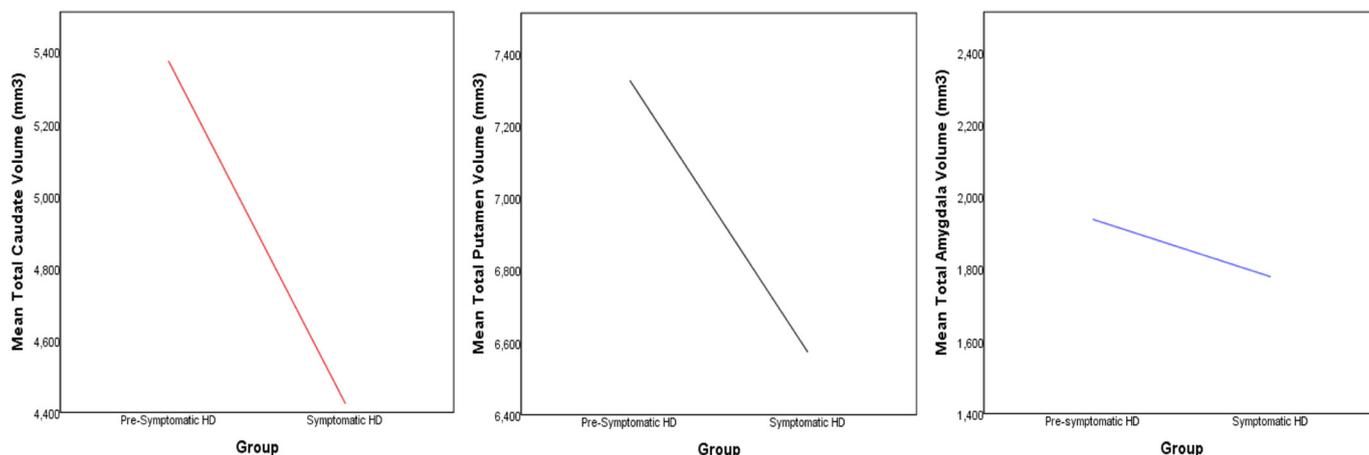

Fig. 4. Mean caudate, putamen, and amygdala volumes for pre-symptomatic and symptomatic HD participants. At a scale of $1000 \, mm^3$, the difference between pre-symptomatic and symptomatic HD participants in caudate and putamen volumes (decrease of 17.7% and 10.3%, respectively) is larger than the difference in amygdala volumes (decrease of 7.6%).

symptoms, particularly anxiety symptoms, begin subtly in the pre-symptomatic phase and continue to peak with worsening motor signs (Duff et al., 2007). Our findings extend on the current literature by showing that the amygdala is related to the increase in anxiety symptoms seen in the stage close to clinical diagnosis.

Amygdala volumes were not associated with odour recognition, despite known associations between the amygdala and olfactory processing (Ledoux, 2004; Rosenkranz and Grace, 2002; Schoenbaum et al., 1999). In our sample, pre-symptomatic and symptomatic HD participants scored similarly on the UPSIT and within a narrow range, so we would not have expected to see associations within our dataset (Georgiou-Karistianis et al., 2013).

5. Conclusions

In conclusion, this study provides findings that are important to understanding how abnormalities in extra-striatal structures, in addition to degeneration in striatal and cortical areas, may contribute to the manifestation of HD in the years before onset of clinical symptoms and across the disease. Furthermore, our work highlights the need for the brain in HD to be considered as a unified whole, rather than separate structures and areas targeted individually by HD.

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