The Value of GRE, ADC and Routine MRI in Distinguishing Parkinsonian Disorders

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ABSTRACT: ***Objectives:*** To study different radiological signs and sequences including apparent diffusion coefficient (ADC) and gradient echo (GRE) to differentiate degenerative parkinsonian syndromes. ***Background:*** Multiple system atrophy (MSA), Parkinson's disease (PD), progressive supranuclear palsy (PSP) and corticobasal degeneration (CBD) differ in the pattern of neurodegeneration and cellular damage. Measuring the ADC, GRE sequences for paramagnetic substances and simple anatomical assessments have been reported individually to assist in separating some of these disorders, but have not been compared. ***Methods:*** Brain MRIs from May 2002 to February 2008 were retrospectively evaluated by raters blinded to the clinical diagnosis for predefined MRI signs on T1, T2 and GRE sequences. ADC values were quantitatively measured. Medical records were objectively analyzed using standard clinical criteria for different parkinsonian syndromes. ***Results:*** 195 cases comprising of 61 PD, 15 MSA-P, 7 MSA-C, 21 PSP, 6 Corticobasal syndrome, 21 not fitting criteria and 64 controls were evaluated. 73% of patients with MSA-P had hypointensity of the putamen (compared to the pallidum) on GRE. The specificity of this sign to diagnose MSA-P was 90% versus PD and 76% versus PSP. When GRE hypointensity was combined with atrophy of the putamen the specificity improved to 98% (versus PD) and 95% (versus PSP) without altering the sensitivity. The ADC values were significantly higher in the middle cerebellar peduncle in cases with MSA-C versus controls, PD and PSP (p<0.001). ***Conclusions:*** The combination of hypointensity and atrophy of the putamen on GRE is useful in differentiating MSA-P from other parkinsonian syndromes.

RÉSUMÉ: La capacité de l'ÉG, du CDA et de l'IRM faits de routine à distinguer les différents troubles parkinsoniens. ***Objectif :*** Le but de l'étude était d'examiner les différents signes et les différentes séquences radiologiques, dont le coefficient de diffusion apparent (CDA) et échos de gradient (ÉG) pour différencier les syndromes parkinsoniens dégénératifs. ***Contexte :*** Les caractéristiques de neurodégénérescence et de dommage cellulaire sont différentes dans l'atrophie multisystémique (AMS), la maladie de Parkinson (MP), la paralysie supranucléaire progressive (PSP) et la dégénérescence cortico-basale (DCB). Des mesures du CDA, des séquences ÉG des substances paramagnétiques ainsi que des évaluations anatomiques simples ont été rapportées séparément pour aider à distinguer certaines de ces maladies, mais elles n'ont jamais été comparées entre elles. ***Méthode :*** Des évaluateurs ont revu rétrospectivement les IRM du cerveau effectuées entre mai 2002 et février 2008, sans connaître le diagnostic clinique posé chez les patients, afin d'identifier des signes prédéfinis à l'IRM en T1, en T2 et dans les séquences ÉG. Les valeurs du CDA ont été mesurées quantitativement. Les dossiers médicaux ont été analysés objectivement au moyen des critères cliniques standards des différents syndromes parkinsoniens. ***Résultats :*** Cent quatre-vingt-quinze cas dont 61 cas de MP, 15 cas d'AMS-P, 7 cas d'AMS-C, 21 cas de PSP, 6 cas de syndrome cortico-basal, 21 cas qui ne rencontraient pas les critères standards et 64 témoins ont été évalués. Soixante-treize pour cent des patients atteints d'AMS-P avaient de l'hypointensité au niveau du putamen par rapport au pallidum à l'ÉG. La spécificité de ce signe pour le diagnostic de l'AMS-P était de 90% par rapport à la MP et de 76% par rapport à la PSP. Quand l'hypointensité à l'ÉG était combinée à l'atrophie du putamen, la spécificité s'élevait à 98% par rapport à la MP et à 95% par rapport à la PSP, sans en modifier la sensibilité. Les valeurs du CDA étaient significativement plus élevées dans le pédoncule cérébelleux chez les cas d'AMS-C par rapport aux témoins et par rapport aux patients atteints de MP et de PSP (p < 0,001). ***Conclusion :*** La combinaison d'hypointensité et d'atrophie du putamen à l'ÉG est utile pour différencier l'AMS-P des autres syndromes parkinsoniens.

Multiple system atrophy (MSA), Parkinson's disease (PD), progressive supranuclear palsy (PSP) and corticobasal degeneration (CBD) can sometimes be clinically indistinguishable, especially early in the clinical course. However all differ in the pattern of neurodegeneration and cellular damage.[1-3] Volumetric studies are able to evaluate atrophy in specific areas of the brain.[4-7] However, these are time consuming and seldom used in clinical practice.

Over the years, based on the pattern of neuronal loss, various radiological signs have been proposed to differentiate parkinsonian disorders.[8-11] While some may be specific they lack sensitivity and others though sensitive are seen in patients with various parkinsonian syndromes. A magnetic resonance imaging

(MRI) rating scale for the diagnosis of the parkinsonian form of MSA (MSA-P) and PSP has been recently proposed, however the diagnostic sensitivity and specificity has not yet been assessed.[12]

Recently, gradient echo (GRE)[13,14] and diffusion weighted imaging (DWI)[15-21] have held a new promise for differentiating these disorders. Diffusion weighted imaging reflects the constrained movement of water molecules associated with myelinated fiber tracts in the normal brain (water molecules move freely along the tracts with restricted movement perpendicular to the fiber tracts), the so-called anisotropy of diffusion.[22] Pathological processes that modify tissue architectural integrity, including neuronal loss and secondary astrogliosis, remove some of the restrictions to water molecule movement, thereby increasing water diffusion and increasing the apparent diffusion coefficient (ADC). Thus, depending on the distribution of neurodegeneration, ADC measurements in different regions of the brain may help in differentiating the neurodegenerative disorders. Higher ADC values have been reported in the putamen in patients with MSA-P and in the middle cerebellar peduncle in patients with the cerebellar form of MSA (MSA-C) compared to patients with PD.[15,17,20,21,23] Deposition of paramagnetic substances like iron is associated with a reduction of the $T2^*$ relaxation time producing a hypointense signal on GRE ($T2^*$) imaging.[13,14,24] Histopathological studies in patients with MSA demonstrate iron deposition in the putamen.[25] Recently, T2 hypointensity in the putamen has been shown to correlate with ferritin deposition in the putamen on post-mortem studies.[26] Hypointensity of the putamen (compared to the pallidum) on GRE imaging has been demonstrated in patients with MSA-P versus those with PD.[13,14] However, there has been a paucity of studies comparing GRE imaging in patients with MSA-P versus PD and other parkinsonian syndromes. Gradient echo and DWI have never been compared simultaneously in different parkinsonian syndromes.

We evaluated the sensitivity and specificity of a variety of radiological signs in differentiating parkinsonian syndromes using conventional MRI, GRE and DWI sequences in patients with MSA, PSP and corticobasal syndrome (CBS) compared to patients with PD and controls.

METHODS

Patient selection

Patients attending the Movement Disorders Clinic at the Toronto Western Hospital and classified as having parkinsonism, PD, MSA, PSP, or CBS/CBD who had undergone MRI of the brain at the University Health Network (UHN) hospitals in Toronto, Canada from May 2002 to February 2008 were included in the study. GRE has been part of the standard MR protocol at UHN since May 2002. Controls had a normal MRI of the brain during investigations for other neurological indications over the same period.

Charts of the patients were reviewed (by JR and PW) and clinical data was extracted from clinic visits closest to the date of the MRI. In cases where more than one MRI scan was available, the first and last MRI were included for analysis and corresponding visit notes were analyzed. Standard diagnostic criteria were applied (Appendix).[27-31] Some of the diagnostic criteria were modified at the outset to include patients with earlier presentations of the disease. In addition to the previously described criteria, we included patients with slowing of downward saccades, abnormalities of vertical OKN and the presence of postural instability within the first year for the diagnosis of PSP without mandating the presence of falls.[28,29] As recommended by the Movement Disorder Society Scientific Issues Committee committee[29] we added a set of less rigid criteria in the diagnostic profile of MSA.[30] In the diagnostic criteria for CBS we dropped dementia from the exclusion criteria, recognising that patients with a clinical presentation of dementia are often found to have corticobasal degeneration on histopathology.[27] For the diagnosis of PD we used the UKPDS Brain Bank criteria, removing the presence of a positive family history from the exclusion criteria.[31] The clinical data was entered into an Excel database that automatically computed the clinical criteria, determining if the diagnosis was fulfilled for any of the parkinsonian syndromes. The expert diagnosis by the movement disorder specialist at the time of the clinic visit was also recorded. In patients with more than one MRI scan the clinical diagnosis at the visit corresponding to the last visit was recorded as the final diagnosis. Patients not fitting the clinical diagnostic criteria for any of the four parkinsonian syndromes were labelled as undiagnosed for the purpose of the study.

MRI protocol

Imaging was performed on 1.5T or 3T MRI systems (HD platforms GE Healthcare Milwaukee) using eight channel head coils following a uniform MRI protocol (supplementary data).

Image analysis

We excluded patients with major deviations in the protocol where more than one sequence was missing including the GRE sequence (typically when T_2 axial + GRE or ADC + GRE were missing). We included patients where one sequence was missing, but excluded them from any analysis that required the missing sequence. Peri-operative MRI's (e.g., from deep brain stimulation in PD) were excluded from the analysis to prevent unblinding of the raters. While we included patients with both 1.5T and 3T MRI scans who fit criteria, there were only four patients with 3T MRIs who fulfilled criteria, (2 PSP, 1 PD and 1 MSA-P).

Two blinded independent raters (PH and MQ) evaluated the MRI scans for all qualitative data (conventional T_1 and T_2 sequence signs (see below) and GRE hypointensity), using standard templates for each sign using examples published in the literature. The raters were blinded to the clinical data of patients and controls. Inter- and intra-rater reliability was also assessed for each of the signs randomly in a select number of patients (inter-rater: 31 scans, intra-rater: 14 scans). Pontine antero-posterior (A-P) diameter measurements and ADC measurements were performed by one unblinded rater (PW).

Putamen atrophy, slit like hyperintensity and the presence of a hot cross bun sign were rated as present unequivocally or not on T_2 axial sequences compared to standard templates. Hypointensity of the putamen on T_2 axial sequences was defined as intensity of the putamen equal to or lower than the intensity of the pallidum excluding regional signal loss in the posterior and lateral parts of the putamen; with or without a band of

hyperintensity within the putamen.[32,33] The presence of a "Hummingbird sign" was accepted if present unequivocally on T_1 mid-sagittal images.[34] The "morning glory sign" was rated as present based on clear evidence of concavity of the lateral margin of the midbrain tegmentum as reported previously.[8] Hyperintensity of the middle cerebellar peduncles (MCP) was accepted if unequivocally present on T_2 axial sequences at the thickest point along a line perpendicular to the MCP at the level of the internal auditory meatus or facial nerve.[15] Pontine A-P diameter was measured on T_1 mid-sagittal images by measuring the largest A-P diameter of the pons perpendicular to the fourth ventricle.[35]

Gradient echo scans were rated using criteria described previously.[13] We considered the sign to be present when the putamen was hypointense compared to the pallidum (grades 2, and 3)[13]. Subsequently those with GRE hypointensity were blindly rated post-hoc (by DJM, AEL) to evaluate the presence of both atrophy (on T_2 and GRE) and GRE hypointensity.

For the measurement of ADC values, elliptical regions of interest (ROIs) were manually placed bilaterally in the MCPs, putamen and lateral ventricles using the axial T_2-weighted images as guidance. The MCPs were measured at their thickest point along the line perpendicular to the MCP at the level of the internal auditory meatus and facial nerve.[15] The putamen was measured with elliptical ROIs, which were completely within the putamen (supplementary Figure 1).

The study was approved by the research ethics board at the University Health Network. Comparisons across the study groups were performed using a series of chi-square tests, GLM tests, and t-tests unless otherwise specified. Post hoc comparisons were made using the Tukey's test for quantitative data and Bonferonni adjusted pairwise comparisons for categorical data. Sensitivity and specificity for regional ADC (rADC) and pontine A-P diameter measurements was calculated using optimal cut-off values determined by receiver operating characteristics (ROC) curve analysis. For inter-rater and intra-rater reliability analysis a kappa value of 0.4-.59 suggested moderate agreement, 0.6-0.79 substantial agreement and 0.8-1.0 outstanding agreement.[36]

RESULTS

A total of 131 patients with parkinsonism scanned from May 2002 to February 2008 and 64 controls (see supplementary data for distribution) were included for the purpose of the study (Table 1). In patients with more than one MRI we included the MRI at the last visit and the corresponding clinical assessment for the analysis.

Due to the heterogeneous nature of the population with parkinsonism it was not possible to have controls age and sex matched for all diseases. Our controls, like the patients with MSA were younger and more likely to be female, compared to an older predominantly male population with PSP and PD. As expected, patients with PD had the longest disease duration and lower Hoehn and Yahr scores compared to those with atypical parkinsonian syndromes (Table 1).

Conventional T_1 and T_2 signs

Among the signs on T2-weighted MRI studying structural changes in the basal ganglia (Supplementary Table 1, Figure 1, a-d), atrophy of the putamen was seen more commonly in patients with MSA-P and PSP compared to patients with PD and controls. Putamen hypointensity was more common in patients with MSA-P compared to controls and patients with PD and PSP. Slit-like hyperintensity in the putamen was significantly more common in patients with MSA-P compared to those with PSP, PD and controls. Atrophy of the putamen alone lacked specificity for the diagnosis of MSA-P (Table 2). Putamen

Table 1: Demographic information and disease severity

Variable	CBS	MSA-C	MSA-P	PD	PSP	Undiagnosed	Controls	Total	Significance
Total N	6 (3.1%)	7[a] (3.6%)	15 (7.7%)	61 (31.3%)	21 (10.8%)	21 (10.8%)	64 (32.8%)	195	
Mean age at MRI	69.2 +/- 7.9	61.4 +/- 7.6	65.5 +/- 12.4	65.8 +/- 10.6	68 +/- 8.4	69.7 +/- 7.8	57.3 +/- 11.3		p < 0.001
Males n (%)	3 (50)	2 (28.6)	8 (53.3)	41 (67.2)	17 (81)	13 (61.9)	23 (35.9)	107 (54.9)	p = 0.001
Mean Disease duration (mths)	48.8 +/- 40.7	36.3 +/- 22.0	46.3 +/- 17.0	84.3 +/- 63.8	55.4 +/- 50.2	54.55 +/- 38.3	N/A	63.3 +/- (52.7)	p=0.012
Median disease duration (mths)	37	31	45	70	39	54	N/A		
Mean H & Y score	3.6 +/-1.7 (N=4)	4.0 +/-0.7 (N=5)	3.5 +/-1.0 (N=15)	2.6 +/-0.9 (N=57)	3.7 +/-0.9 (N=19)	2.6 +/-1.0 (N=15)	N/A	(N=115)	p < 0.001

CBS = Corticobasal syndrome, MSA-C = Multiple system atrophy cerebellar type, MSA-P = Multiple system atrophy parkinsonian type, PD = Parkinson's disease, PSP = Progressive supranuclear palsy, H & Y = Hoehn and Yahr. Controls were significantly younger than patients with PSP (p=0.001) and PD (p<0.0001). Controls were more likely to be female as compared to patients with PSP (p=0.003) and PD (p=0.0005). Patients with PD had lower H & Y scores compared to patients with MSA-P (p=0.0004), MSA-C (p=0.0034) and PSP (p<0.0001). [a.] 5/7 had additional parkinsonism,1/7 had only bradykinesia, but none of the other features, 1/7 had pure cerebellar ataxia with no parkinsonian signs.

Table 2: Sensitivity and specificity of different radiological signs

Sign	Disease for which sensitivity assessed	Sensitivity	Specificity compared to PD	Specificity compared to PSP	Specificity compared to controls	Specificity compared to MSA
Atrophy of putamen on T_2	MSA-P	**73.3**	73.8	47.6	93.8	NA
T_2 Hypointensity of Putamen	MSA-P	**40.0**	90.2	90.5	96.9	NA
T_2 Slit-like hyperintensity of putamen	MSA-P	**60.0**	93.4	85.7	95.3	NA
GRE Hypointensity of putamen	MSA-P	**73.3**	90.0	76.2	93.7	NA
GRE hypointensity + atrophy (GRE and T_2) of putamen	MSA-P	**73.3**	98.3	95.2	100	NA
rADC value of putamen $\geq 0.840 \times 10^{-3} mm^2/sec$[a]	MSA-P	**33.3**	73.7	61.9	87.5	NA
Hot cross bun sign	MSA-C	**71.4**	100	90.5	100	NA
Hyperintensity of MCP	MSA-C	**42.9**	100	100	100	NA
Pontine AP diameter below 21 mm[a]	MSA-C	**71.4**	98.4	90.5	98.4	NA
rADC value of MCP $\geq 0.940 \times 10^{-3} mm^2/sec$[a]	MSA-C	**66.7**	91.4	100	87.5	NA
Hummingbird sign	PSP	**47.6**	91.8	NA	100	86.4
Morning glory sign	PSP	**9.5**	100	NA	100	100.0

rADC= regional apparent diffusion coefficient, GRE= Gradient echo imaging, MSA-C = Multiple system atrophy cerebellar type, MSA-P = Multiple system atrophy parkinsonian type, PSP = Progressive supranuclear palsy, MCP= Middle cerebellar peduncle. [a] Cut off values calculated using ROC curves to try and achieve the best specificity and sensitivity for that value.

hypointensity on T_2 sequences on the other hand was specific, but lacked sensitivity in detecting MSA-P. Slit-like hyperintensity was seen in 60% of the patients with MSA-P with a good specificity (Table 2).

We assessed two signs of changes in midbrain anatomy (Supplementary Table 1, Figure 1, e-h), namely the presence of the "hummingbird sign"[34] and concavity of the midbrain tegmentum, the "morning glory sign"[8]. The Hummingbird sign was more common in patients with PSP as compared to controls and those with PD, but not MSA-P. The morning glory sign was very rare; only two patients with PSP and one patient who did not fit criteria for PSP at the time of assessment (but had a clinical syndrome compatible with PSP) had concavity of the midbrain tegmentum. While both signs seem highly specific for the diagnosis of PSP, they both lack sensitivity with hummingbird sign present in less than 50% and the morning glory sign in less than 10% of the clinically diagnosed cases of PSP (Table 2).

Among signs assessing pontine and cerebellar anatomy, the presence of the hot cross bun sign and hyperintensity of the MCP were significantly more common in patients with MSA-C compared to controls, patients with PD and PSP. The mean pontine A-P diameter was significantly lower in patients with MSA-C compared to controls and patients with PD, PSP and CBS (Supplementary Table 1, Figure 1 k-l). Both the hot cross bun sign and a low pontine A-P diameter (Figure 1 i-j) were sensitive for the diagnosis of MSA-C (Table 2). While these signs were specific when compared to patients with other parkinsonian syndromes, we did not have patients with other degenerative or genetic ataxias for comparison making interpretation of specificity difficult.

At least moderate agreement was obtained in all signs among the two raters and also within the same rater (Supplementary Table 2). Inter-rater and intra-rater agreement was less for signs involving putamen anatomy as compared to signs like the hummingbird sign and hot cross bun sign where it is easier to interpret if the sign is present or absent.

GRE scans

As seen in Table 3, 73.3% of the patients with MSA-P had hypointensity of the putamen on GRE scans compared to the pallidum (MSA-P versus controls p < 0.0001, MSA-P versus PD p<0.0001, MSA-P vs PSP p=0.042). A post-hoc blinded analysis (by two further reviewers, DJM, AEL) was conducted in all cases with hypointensity of the putamen (compared to the pallidum) rating putamenal atrophy visually using both GRE and T_2-weighted axial sequences (Figure 2, Table 3 and brief case reports in Supplementary Information). The combination of putamenal atrophy and GRE hypointensity was significantly more common in patients with MSA-P compared to PD (p<0.0001), PSP (p=0.0006), and controls (p<0.0001). When atrophy and hypointensity were combined, the specificity of GRE images in diagnosing MSA-P was significantly higher than with using hypointensity alone (Table 2).
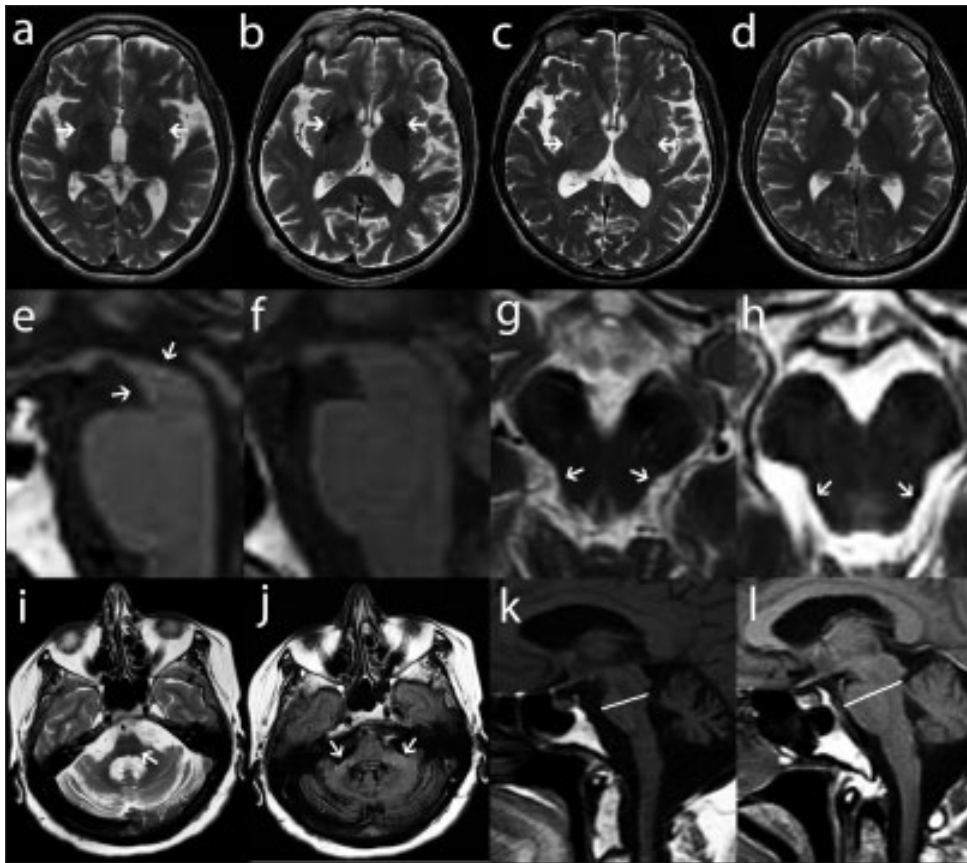
Figure 1: MR Images demonstrating different signs on structural imaging. Images a-d: T2-weighted axial MRI images of the basal ganglia a. Atrophy of the putamen (also see slit hyperintensity), b. Hypointensity of the putamen c. Slit-like hyperintensity of the putamen d. Control MRI (normal). Images e,f: T1-weighted sagittal images showing the hummingbird sign in a patient with PSP (e) compared to a normal control (f). Images g, h: T2-weighted axial images of the midbrain showing tegmental concavity (the morning glory sign) in a patient with PSP (g) compared to convexity in a patient with PD (h). Images i, T2-weighted and j, FLAIR axial MRI images of patients with MSA-C of the pontocerebellar region, showing a hot cross bun sign (i) and hyperintensity of the middle cerebellar peduncle (j). Images k, l: T1-weighted sagittal images showing pontine atrophy in a patient with MSA-C (k) compared to a normal pons in a patient with PD (l).

Antero-posterior gradient of hypointensity in the putamen was seen commonly among patients with both MSA-P and PSP and to a lesser extent among other groups, significantly reducing the specificity of this sign (Table 3).

ADC maps

Comparing rADC (regional ADC) values among the regions of interest (Table 4), patients with MSA-C had significantly higher rADC values in the MCP compared to patients with PD ($p=0.013$), PSP ($p=0.016$) and controls ($p=0.014$). However, rADC values in the putamen in patients with MSA-P were no different from those in patients with PD, PSP or CBS (Table 4).

We plotted ROC curves of the rADC values assigning cut off values for the MCP ($0.940 \times 10^{-3}mm^2/sec$) and the putamen ($0.840 \times 10^{-3}mm^2/sec$); using these, rADC assessment in the MCP was specific for the diagnosis of MSA-C compared to the other parkinsonian groups, but lacked sensitivity (Table 2).

Table 3: Comparison of the intensity on gradient echo imaging

Characteristic	CBS	Control	MSA-C	MSA-P	PD	PSP	Undiagnosed	Significance
Total No of cases	6	64	7	15	60[a]	21	21	
Hypointensity of the putamen on GRE (compared to pallidum)	1 (16.7)	4 (6.3)	1 (14.3)	**11 (73.3)**	6 (10.0)	**5 (23.8)**	5 (23.8)	p< 0.0001
Putamen atrophy (GRE and T_2) + hypointensity on GRE (compared to pallidum)	0	0	1 (14.3)	**11 (73.3)**	1 (1.7)	1 (4.8)	0	p< 0.0001
Presence of a rostro caudal gradient of hypointensity (GRE) in the putamen	2 (33.3)	17 (26.6)	3 (42.9)	**13 (86.7)**	20 (32.8)	**14 (66.7)**	13 (61.9)	p=0.0005

GRE = gradient echo sequence, CBS = Corticobasal syndrome, MSA-C = Multiple system atrophy cerebellar type, MSA-P = Multiple system atrophy parkinsonian type, PD = Parkinson's disease, PSP = Progressive supranuclear palsy. [a]1 patient with PD did not have a gradient echo sequence (denominator for PD = 60). Numbers in parentheses are percentages.

Table 4: Comparison of ADC values in the putamen, middle cerebral peduncle and CSF

rADC value[a]	CBS	Controls	MSA-C	MSA-P	PD	PSP	Undiagnosed	Significance
N[b]	5	64	6	15	57	21	21	
MCP (right)	0.86 +/- 0.16	0.75 +/- 0.13	0.95 +/- 0.10	0.83 +/- 0.12	0.72 +/- 0.13	0.71 +/- 0.10	0.70 +/- 0.10	<0.0001
MCP (left)	0.84 +/- 0.13	0.75 +/- 0.15	0.90 +/- 0.13	0.78 +/- 0.12	0.71 +/- 0.12	0.72 +/- 0.10	0.74 +/- 0.09	0.0054
MCP (mean)	0.85 +/- 0.15	0.75 +/- 0.13	0.93 +/- 0.11	0.80- +/- 0.10	0.69 +/- 0.17	0.71 +/- 0.09	0.72 +/- 0.08	0.0339
Putamen (right)	0.86 +/- 0.13	0.72 +/- 0.10	0.73 +/- 0.09	0.81 +/- 0.15	0.80 +/- 0.09	0.80 +/- 0.09	0.79 +/- 0.14	0.0069
Putamen (left)	0.87 +/- 0.12	0.75 +/- 0.09	0.80 +/- 0.14	0.88 +/- 0.20	0.84 +/- 0.16	0.84 +/- 0.09	0.81 +/- 0.13	0.0023
Putamen (mean)	0.86 +/- 0.11	0.73 +/- 0.09	0.76 +/- 0.13	0.85 +/- 0.17	0.79 +/- 0.215	0.82 +/- 0.08	0.80 +/- 0.12	0.0184
CSF (right)	3.51 +/- 0.27	3.35 +/- 0.34	3.41 +/- 0.33	3.37 +/- 0.29	3.41 +/- 0.28	3.36 +/- 0.26	3.03 +/- 0.93	0.0336
CSF (left)	3.65 +/- 0.21	3.49 +/- 0.29	3.54 +/-0 .37	3.40 +/- 0.28	3.44 +/- 0.26	3.51 +/- 0.32	3.47 +/- 0.28	0.6550

[a] rADC values (x 10^{-3} mm^2/sec) are reported as mean +/- standard deviation. [b] Not all patients had ADC sequences. Denominators reflect only those with ADC sequences. rADC= regional apparent diffusion coefficient, CBS = Corticobasal syndrome, MSA-C = Multiple system atrophy cerebellar type, MSA-P = Multiple system atrophy parkinsonian type, PD = Parkinson's disease, PSP = Progressive supranuclear palsy, MCP= Middle cerebellar peduncle, CSF = cerebrospinal fluid.

Predictive role of imaging in early and undiagnosed cases

Four of six cases fulfilling the criteria of MSA-P at the final visit but not at the first clinical visit (mean duration between visits was 20.5 months) had GRE hypointensity and atrophy of the putamen at the time when the clinical criteria for MSA-P were not met. The remaining two patients not showing GRE hypointensity at the first visit continued to have normal scans 20 months later. On the other hand only one of four cases having serial imaging and fulfilling criteria for MSA-P at all time points showed GRE hypointensity and atrophy on the second MRI scan not present in the first.

There were 21 cases in our study that did not fulfill our diagnostic criteria for the parkinsonian syndromes at their last visit (Supplementary Table 3). Four cases showed MRI signs that are seen in patients with MSA. All had a clinical syndrome which could be compatible with MSA, though they did not fulfill the prespecified clinical criteria. Two of the patients with postural instability and gait problems showed MRI signs seen in PSP, both had a clinical picture suggestive of PSP (supplementary Table 3).

DISCUSSION

Standard T1/T2 MRI signs

Several studies have evaluated different signs on standard T$_2$-weighted MRI. Among signs evaluating patients with MSA-P, hypointenstiy of the putamen or a linear slit like hyperintensity of the putamen have been most commonly used.[10,11,32,33,37-41]

Putamen atrophy has been evaluated accurately using volumetric MRI, however this is extremely time consuming and difficult to put into clinical practice.[7] Studies evaluating the presence of putamen atrophy in MSA based on a visual rating have found the prevalence to be highly variable from 31-100%.[11,18,33,39,42] Using templates from previous studies, we found a lack of specificity for the diagnosis of MSA-P, especially when compared to PSP (Table 2).[11]

Hypointensity of the putamen on T$_2$-weighted imaging has been reported in 40-90% of MSA cases depending on patient selection (MSA-P alone or all cases with MSA) and the definition used (eg. signal intensity compared to pallidum).[10,11,32,33,37-41] Most of these studies reported relatively good specificity when comparing MSA to PD. The few studies that included patients with PSP, showed a lowered specificity when compared to PSP, contrary to our study. It is possible that the use of templates of putamen hypointensity on T$_2$-weighted MRI, as in our study could have been responsible in improving the specificity in diagnosing MSA-P, but at the same time having a lower sensitivity.

Linear slit-like hyperintensity of the putamen was found in 60% of our MSA cases compared to 26-83% in the literature.[10,11,14,18,32,33,38,39,43,44] In our study, largely using 1.5 T scans (only 4/195 had 3T), this sign had good specificity compared to both PD and MSA (Table 2). While this sign may be useful as a diagnostic tool for MRI's done on 1.5T MRI machines, we have often seen a long linear rim on the lateral margin of the putamen on T2-weighted scans done on a 3T scanner in patients with various diagnoses and a study of healthy adults showed significant false positive hyperintense putamen rims on 3T compared to 1.5T MRI scans.[45] Recently an increase in the MRI field strength resulted in a decrease in the occurrence of hyperintensity both at the dorsolateral outer putaminal margin and an increase in the occurrence of hypointensity at the dorsolateral putaminal margin in patients with MSA.[46]

The hot-cross bun sign was present in 71% of patients with MSA-C in our study compared to 50-80% of the patients with MSA-C, or MSA in the literature.[39,44,47] It has been proposed that this sign is due to selective loss of myelinated transverse pontocerebellar fibers and neurons in the pontine raphe with preservation of the pontine tegmentum and corticospinal tracts[48] as well as gliosis of the middle part of the pontine reticular formation, and the pontocerebellar fibers.[49] While there was a good specificity for this sign in our study population, we did not
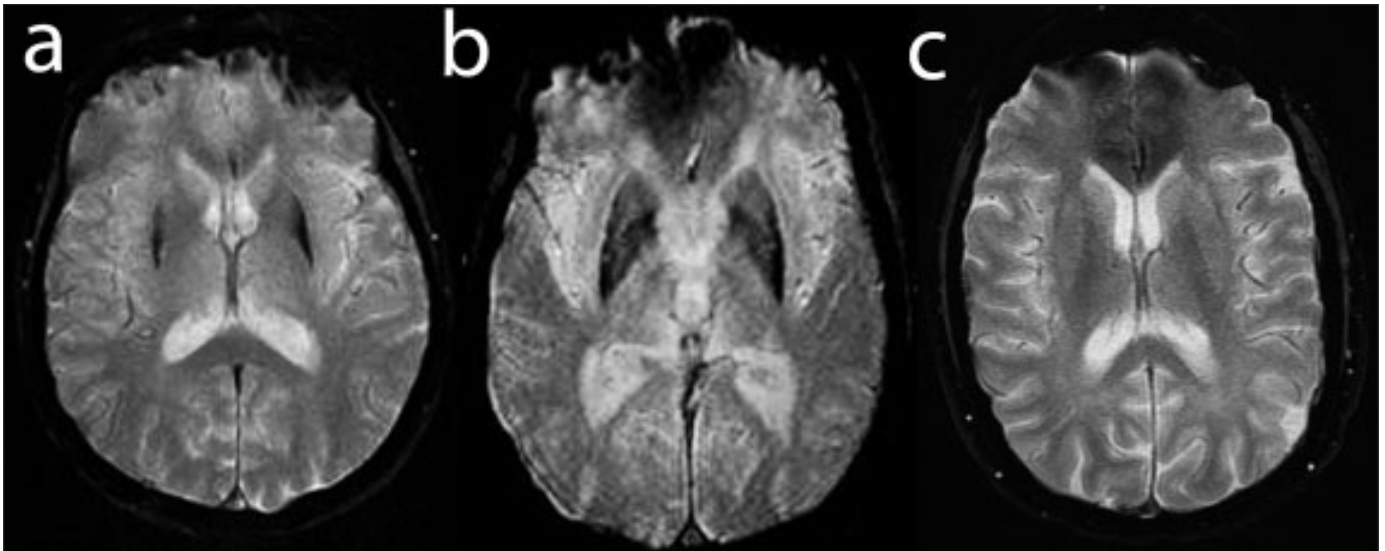
Figure 2: Gradient Echo Imaging in parkinsonism. A patient with MSA-P showing both atrophy and hypointensity of the putamen bilaterally (a), a patient with PSP showing hypointensity of the putamen without atrophy (b), a patient with PD with a normal putamen on the GRE sequence (c).

include patients with spinocerebellar ataxias where the hot cross bun sign has also been reported.[50,51]

We found hyperintensity of the MCP in 43% of the patients with MSA-C; this has been reported in 21-50% of patients with MSA,[33,38,44] higher in studies looking only at MSA-C.[38] While the specificity of this sign was found to be good when compared to patients with parkinsonism in our study, this sign has been reported in 14% of MRIs done for tinnitus or hearing loss[52] and it has also been reported in spinocerebellar ataxias, adrenoleukodystrophy, HIV encephalopathy and Fragile X tremor ataxia syndrome.[53,54]

Pontine A-P diameter[35,55] below 21mm had a good sensitivity and specificity for MSA-C but did not differentiate other forms of parkinsonism.

The hummingbird sign was less sensitive[56] and the morning glory sign[8] was distinctly uncommon, suggesting that it is a late sign that is of little use in the early differentiation of parkinsonian disorders. However both were specific for the diagnosis of PSP.

GRE signs

The sensitivity and specificity profile of GRE sequences in differentiating MSA from PD was similar to previous reports (35-69% and 88-89%).[13,14,57,58] It has been suggested that the specificity for MSA could be improved if this sign were combined with a slit like T_2 weighted hyperintensity of the putamen.[14] Two studies evaluating putaminal hypointensity using GRE or susceptibility-weighted imaging (SWI) sequences in MSA-P included patients with PSP in their study.[59,60] As in these studies, we found hypointensity of the putamen in some of the patients with PSP, significantly reducing the specificity of the sign (Table 2, 3). However, we were impressed by the differences in the morphology with atrophy of the putamen in MSA-P (Figure 2) compared to those in PSP. Blinded post-hoc

analysis confirmed the presence of the combination of putamenal atrophy (T_2 and GRE) and GRE hypointensity (Table 3) with moderately good sensitivity (73%) and excellent specificity for the diagnosis of MSA-P compared with PD (specificity 98.3%), PSP (specificity 95.2%) and controls (specificity 100%).

This sign was also present in four out of six patients with MSA-P, prior to fulfilling clinical criteria, indicating that it may be a useful sign for the diagnosis of MSA-P in the early stages of the disease.

ADC maps

Most studies measuring rADC values in the putamen found a higher ADC value in patients with MSA-P as compared to PD and controls.[16,18,20,23,43,61] When rADC values of the putamen of MSA-P and PSP were compared the results have been mixed; showing no significant difference,[17,23] or a lower specificity.[16] As in at least one previous study,[17] we did not find higher rADC values in the putamen in patients with MSA-P. Methodological differences in slice thickness (5mm compared to 2mm in other studies) and ROI measurement (we used only a part of the putamen that included the anterior portion for analysis compared to the entire putamen or exclusively the posterior portion) make it difficult to compare the results of our retrospective evaluation with other prospective studies.

However, despite these methodological differences, we were able to reproduce the higher ADC values in the MCP in patients with MSA-C as compared to other parkinsonian syndromes reported earlier[16,17,61,62] While this sign is useful when comparing patients with other parkinsonian syndromes, we did not evaluate patients with other cerebellar disorders. Higher ADC values have been described in patients with DRPLA in all cerebellar peduncles, but not in patients with late cerebellar cortical atrophy.[63]

Limitations of the study

Our study suffers from certain limitations. Though we tried to match the controls by age and sex it was not possible to do so for all the diseases due to the heterogeneous nature of the disorders. Most of the signs in our study however, pertained to patients with MSA where the controls were age and sex matched. Due to the retrospective nature of our study we were unable to use high resolution multidirectional DWI protocols applied in prospective trials studying diffusivity in patients in parkinsonian syndromes.[18,20,23] The methods of measuring ROI's were also different. These differences make it more difficult to compare our results with those from other centers, but show that DWI using routine screening MRI protocols did not adequately differentiate MSA from other disorders. Most of the scans were acquired on 1.5T MRI scanners. A prospective study using three Tesla scans would have provided a better assessment of imaging capabilities now available in most neurosciences centers. However, our study does provide useful and practical data for scans still utilized in many neurological practices. Newer comparative analyses (e.g., midbrain to pontine diameters[64], diffusion tensor imaging[65] also need to be evaluated in similar large populations to assess their sensitivity and specificity in the diagnosis of various parkinsonian disorders. We used established clinical criteria. However in the absence of pathological confirmation, misdiagnosis in some of these cases cannot be excluded.

CONCLUSIONS

Hypointensity of the putamen on GRE is extremely valuable especially when combined with atrophy of the putamen in differentiating MSA-P from other forms of parkinsonism. This sign is quick, easy to interpret and can be assessed retrospectively on routine MRI studies.

Our ADC sequence evaluation failed to show useful changes in the striatum in MSA-P. Perhaps not surprisingly, ADC and T_2-weighted abnormalities in posterior fossa structures found in MSA-C differentiated this disorder from other forms of parkinsonism but further studies need to compare this to other degenerative and genetic cerebellar disorders. The hummingbird and morning glory signs, though not sensitive, were fairly specific for the presence of PSP.

ACKNOWLEDGMENT

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Appendix: Clinical Criteria for diagnosis of Parkinsonian Syndromes

For the purpose of this study we have used the most commonly used diagnostic criteria or suggested modifications of the same in each of the disorders. We have however modified the criteria in some cases at the beginning of the study so as to include more recent knowledge about the disorders and also with a hope to be able to classify more of the cases which would have been excluded by some of the more stringent criteria.

A} PSP - NINDS Clinical research criteria[28] and SIC modifications[29]

Mandatory Inclusion Criteria
a. Gradually progressive disorder
b. Age at onset at 40 or later
c. For…..
   Possible (clinically probable)[29] - either vertical supranuclear palsy or both slowing of vertical saccades & postural instability with falls < 1 yr disease onset
   Probable (clinically definite)[29] - Vertical supranuclear palsy and prominent postural instability with falls within first year of disease onset
   Definite - All criteria for possible or probable PSP are met and histopathologic confirmation at autopsy

Exclusion Criteria - For possible and probable:
   1. Recent history of encephalitis;
   2. Alien limb syndrome;
   3. Cortical sensory deficits;
   4. Focal frontal or temporoparietal atrophy;
   5. Hallucinations or delusions unrelated to dopaminergic therapy;
   6. Cortical dementia of Alzheimer type;
   7. Prominent, early cerebellar symptoms or unexplained dysautonomia;
   8. or evidence of other diseases that could explain the clinical features

Supportive criteria
   1. Symmetric akinesia or rigidity, proximal more than distal
   2. Abnormal neck posture, especially retrocollis;
   3. Poor or absent response of parkinsonism to levodopa;
   4. Early dysphagia & dysarthria;
   5. Early onset of cognitive impairment including > 2 of: apathy, impairment in abstract thought, decreased verbal fluency, utilization or imitation behavior, or frontal release signs

Clinical criteria[29]
   1. Clinically Definite as the old probable
   2. Clinically Probable as the old possible
   3. New Category of Clinically Possible PSP as
         a. Gradually progressive disorder
         b. Onset over 40 years of age
         c. Clear slowing of vertical downward saccades and marked abnormalities of vertical OKN#
         d. Postural instability/ disequilibrium within the first year (falls are unnecessary)
         e. No exclusion criteria

#We have made one change to the SIC recommendations adding slowing of downward saccades and abnormalities of vertical OKN with the presence of postural instability within the first year without mandating the presence of falls for the diagnosis of PSP as previously required in the SIC modifications.

B} MSA - Consensus criteria for the diagnosis of MSA[30]
Section ONE – features and criteria

   I. Autonomic and urinary dysfunction

A. *Autonomic and urinary features*.
   Orthostatic hypotension (by 20 mm Hg systolic or 10 mm Hg diastolic); urinary incontinence or incomplete bladder emptying
B. *Criterion for autonomic failure or urinary dysfunction in MSA*.
   Orthostatic fall in blood pressure (by 30 mm Hg systolic or 15 mm Hg diastolic) and/or urinary incontinence (persistent, involuntary partial or total bladder emptying, accompanied by erectile dysfunction in men)

   II. Parkinsonism;

A. *Parkinsonian features*
   1) Bradykinesia (slowness of voluntary movement with progressive reduction in speed and amplitude during reciprocal actions)
   2) Rigidity
   3) Postural instability (not caused by primary visual, vestibular, cerebellar or proprioceptive dysfunction
   4) Tremor (postural, resting or both)

B. Criteria for Parkinsonism in MSA
   Bradykinesia plus at least one of items of 2-4

   III. Cerebellar dysfunction;

A. Cerebellar features
   1) Gait ataxia;
   2) Ataxic dysarthria;
   3) Limb ataxia;
   4) Sustained gaze-evoked nystagmus

B. Criterion for Cerebellar dysfunction
   Gait ataxia plus at least one of items of 2-4

   IV. Corticospinal tract dysfunction

A. Corticospinal tract features
   Extensor plantar responses + Hyperreflexia;
B. No corticospinal tract features are used in defining the diagnosis of MSA

   V. Set of softer features[#]

   a) REM sleep behavior disorder
   b) Stidor
   c) Myocolonus
   d) Severly hypophonic speech
   e) Marked antecollis (head drop)

Section TWO: Diagnosis of probable and possible MSA

Possible MAJOR: One criterion plus two features from separate other domains. When the criterion is Parkinsonism, a poor levodopa response qualifies as one feature (hence, only one additional feature is required)

Possible MINOR: Same as the above – however ≥ 2 softer features may be substituted as one feature in the above criteria

Probable A: One criterion for autonomic failure/urinary dysfunction plus poorly levodopa responsive Parkinsonism or cerebellar dysfunction

Probable a: autonomic feature + Parkinsonism criteria + cerebellar feature + ≥ 2 softer features

Definite: Pathologically confirmed by the presence of a high density of glial cytoplasmic inclusions in association with a combination of degenerative changes in the nigrostriatal and olivopontocerebellar pathways

Section THREE: Exclusion criteria
For possible and probable:

History
   1. Symptomatic onset < 30 yr of age
   2. Family history of a similar disorder;
   3. Systemic diseases or other identifiable causes for features listed above
   4. Hallucinations unrelated to medication

Clinical examination
   5. DSM criteria for dementia;
   6. Prominent slowing of vertical saccades or vertical supranuclear gaze palsy
   7. Evidence of focal cortical dysfunction such as aphasia, alien limb syndrome, and parietal dysfunction

Laboratory Investigation
   8. Metabolic, molecular genetic, and imaging evidence of an alternative cause of features listed above.

[#]We added a set of softer features recommended by the SIC in their evaluation of Criteria for MSA[29] with the aim to pick up the diagnosis and classify more earlier presentations of the disease.

C} Cortico-basal Degeneration (CBD) Diagnostic Criteria by Boeve et al[27]

Core features
   •Insidious onset and progressive course
   •No identifiable cause (e.g. tumor; infarct)
   •Cortical dysfunction as reflected in at least one of the following

o Focal or asymmetrical limb apraxia
o Alien limb phenomenon
o Cortical sensory loss
o Visual or sensory neglect
o Constructional apraxia
o Focal or asymmetric myoclonus
o Apraxia of speech/ nonfluent aphasia

• Extrapyramidal dysfunction as reflected by at least one of the following
o Focal or asymmetrical appendicular limb rigidity lacking prominent and sustained L Dopa response
o Focal or asymmetrical appendicular dystonia

Supportive investigations
• Variable degrees of focal or lateralized cognitive dysfunction, with relative preservation of learning and memory, on neuropychometric testing
• Focal or asymmetric atrophy on computed tomography or magnetic resonance imaging, typically maximal in parietofrontal cortex
• Focal or asymmetric hypoperfusion on single-photon emission computed tomography and positron emission tomography, typically maximal in parietofrontal cortex +/- basal ganglia +/- thalamus

D} Parkinson's disease (PD) – as per the UKPDS Brain bank Criteria[31]

Step 1 - Diagnosis of Parkinsonism
Bradykinesia and at least one of the following
• Bradykinesia
• Muscular rigidity
• 4-6 Hz. Resting tremor
• Postural instability not caused by primary visual, vestibular, cerebellar or Proprioceptive dysfunction

Step 2 - Features tending to exclude Parkinson's disease as a cause of the Parkinsonism#
1. History of repeated strokes with stepwise progression of parkinsonian features
2. History of repeated head injury
3. History of definite encephalitis
4. Neuroleptic treatment at onset of symptoms
5. Sustained remission
6. Strictly unilateral features after 3 years
7. Supranuclear gaze palsy
8. Cerebellar signs
9. Early severe autonomic involvement
10. Early severe dementia with disturbances of memory, language and praxis
11. Babinski's sign
12. Presence of a cerebral tumour or communicating hydrocephalus on computed tomography scan
13. Negative response to large doses of levodopa (if malabsorption excluded)
14. MPTP exposure

The criteria of '>1 affected relatives' as an exclusion criteria was removed

Step 3 - Features that support a diagnosis of Parkinson's disease (three or more required for diagnosis of definitive Parkinson's disease)
1. Unilateral onset
2. Rest tremor present
3. Progressive disorder
4. Persistent asymmetry affecting the side of onset most
5. Excellent (70–100%) response to levodopa
6. Severe levodopa-induced chorea
7. Levodopa response for ≥5 years
8. Clinical course of ≥10 years

Supplementary Data

65. Canu E, Agosta F, Baglio F, Galantucci S, Nemni R, Filippi M. Diffusion tensor magnetic resonance imaging tractography in progressive supranuclear palsy. Mov Disord. 2011;26:1752-5.

METHODS

Controls included 21 cases who had undergone MRI scans of the brain for the complaints of headaches, 16 for transient neurological deficits including sensory and motor complaints, seven with vertigo, five with cranial nerve related symptoms including earache and vision impairment, five with history of head injuries, four with episodes of loss of consciousness, four with requests to look for space occupying lesions, and two for screening due to familial disorders. All the MRI scans had previously reported as normal.

In most cases the following sequences were acquired (in the axial plane except for the T_1 sequence): T_1-weighted images were acquired using a conventional spin echo with the following parameters: repetition time 500 msec, echo time 8 msec, slice thickness of 5mm, slice gap of 2mm, field of view (FOV) 240mm, matrix 256 X192; T_2-weighted images were acquired with fast spin echo (FSE) with the following parameters: repetition time 4,450 msec, echo time 90 msec, slice thickness of 5mm, slice gap of 2mm, FOV 220mm, matrix 256 X 256, echo train length of 17. FSE images were obtained using the following parameters: repetition time 10,000 msec, echo time 150 msec, inversion time 2200 msec, slice thickness of 5mm, slice gap of 2mm, FOV 220mm, matrix 256 X 192. DWI imaging was recorded using spin-echo echoplanar imaging (EPI) sequence with the following parameters: b-value of 1000 s/mm², repetition time 8,000 msec, echo time 63 msec, slice thickness of 5mm, no gap, FOV 260mm, matrix 128 X 128s. GRE imaging was recorded using a gradient-echo EPI sequence[2] and the following parameters: repetition time 4,000 msec, echo time 30 msec at 1.5T 25msec at 3T, slice thickness of 5mm, no gap, FOV 240mm, matrix 256 X 384s

RESULTS

Among raters moderate agreement was found for the presence of putamen hypointensity on GRE scans, with complete agreement within the same rater when rated at different time points (supplementary Table 2).

Brief case reports of patients not fulfilling diagnostic criteria for MSA-P but who had putamenal atrophy and GRE hypointensity.

One undiagnosed patient (case 195) was a 66-year-old gentleman with asymmetric parkinsonism of three year duration with a poor response to L-Dopa. He had a jerky tremor, erectile dysfunction and marked constipation. He had no urinary incontinence or orthostatic symptoms. He did not fulfill criteria for MSA at the time of assessment. The second patient (case 22) was a 53-year-old lady with tremor dominant symptoms who fulfilled criteria for PD. She had been followed for four years after disease onset. Apart from a jerky tremor and some stimulus sensitive myoclonus in the fingers, there were no other atypical features. She had been on a very low dose of L-Dopa due to nausea making an assessment of her response difficult. The last patient (case 37) fulfilled the criteria for PSP. He was an 80-year-old gentleman with L-Dopa unresponsive akinetic rigid state with early falls, abnormal extraoccular movements and bulbar involvement.
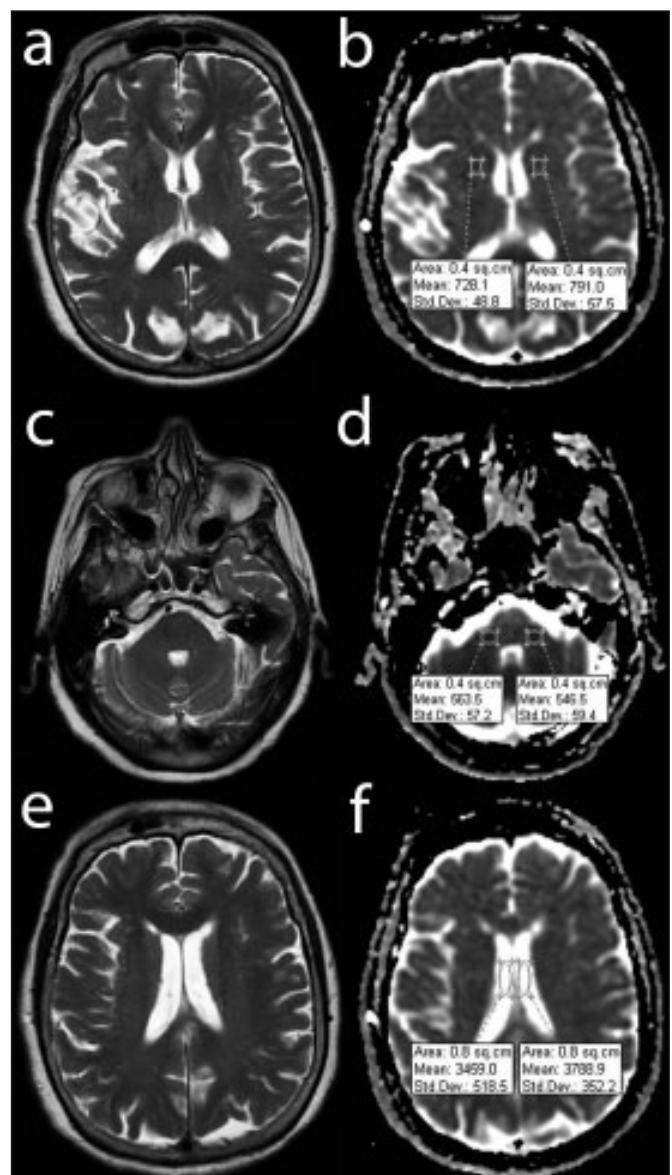
Supplementary Figure: *Region of Interest mapping for ADC. T2 axial (a,c,e) and corresponding ADC images (b,d,f) showing the calculation of ADC values in patients. The ROIs were applied to the area of the putamen (b), Middle cerebellar peduncle (d) and the lateral ventricles bilaterally for the calculation of mean ADC of the region of interest using the corresponding T2 axial images for anatomical reference.*

Supplementary Table 1: Signs on T1/ T2 MRI

Characteristic	CBS	Control	MSA-C	MSA-P	PD	PSP	Undiagnosed	Total	Significance
Number of cases	6	64	7	15	61	21	21	195	
T_2 Atrophy of putamen (%)	1 (16.7)	4 (6.3)	3 (42.9)	11 (71.3)	16 (26.2)	11 (52.4)	7 (31.3)	54 (24.6)	<0.0001
T_2 hypointensity of putamen[a,b] (%)	1 (16.7)	2 (3.1)	0	6 (40)	6 (9.8)	2 (9.5)	2 (9.5)	19 (9.7)	0.013
slit-like hyperintensity of putamen[a,b] (%)	0 (0)	3 (4.7)	0 (0)	9 (60)	4 (6.6)	3 (14.3)	5 (23.8)	26 (13.3)	p<0.0001
Presence of Humming-bird sign (%)	0	0	0	3 (20)	5 (8.2)	10 (47.6)	2 (9.5)	20 (10.3)	p <0.0001
Presence of morning glory sign (%)	0	0	0	0	0	2 (9.5)	1 (4.8)	3 (1.5)	p <0.0001
Presence of the Hot-cross-bun sign[c] (%)	0	0	5 (71.4)	2 (13.3)	0	2 (9.5)	1 (4.8)	10 (5.1)	p <0.0001
Presence of hyperintensity of MCP[c] (%)	1 (16.7)	0	3 (42.9)	2 (13.3)	0	0	1 (4.8)	7 (3.6)	p <0.0001
Mean pontine AP diameter +/- SD (mm)	2.25 +/- 0.18	2.32 +/- 0.14	1.94 +/- 0.21	2.16 +/- 0.23	2.36 +/- 0.15	2.19 +/- 0.18	2.26 +/- 0.17	2.28 +/-0.19	p<0.0001

CBS = Corticobasal syndrome, MSA-C = Multiple system atrophy cerebellar type, MSA-P = Multiple system atrophy parkinsonian type, PD = Parkinson's disease, PSP = Progressive supranuclear palsy. [a] Taken as all cases with the presence of either unilateral or bilateral signs. [b] It was not possible to assess the characteristics in 2 cases of PD due to technical difficulties. (denominator for PD was 59, total cases 193) [c] It was not possible to assess the characteristics in 1 case with PD due to technical difficulties. (denominator for PD was 60)

Supplementary Table 2: Intra-rater and inter-rater reliability analysis of the different signs

Variable	Intra-rater reliability – kappa value[a]	Inter-rater reliability – kappa value[b]
Atrophy of putamen on T_2[c]	0.5758	0.5205
T_2 Hypointensity of Putamen[c]	0.4167	0.7143
T_2 Slit-like hyperintensity of putamen[c]	0.5758	0.5946
Hummingbird sign	1.0	0.6364
Loss of tegmental convexity	1.0	1.0
Hot cross bun sign	1.0	0.7671
Hyperintensity of MCP	1.0	0.6364
GRE Hypointensity of putamen	1.0	0.4576

GRE = gradient echo imaging, MCP = middle cerebellar peduncle. [a] 31 scans rated by each of the two blinded raters twice. [b] 14 scans rated by the two blinded raters twice at different time points. [c] Taken as all cases with the presence of either unilateral or bilateral signs. Interpretation of kappa values: 0.4-0.59 mod agreement; 0.6-0.79 substantial agreement, 0.8-1.0 outstanding agreement.

Supplementary Table 3: Distribution of undiagnosed cases and role of MRI

Clinical Syndrome among the undiagnosed group	N	No. with relevant MRI signs	Description of relevant abnormal MRI Scans
Parkinsonism with autonomic features not fitting MSA criteria	4	2	Case 195[a] had GRE hypointensity and atrophy of the putamen on two MRI scans a year apart. Case 147[b] had a hot cross bun sign and T_2 hyperintensity of the MCP
Sporadic ataxia not fitting criteria for MSA	4	2	2 (case 35[c], 81[d]) had significantly higher rADC values in the MCP compared to the cut off value (0.94 X 10-3 mm²/sec)
Parkinsonism with predominant gait disturbance and postural instability not fulfilling criteria of PD or PSP	9	2	2 showed the presence of a hummingbird sign (case 1[e], 176[f]), case 1 also had a morning glory sign
Corticobasal like syndrome not fulfilling criteria	3	0	
Asymmetric Parkinsonism with a resting tremor not fulfilling criteria for PD yet.	1	0	
Total Undiagnosed cases	21	6	

CBS = Corticobasal syndrome, MSA = Multiple system atrophy, PD = Parkinson's disease, PSP = Progressive supranuclear palsy GRE = gradient echo imaging, MCP = middle cerebellar peduncle. [a]Case 195 was a 66 year old gentleman with asymmetric parkinsonism of three year duration with a poor response to L-Dopa. He had a jerky tremor, erectile dysfunction and marked constipation. He had no urinary incontinence or orthostatic symptoms. He did not fulfill criteria for MSA at the time of assessment. [b]Case 147 was a 60 year old lady presenting with parkinsonism with a right foot tremor. She also had ataxia and mild autonomic dysfunction not fulfilling the criteria for MSA. She had a long disease course with stridor. [c]Case 35 had asymmetric parkinsonism with mild autonomic features and dementia in addition to ataxia. He died within 4 years of his illness. [d]Case 81 had ataxia with no autonomic features or parkinsonism. [e]Case 1 had forward falls, forgetfulness, diminished vertical saccades and pursuits and parkinsonism. He also had urinary incontinence and enlarged ventricles on the MRI raising the question of NPH, though clinically he had more features of PSP. [f]Case 176 had L-Dopa unresponsive parkinsonism, with early forward falls, dysphagia, dementia, urinary incontinence, postural rather than resting tremors. Over 3 years she progressed to an akinetic rigid mute state with dementia. Her vertical gaze was restricted.