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Phenotypic spectrum of probable and genetically-confirmed idiopathic basal ganglia calcification

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Idiopathic basal ganglia calcification is characterized by mineral deposits in the brain, an autosomal dominant pattern of inheritance in most cases and genetic heterogeneity. The first causal genes, *SLC20A2* and *PDGFRB*, have recently been reported. Diagnosing idiopathic basal ganglia calcification necessitates the exclusion of other causes, including calcification related to normal ageing, for which no normative data exist. Our objectives were to diagnose accurately and then describe the clinical and radiological characteristics of idiopathic basal ganglia calcification. First, calcifications were evaluated using a visual rating scale on the computerized tomography scans of 600 consecutively hospitalized unselected controls. We determined an age-specific threshold in these control computerized tomography scans as the value of the 99th percentile of the total calcification score within three age categories: <40, 40–60, and >60 years. To study the phenotype of the disease, patients with basal ganglia calcification were recruited from several medical centres. Calcifications that rated below the age-specific threshold using the same scale were excluded, as were patients with differential diagnoses of idiopathic basal ganglia calcification, after an extensive aetiological assessment. Sanger sequencing of *SLC20A2* and *PDGFRB* was performed. In total, 72 patients were diagnosed with idiopathic basal ganglia calcification, 25 of whom bore a mutation in either *SLC20A2* (two families, four sporadic cases) or *PDGFRB* (one family, two sporadic cases). Five mutations were novel. Seventy-one per cent of the patients with idiopathic basal ganglia calcification were symptomatic (mean age of clinical onset: 39 ± 20 years; mean age at last evaluation: 55 ± 19 years). Among them, the most frequent signs were: cognitive impairment (58.8%), psychiatric symptoms (56.9%) and movement disorders (54.9%). Few clinical differences appeared between *SLC20A2* and *PDGFRB* mutation carriers. Radiological analysis revealed that the total calcification scores correlated positively with age in controls and patients, but increased more rapidly with age in patients. The expected total calcification score was greater in *SLC20A2* than *PDGFRB* mutation carriers, beyond the effect of the age alone. No patient with a *PDGFRB* mutation exhibited a cortical or a vermis calcification. The total calcification score was more severe in symptomatic versus asymptomatic individuals. We provide the first phenotypical description of a case series of patients with idiopathic basal ganglia calcification since the identification of the first causative genes. Clinical and radiological diversity is confirmed, whatever the genetic status. Quantification of calcification is correlated with the symptomatic status, but the location and the severity of the calcifications don't reflect the whole clinical diversity. Other biomarkers may be helpful in better predicting clinical expression.

Keywords: Fahr's disease; calcification; SLC20A2; PDGFRB; ageing

Abbreviations: BGC = basal ganglia calcification; IBGC = Idiopathic BGC; TCS = total calcification score

Introduction

Basal ganglia calcification (BGC) was first described in 1850 (Delacour, 1850) and is known to be caused by numerous conditions such as phosphocalcic metabolic disorders, mitochondrial diseases, numerous hereditary and non-hereditary congenital syndromes and acquired conditions (Baba *et al.*, 2005; Manyam, 2005). BGC has also been reported in normal ageing and seems to be common in older people (Simoni *et al.*, 2008; Yamada *et al.*, 2013). Nevertheless, thus far no published work has reported a method for determining when BGC can be attributed to normal ageing or to a pathological process. As BGC can be clinically silent, its classification as a normal part of ageing can therefore not be based exclusively on neuropsychiatric examination.

Idiopathic BGC (IBGC), also known as Fahr's disease, is clinically heterogeneous. Patients with calcification may exhibit neurological and/or psychiatric symptoms with diverse severity and ages of onset. Others can remain asymptomatic throughout life (Yamada and Hayashi, 2000; Manyam, 2005). For these reasons, the diagnosis is based on (i) the presence of BGC, which may or may not involve other brain areas, regardless of the clinical status; and (ii) the exclusion of the other causes of BGC. As a consequence, an extensive clinico-biological aetiological assessment is necessary to diagnose probable IBGC. Definite IBGC can now be diagnosed in patients bearing a causative mutation in one of the two recently identified genes: *SCL20A2* (Wang *et al.*, 2012) and *PDGFRB* (Nicolas *et al.*, 2013). Nevertheless, these two genes do not account for all cases of IBGC, confirming the genetic heterogeneity of the disease previously suggested by linkage studies (Oliveira *et al.*, 2004; Kostic *et al.*, 2011). Therefore, aetiological assessment remains necessary; firstly to select patients for genetic screening and secondly to attribute a probable diagnosis of IBGC in cases not related to a mutation in one of the two recently described genes. This type of assessment entails searching for known causes of BGC, including calcification associated with normal ageing.

Reports have shown wide clinical diversity within and between families. Calcification severity also seems to be diverse. Only one case series was published on different phenotypes associated with the disease before the discovery of the first causative genes (Manyam *et al.*, 2001). Their identification opens the door to clinical and radiological studies of patients affected by genetically confirmed IBGC. With the aim to describe the phenotype associated with definite IBGC as well as probable IBGC (with unknown genetic cause), we selected the patients bearing BGC with no known cause following an etiological assessment, and included only cases exhibiting calcifications rated above the age-specific threshold, determined by using data from controls. To rate the calcifications on CT scans, we used a new visual rating scale in controls and IBGC patients.

Patients and methods

Visual rating of calcifications

Axial view CT scans were analysed while blinded to identity, by two investigators (G.N. and D.H.), separately and then jointly, using an

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original visual grading system. Consensus was reached when the two investigators did not attribute the same score for each location. As this system was designed to improve the accuracy of IBGC diagnoses, we focused on brain parenchymal calcification. Calcifications were rated when there was a spontaneously hyperdense area with a Hounsfield value consistent with a calcification. The following locations were analysed: left and right lenticular nucleus, left and right caudate nucleus, left and right thalamus, left and right cerebral subcortical white matter, left and right internal capsule only if independent of other calcifications, cerebral cortex, left and right cerebellar hemisphere, vermis, left and right midbrain, pons and medulla. The scores were attributed according to specific definitions with visual examples (see Supplementary material for a comprehensive description): 0 = absent calcification; 1 = punctate; 2 = faint; 3 = moderate; 4 = severe; 5 = severe and confluent. The scores were compiled into a total calcification score (TCS) by addition.

Control patients

To determine age-specific thresholds, we rated the CT scans of control patients hospitalized in our university department of neurology during a 6-month period. The study was approved by our local ethics committee. All CT scans were performed using the same protocol (General Electric Lightspeed 64-slice scanner, 120 kVp, 280 mAs, 1.25 mm slice thickness, 0.9 mm interslice gap, non-contrast enhanced). We then sorted the CT scans into three categories according to the age of the control patients: $<$40.0 years, 40.0–59.9 years, $\geqslant$60.0 years of age. A total of 200 CT scans were rated in each age category. The main diagnosis or clinical context was collected for each control patient. If a large area involving the location(s) to be scored was not visible because of a pathological process (e.g. extensive haematoma or tumour), the CT scans were not included in the study. Scans showing brain calcification clearly attributable to a previously known pathological process (e.g. calcified tumour) were also not included. We determined the threshold for each category of age to be the value of the 99th percentile of the TCS.

Patients with idiopathic basal ganglia calcification

Patients were recruited from multiple centres and all patients and/or relatives gave informed, written consent. Each individual who participated in the study underwent a semi-structured interview, a neurological examination and a CT scan (with diverse acquisition protocols). The calcifications were reviewed and rated by the same investigators as for the scans of the control patients, using our visual rating scale. In accordance with previous studies, IBGC status was determined based first on radiological criteria (Kostic *et al.*, 2011; Nicolas *et al.*, 2013). The diagnosis of probable IBGC was retained if: (i) patients were affected by bilateral lenticular calcifications; (ii) the TCS was above the age-specific threshold ($\geqslant$99th percentile); and (iii) no cause was retrieved after an extensive aetiological assessment in the proband (Nicolas *et al.*, 2013). Genetic analyses were performed in the probands first diagnosed with probable IBGC. Patients with a mutation within *SLC20A2* or *PDGFRB* were then considered to be affected by definite IBGC, as well as their relatives bearing both mutation and BGC. Patients with IBGC were considered symptomatic if they presented at least one sign among the list provided in the previous case series (Manyam *et al.*, 2001). Symptomatic patients were evaluated with a neuropsychological test battery, which included, depending on age: Mini Mental State Examination, Mattis Dementia Rating Scale (if age $\geqslant$50), evaluation of episodic memory and assessment of executive functions. Episodic verbal memory was assessed using Free and Cued Recall Test (if age $\geqslant$50). Cueing was considered efficient when patients with impaired free recall achieved normal total recall (Sarazin *et al.*, 2007; Godefroy *et al.*, 2009). Executive functions were evaluated using the Frontal Assessment Battery, Stroop Test, Trail Making Test, Modified Card Sorting Test and verbal fluencies. Patients were considered to suffer from cognitive dysexecutive syndrome according to normative data (Godefroy *et al.*, 2010). Complementary neuropsychological evaluation included, when available: Oral Naming Test, Gestural Praxis Evaluation and Rey Figure Copy.

Genetic analyses

DNA was extracted from peripheral blood. Sanger sequencing of *SLC20A2* and *PDGFRB* was performed in each proband. Segregation of the novel variants (not retrieved in the dbSNP132 and EVS databases) was studied when DNA from relatives was available. We concluded that a variant was a disease-causative mutation if: it was not retrieved among the dbSNP132 and EVS databases or our 173 in-house controls (obtained by whole-exome sequencing); the variant was highly disruptive (e.g. introducing a frameshift or altering the splice); or a missense variant was predicted to alter the protein function by at least two software programmes (PolyPhen2, Mutation taster and SIFT) or by one software programme and the base was highly conserved across species (according to the PhyloP score) (Liu *et al.*, 2011).

Statistics

Inter-rater agreement was assessed using Cohen's weighted kappa reliability test. Correlations between age and TCS were evaluated using a Kendall correlation test. Comparisons between the three groups of age in controls were performed using Fisher exact tests. We used *t*-tests to compare the mean scores between symptomatic and asymptomatic patients with IBGC and differences between the mean ages of the *SLC20A2* and *PDGFRB* mutation carriers. To account for the combined effects of age and genotypes (definite IBGC with *SLC20A2* or *PDGFRB* mutation, and probable IBGC with unknown genetic cause) on the quantification scores (TCS and scores from each location), we adopted a negative binomial model, which allows for over-dispersion of count data (Ver Hoef and Boveng, 2007; Zeileis *et al.*, 2008).

We used the statistical software R.

Results

Control patients

Inter-rater agreement was excellent (weighted kappa = 0.969). Of the CT scans analysed consecutively, 30 showed a large pathological process and were thus impossible to rate. These 30 control patients were therefore not included in the study. Three more control patients were not included because of a known cause of brain calcification (unilateral thalamus calcification in a 45-year-old patient, located in a sequelae of a toxoplasma infection, brainstem tumour calcification in a 47-year-old patient, and another calcified tumour in the cerebral cortex of a 59-year-old patient). Finally, 200 control patients were included in each group (Table 1). The main diagnosis or clinical context of the CT scans in the 600

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control patients is provided in Supplementary Table 1. Ninety control patients (15%) had a TCS > 0. Among them, 87 had lenticular calcifications (50 unilateral, 37 bilateral), with a maximum score of 3 for each lenticular calcification (Table 1). Five control patients showed calcifications in areas other than lenticular nuclei; they were all >60 years old (Supplementary Note 1). In the group of control patients who were <40, only one (0.5%), aged 28, presented a calcification. In the group of patients between 40 and 60 years old, 29 (14.5%) had a TCS > 0. In the group of control patients over 60 years old, 60 (30.0%) had a TCS > 0.

There were significantly more patients with calcifications in the older groups ($P < 0.0001$ for each comparison, except that between 40–60 years and >60 for which $P = 0.0002$) and there was a positive correlation between age and TCS (Kendall tau = 0.2873883, $P < 0.0001$).

The 99th percentile of the TCS were: 0 in <40 years, 4 in 40–60 years, and 5 in >60 years.

Patients with idiopathic basal ganglia calcification

Of the 53 index patients referred for bilateral lenticular calcifications, 15 were excluded from our study: three scored a TCS under the age-specific threshold (they did not have family history of neuropsychiatric disorders and were diagnosed respectively with stroke, vascular dementia and Alzheimer's disease with compatible CSF biomarkers) and 12 were attributed another cause following the aetiological assessment [pseudohypoparathyroidism type 1a ($n = 2$), type 1b ($n = 1$); hyperparathyroidism with renal insufficiency ($n = 2$), with parathyroid gland hyperplasia ($n = 2$); brain irradiation *in toto* ± chemotherapy ($n = 2$); Down syndrome ($n = 1$); neurolupus ($n = 1$) and uncharacterized autoimmune disease ($n = 1$)].

Thus, the diagnosis of probable IBGC was first retained in 38 patients. Among them, nine belonged to a pedigree with a demonstrative autosomal dominant inheritance of IBGC and six belonged to families in which the number of relatives with available brain imaging was insufficient to ascertain a specific pattern of inheritance. Taking into account the affected relatives, the IBGC series was comprised of 38 patients from the nine autosomal dominant families, 11 patients from the six unspecified families, and 23 with a sporadic presentation (total = 72).

Genetic analyses

We previously reported two families with an *SLC20A2* mutation (of four and two affected individuals) (Nicolas *et al.*, 2013). We found four new *SLC20A2* mutations in four cases with a sporadic presentation (Table 2). None of the four new *SLC20A2* mutations have previously been reported by others (Schottlaender *et al.*, 2012; Wang *et al.*, 2012; Hsu *et al.*, 2013; Lemos *et al.*, 2013; Zhang *et al.*, 2013). All relatives of an *SLC20A2* mutation carrier who also carried the mutation exhibited BGC and were thus included.

We previously reported two missense mutations within *PDGFRB* in a large family (13 affected individuals, Family 870) and a case

Table 1 Demographic characteristics and calcification scores of the controls

Group	Mean age ± SD	Sex (males / females)	Presence of calcifications		Lenticular calcification, unilateral, *n* / max score	Lenticular calcification, bilateral, *n* / max score	Caudate calcification, *n* / max score	Cerebellar hemisphere calcification, *n* / max score	Cortical calcification, *n* / max score	Other locations	TCS: 1st percentile (threshold)
			Yes *n* (%)	Maximum TCS							
<40 years ($n = 200$)	29.6 ± 6.4	75/125	1 (0.5)	1	1 / 1	0	0	0	0	0	**0**
40–60 years ($n = 200$)	50.7 ± 5.8	101/99	29 (14.5)	6	15 / 3	14 / 6	0	0	0	0	**4**
⩾60 years ($n = 200$)	75.1 ± 8.2	108/92	60 (30.0)	6	34 / 3	23 / 6	1 / 3	3 / 1	1 / 3	0	**5**

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Table 2 Mutations within *SLC20A2* and *PDGFRB* in the definite cases of IBGC. Control exomes are from 173 individuals with non-neurological or psychiatric disease

Individual or family identification	Affected (*n*)	Gene	Mutation	c.DNA*	Protein*	dbSNP132, EVS, control exomes (*n* = 173)	Mutation Taster	Polyphen2	SIFT	PhyloP
Family 1038 (Ref 1)	2	*SLC20A2*	Missense	c.551C > T	p.Pro184Leu	Absent	**Disease causing (0.99)**	**Probably damaging (0.98)**	**Deleterious (0.05)**	**5.77**
Family 752 (Ref 1)	4	*SLC20A2*	**Splice**	c.431-1G > T	NA	Absent	NA	NA	NA	**5.69**
Case 413 (new)	1	*SLC20A2*	Missense	c.1711G > A	p.Glu571Ser	Absent	**Disease causing (0.99)**	**Probably damaging (1.00)**	**Deleterious (0.00)**	**5.53**
Case 485 (new)	1	*SLC20A2*	Missense	c.581A > G	p.Asn194Ser	Absent	**Disease causing (0.99)**	Benign (0.155)	Tolerated (0.29)	**4.73**
Case 424 (new)	1	*SLC20A2*	**Frameshift**	c.1527delT	p.Asn509LysFs*7	Absent	**Disease causing (1.00)**	NA	NA	0.21
Case 1352 (new)	1	*SLC20A2*	Missense	c.82G > A	p.Asp28Asn	Absent	**Disease causing (0.99)**	**Probably damaging (1.000)**	**Deleterious (0.00)**	**5.69**
Family 870 (Ref 1)	13	*PDGFRB*	Missense	c.1973T > C	p.Leu658Pro	Absent	**Disease causing (0.99)**	**Probably damaging (1.00)**	**Deleterious (0.00)**	**4.89**
Case 1060 (Ref 1)	1	*PDGFRB*	Missense	c.2959C > T	p.Arg987Trp	Absent	**Disease causing (0.95)**	**Probably damaging (1.00)**	**Deleterious (0.00)**	**1.50**
Case 1106 (new)	1	*PDGFRB*	Missense	c.3212A > T	p.Glu1071Val	Absent	Polymorphism (0.99)	**Possibly damaging (0.745)**	**Deleterious (0.01)**	**3.51**

Ref 1 = Nicolas *et al.*, 2013. Items to be included in our criteria for disease-causative mutations are in bold type.
*The nomenclature of the mutations refers to transcripts NM_006749 (*SLC20A2*) and NM_2609 (*PDGFRB*).

with a sporadic presentation (Nicolas *et al.*, 2013). In Family 870, all relatives carrying the familial mutation presented BGC with diverse severity (see brain imaging section) and were therefore included. We found a new *PDGFRB* missense mutation in a patient with a sporadic presentation (Table 2).

All 25 patients with a mutation in *SLC20A2* or *PDGFRB* were therefore diagnosed with definite IBGC. A total of 47 patients had no mutation in the known IBGC-causative genes, and were diagnosed with probable IBGC (Table 3, 'unknown').

Neurological and psychiatric symptoms

Most (70.8%) of our patients were symptomatic with a mean age of 54.9 ± 18.8 years at last evaluation and clinical onset at a mean age of 39.2 ± 20.0 years; 29.2% were asymptomatic (mean age at the time of their last evaluation: 49.3 ± 18.3 years, $P = 0.94$) (Table 3). The most frequent symptoms were cognitive impairment (58.8% of the symptomatic patients), psychiatric signs (56.9%) and movement disorder (54.9%) (Table 3).

The most frequently impaired cognitive field was memory (70%). In patients with memory impairment, episodic memory disorder was characterized by impaired encoding and retrieval with efficient cueing in 9 of 21 patients (43%). This pattern is consistent with frontal subcortical dysfunction. Conversely, 10 patients (47%) had severe episodic memory impairment without efficient cued recall [for 2 of 21 (10%), memory impairment was not assessed by free and cued recall test]. Sixty per cent of the patients with cognitive impairment presented a dysexecutive syndrome. Among the symptomatic patients with complementary neuropsychological evaluation, oral naming was pathological in 2 of 13, gestural praxis in 3 of 10, and Rey figure copy in 7 of 16, according to normative data. One sporadic patient and two patients of two autosomal dominant families presented intellectual disability.

Psychiatric signs included mostly mood disorders (59%), ranging from one or recurrent depressive episode(s) to bipolar disorder, and psychotic symptoms (28%) ranging from some positive signs to psychosis. Other psychiatric symptoms included personality disorders, attention deficit hyperactivity disorder, uncharacterized behavioural disturbances and somatoform symptoms. No obsessional compulsive disorder was diagnosed. Interestingly, psychosis was clinically undistinguishable from schizophrenia in five patients. Among them, three belonged to a pedigree with autosomal dominant inheritance (unknown genetic cause) (Le Ber *et al.*, 2007). The other two had a sporadic presentation: first, a patient with a mutation within *PDGFRB* (age of psychosis onset = 16 years), who later experienced two ischaemic strokes at the age of 62; and second, a woman without known genetic cause (age of psychosis onset = 35 years) presenting intellectual disability, depressive elements and who insidiously developed extrapyramidal, pyramidal sign and gait disorder with static cerebellar syndrome in her early 50 s. Three additional patients had some psychotic signs: one patient, with a sporadic presentation and no known genetic cause, had positive signs (delusion, hallucinations), apathy and then a major depressive episode without disorganized speech or behaviour (Nicolas *et al.*, 2012). The other two developed few positive signs (visual hallucinations in one and a transient delusion in the second).

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Table 3 Demographic and clinical characteristics of patients with definite (*SLC20A2* or *PDGFRB* mutation) and probable (unknown genetic cause) IBGC

	Definite IBGC		Probable IBGC	Total	Manyam *et al.* (2001)
Gene	*SLC20A2*	*PDGFRB*	Unknown	All	Unknown
Cases (*n*)	10	15	47	72	99
Inheritance					
Autosomal dominant, *n* families (cases)	2 (6)	1 (13)	6 (19)	9 (38)	NA (73)
Familial, unspecified, *n* families (cases)	0 (0)	0 (0)	6 (11)	6 (11)	NA (12)
Sporadic cases	4	2	17	23	14
Sex (male/female)	1.50	1.50	1.35	1.40	1.36
Asymptomatic, *n* (%)	**3 (30.0)**	**9 (60.0)**	**9 (19.1)**	**21 (29.2)**	**32 (32)**
Age at last evaluation (mean, SE)	67.0 (8.0)	43.3 (5.0)	49.4 (6.7)	49.3 (4.0)	32 (20)
Symptomatic, *n* (%)	**7 (70.0)**	**6 (40.0)**	**38 (80.9)**	**51 (70.8)**	**67 (68)**
Age at onset (mean, SE)	37.3 (8.9)	24.5 (8.6)	41.9 (3.0)	39.2 (2.8)	NA
Age at onset (range)	10–77	6–21	3–80	3–80	
Age at last evaluation (mean, SE)	67.0 (3.9)	46.2 (9.9)	54.1 (3.0)	54.9 (2.6)	47 (15)
Cognitive, *n* (%[a])	**5 (71.4)**	**2 (33.3)**	**23 (60.5)**	**30 (58.8)**	**26 (39)**
Dysexecutive	3	2	13	18 (60.0[b])	NA
Memory	4	2	15	21 (70.0[b])	NA
Intellectual disability	1	0	2	3 (10.0[b])	NA
Uncharacterized/other	2	1	11	14 (46.7[b])	NA
Psychiatric, *n* (%[a])	**5 (71.4)**	**5 (83.3)**	**19 (50.0)**	**29 (56.9)**	**21 (31)**
Psychotic signs	1	1	6	8 (27.6[c])	NA
Mood disorder	2	2	13	17 (58.6[c])	NA
Other	2	2	2	6 (20.7[c])	NA
Movement disorder, *n* (%[a])	**5 (71.4)**	**2 (33.3)**	**21 (55.3)**	**28 (54.9)**	**37 (55)**
Akinetic-hypertonic	4	2	16	22 (78.6[d])	21 (57[b])
Tremor	1	1	5	7 (25.0[d])	3 (8[b])
Chorea	0	1	3	4 (14.3[d])	7 (19[b])
Dystonia	1	1	5	7 (25.0[d])	3 (8[b])
Athethosis	0	0	0	0 (0.0[d])	2 (5[b])
Orofacial Dyskinesia	1	0	2	3 (10.7[d])	1 (3[b])
Other	1	0	1	2 (7.1[d])	NA
Gait, *n* (%[a])	**3 (42.9)**	**1 (16.7)**	**18 (47.4)**	**22 (43.1)**	**12 (18)**
Speech, *n* (%[a])	**1 (14.3)**	**2 (33.3)**	**17 (45.9)**	**20 (39.2)**	**24 (36)**
Cerebellar, *n* (%[a])	**0 (0.0)**	**1 (16.7)**	**13 (35.1)**	**14 (27.5)**	**24 (36)**
Pyramidal, *n* (%[a])	**1 (14.3)**	**1 (16.7)**	**10 (26.3)**	**12 (23.5)**	**15 (22)**
Seizures, *n* (%[a])	**0 (0.0)**	**0 (0.0)**	**4 (10.5)**	**4 (7.8)**	**6 (9)**
Gastro-intestinal, *n* (%[a])	**0 (0.0)**	**0 (0.0)**	**4 (10.5)**	**4 (7.8)**	**8 (12)**
Genito-urinary, *n* (%[a])	**0 (0.0)**	**1 (16.7)**	**3 (7.9)**	**4 (7.8)**	**9 (13)**
Sensory/pain, *n* (%[a])	**1 (14.3)**	**0 (0.0)**	**1 (2.6)**	**2 (3.9)**	**11 (16)**

Uncharacterized cognitive impairments include patients with pathological global scores without available tests examining memory or executive functions. Other cognitive impairments include pathological oral naming, Rey figure copy, and gestural praxis. For comparison, data from the case series reported by Manyam *et al*. (2001) are recalled on the right (38 cases seen in a registry and 61 cases from literature review) but may not be strictly compared for each point (in Manyam's series, details were not provided and patients with 'abnormal development' were not included, so the patients with intellectual disability, classified here among the patients with cognitive impairment, are not comparable).

[a]Among symptomatic patients.

[b]Among patients with cognitive impairment.

[c]Among patients with psychiatric signs.

[d]Among patients with movement disorders.

Movement disorders included mostly akinetic-hypertonic syndrome (78.6%) with or without tremor. Motor improvement with dopatherapy was noted in five cases (by interview of patients and their neurologist). An acute levodopa challenge was performed on two of these five patients: Unified Parkinson's Disease Rating Scale III motor score improved by 25% and 47%. The latter is the proband of the family with a *PDGFRB* mutation, and presented ON/OFF fluctuations and dyskinesia during the course of the disease, which was clinically indistinguishable from idiopathic Parkinson's disease (Nicolas *et al.*, 2013). For five other patients, no subjective motor improvement was reported. For the remaining 12, dopatherapy was not introduced, in most cases because Parkinsonism's severity was mild. Hyperkinetic signs were less frequent and included chorea, dystonia, and orofacial dyskinesia.

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Gait (43.1%) and speech (39.2%) disorders were frequent and of multiple origin, mostly due to akinetic-hypertonic syndrome and cerebellar signs. Pyramidal signs were present in 23.5% of symptomatic patients, associated only once with paresis. Four patients presented seizures, ranging from one episode to several seizures but none presented hard-to-treat epilepsy.

The most frequently associated signs were: cognitive impairment with movement disorder (20 patients), cognitive impairment with psychiatric signs (17 patients), cognitive impairment with gait disorder (17 patients) and movement disorder with gait disorder (16 patients).

Clinical onset appeared to be earlier when associated with *PDGFRB* mutations, compared with *SLC20A2*, but the difference was not statistically significant. Seventy per cent of the *SCL20A2* patients versus 40% of the *PDGFRB* patients were symptomatic, but the almost significant older age of evaluation of the *SLC20A2* patients ($P = 0.095$ for symptomatic and $P = 0.071$ for asymptomatic patients) could account at least for a part of the difference.

No patient with a *SLC20A2* or a *PDGFRB* mutation experienced seizures, and none of the *SLC20A2* mutation carriers presented cerebellar signs despite the presence of extensive cerebellar calcifications. Movement disorders seemed more common in *SLC20A2* mutation carriers.

Migraine, with or without aura, was reported in 15 patients in our study (nine classified among the symptomatic patients, and six classified among the asymptomatic patients).

Interestingly, four patients diagnosed with probable IBGC presented cerebral haemorrhage(s), which led to the fortuitous discovery of BGC (Supplementary Note 2).

Brain imaging

In patients with IBGC, inter-rater agreement was acceptable (weighted kappa = 0.67). Calcification was most frequently found in (besides lenticular nuclei, 100%, by definition): caudate nuclei, cerebellar hemispheres, and cerebral white matter (each 55%), thalami (43%), cortex (33%), vermis (32%), midbrain (10%), pons (7%) and medulla (3%) (Fig. 1 and Supplementary Table 2). Calcification of the internal capsules was not included in the figures and the Supplementary tables because in this location it systematically involved adjacent location and consequently had to forego scoring.

Among the patients with a definite diagnosis of *PDGFRB*-related IBGC, one patient presented calcifications under the age-specific threshold. In order to consider the effects of age and genotype on TCS, we studied both definite and probable IBGC patients, and excluded the aforementioned patient from the statistics. The TCS correlated positively with age (tau = 0.28, $P = 0.0009$). Using a negative binomial regression for overdispersed count data, we found that the mean TCS increased more rapidly in patients than control subjects significantly ($P < 0.0001$, Fig. 2A).

Patients with an *SLC20A2* mutation had a greater expected TCS than those with a *PDGFRB* mutation, with respect to age at CT scan ($P = 0.0004$) (Fig. 2B). In particular, the mean scores of the following locations were greater in the *SLC20A2* group: lenticular ($P = 0.0111$), caudate ($P = 0.0105$) and subcortical white matter ($P = 0.0189$), the mean scores of the thalamus and the cerebellar hemispheres approaching significance ($P = 0.0916$ and $P = 0.06934$, respectively). Cortical calcifications were located in the occipital cortex and/or in the depth of the sulci (Fig. 3). Interestingly, no patient bearing a *PDGFRB* mutation presented cortical calcification, contrary to 77% of the patients with an *SLC20A2* mutation (Fig. 1B). This was also the case of the vermis calcifications, which were present in 77% of the *SLC20A2* and 0% of the *PDGFRB* mutation carriers (Fig. 1B). Examining the cerebellum more closely, we noted that cerebellar calcifications in the *PDGFRB* mutation carriers were restricted to or centred on the dentate nuclei. However, cerebellar calcification of the *SLC20A2* mutation carriers seemed to be restricted to the dentate nuclei only in one case, and more widespread in the others. The brainstem was rarely involved; calcification was seen in the midbrain of seven patients, including one *SLC20A2* mutation carrier with faint bilateral calcifications in the upper part of the midbrain, and another with similar appearance and no genetic cause. The other five (three siblings and two unrelated patients) did not carry a mutation in either of the two genes and were the only patients showing calcification in the pons, in addition to all other locations, except for medulla (that was calcified in only one of these five patients). The medulla was also calcified in an unrelated patient devoid of other brainstem calcification (unknown genetic cause).

Among the 48 symptomatic and the 20 asymptomatic patients (Supplementary Table 2), the mean age at the time the CT scans were performed was not significantly different (mean $\pm$ SD: 54.5 ± 17.8 versus 47.3 ± 18.1), and the mean age of clinical onset of symptomatic patients was significantly lower than the mean age at the last clinical evaluation of asymptomatic patients (one-sided *t*-test: $P = 0.027$). The TCS was significantly higher in symptomatic compared to asymptomatic patients {mean [95% confidence interval (CI)]: 33.6 (27.3–39.9) versus 14.5 (8.3–20.7), $P = 0.0007$}. Similar results were observed pertaining to the location of calcification excepting midbrain, pons, and medulla, which were involved only in symptomatic patients but the number of patients exhibiting such calcification was too small to obtain significant results (Fig. 1C and Supplementary Table 2).

Discussion

This study is the first systematic description of the phenotype associated with IBGC since the discovery of the first causative genes. We found that patients with *SLC20A2* mutations exhibited more severe calcifications than those with *PDGFRB* mutations. Moreover, a different pattern of calcifications appeared between these two groups, the absence of cortical and vermis calcification in *PDGFRB* mutation carriers having to be confirmed in further studies. Clinical and radiological diversity is confirmed, even among patients carrying the same mutation. Importantly, the severity of the calcifications correlated positively with age, and a more severe TCS was found in symptomatic versus asymptomatic individuals.

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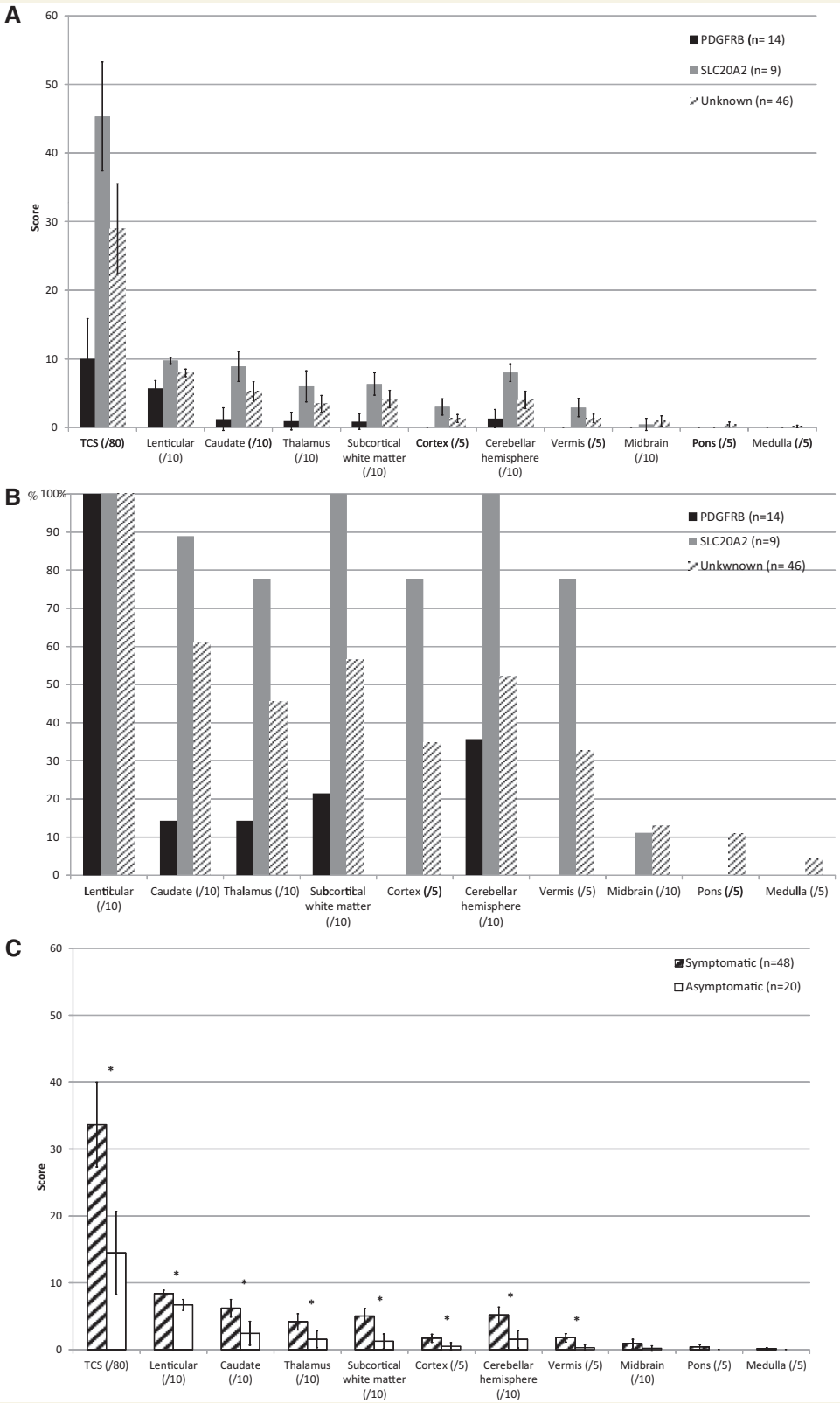

Figure 1 Calcification scores of patients with IBGC. Mean scores (95% CI) (**A**) and percentage of IBGC patients (**B**) affected by a calcification in each location (score > 0), among the patients with a mutation in *PDGFRB*, *SLC20A2* or with unknown genetic cause. Mean scores (95% CI) of symptomatic and asymptomatic patients with IBGC (**C**, $*P < 0.05$).

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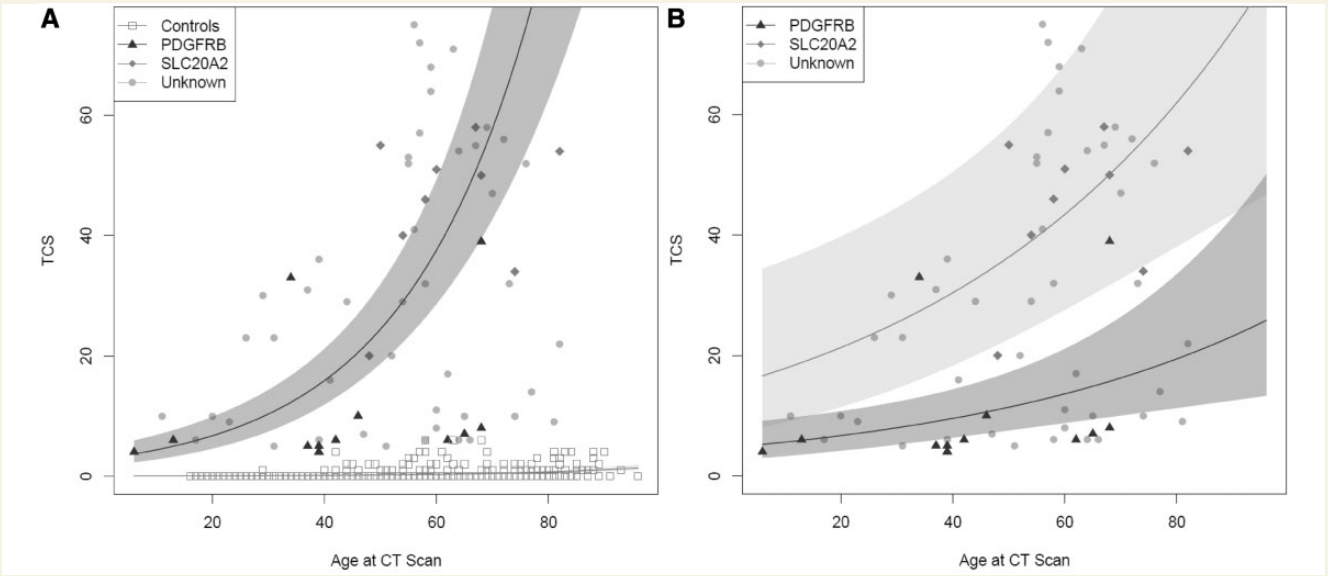

Figure 2 Total calcification scores. (**A**) Definite (*PDGFRB* and *SLC20A2*) and probable (unknown genetic cause) IBGC patients and control subjects (white squares). Positive correlation is seen in each group and the TCS increase more rapidly in patients with IBGC than controls ($P < 0.0001$). (**B**) Patients with IBCG by genotype. The curves represent the TCS predictions under a negative binomial model accounting for age and genotype for *PDGFRB* and *SLC20A2*. Grey areas represent 95% CI for the true expected TCS conditional on age and IBGC versus control status (**A**) or genotype (**B**). Note that these indicate the statistical confidence in the estimated mean effect; they do not encompass the population variation of observed TCS around their expectation. In **B**, the light grey zone is related to the *SLC20A2* mutation status and the dark grey zone to the *PDGFRB* mutation status. Under this model, patients with a mutation in *SLC20A2* exhibit significantly more severe calcifications than those with a mutation in *PDGFRB* ($P = 0.0004$).

Clinical phenotype

Three categories of signs were present in more than half of the symptomatic individuals: cognitive impairment, psychiatric signs and movement disorders. These signs were already predominant in the previous case series (Manyam *et al.*, 2001), but with an apparent lesser proportion of symptomatic patients exhibiting cognitive (39%) and psychiatric signs (31%) compared with ours. Cognitive impairment is a known symptom of IBGC and other diseases affecting the basal ganglia (Sobrido *et al.*, 1993–2004). In IBGC, some major cortico-subcortical loops may be disrupted, which is consistent with the subcortical memory impairments and dysexecutive syndromes seen in some of our patients and in other studies (Lopez-Villegas *et al.*, 1996). Conversely, some patients exhibit cortical calcification, in particular within the occipital lobe. Other neuropsychological tests focused on visuoconstructive functions would be of interest to explore their impact. We also found three patients with intellectual disability in our case series, including one with an *SLC20A2* mutation, as previously reported (Wang *et al.*, 2012). We note that intellectual disability cannot be compared to the study by Manyam *et al.* (2001), in which patients with 'abnormal development' were not included.

Psychiatric signs seemed to be more frequent in our series than in the previously published one (Manyam *et al.*, 2001), despite the fact that our patients were referred mostly by neurologists. Large diverse psychiatric signs have been associated with IBGC, such as psychotic signs (Geschwind *et al.*, 1999; Le Ber *et al.*, 2007; Nicolas *et al.*, 2012), personality disorders, obsessive-compulsive disorder, and mood disorders, including bipolar disorder (Trautner *et al.*, 1988; Konig, 1989; Lopez-Villegas *et al.*, 1996). More generally, BGC were significantly associated with psychotic symptoms in a population of non-demented 85-year-old individuals, but not with anxiety or mood disorders (Ostling *et al.*, 2003). In another study, 4 of 18 patients with BGC had mood disorders (bipolar disorder and depression) and six had obsessive-compulsive disorder (Lopez-Villegas *et al.*, 1996). In our case series, mood disorder and psychotic symptoms were the most frequent psychiatric signs, although we might have overestimated the proportion of mood disorders veritably due to IBGC, as these syndromes—especially depression—are frequent in the general population. Nevertheless, it seems that patients with IBGC of each genetic cause and with no known cause may all exhibit mood disorders or psychotic signs. Of note, obsessive-compulsive disorder seems to be a rare manifestation of IBGC and has been reported in only one *SLC20A2* mutation carrier to our knowledge (Hsu *et al.*, 2013).

As in the series by Manyam *et al.* (2001), akinetic-hypertonic syndrome was the most frequent movement disorder, whereas tremor and other hyperkinetic movements were less common.

Until now, there have been only two studies published with phenotypic data about *SLC20A2* mutation carriers (Wang *et al.*, 2012; Hsu *et al.*, 2013). It is interesting to note that seizures, which were not reported in our *SLC20A2* mutation carriers, were described in only two children, who were also affected by intellectual disability/developmental delay (Wang *et al.*, 2012). Clinical onset of other patients was primarily during adulthood.

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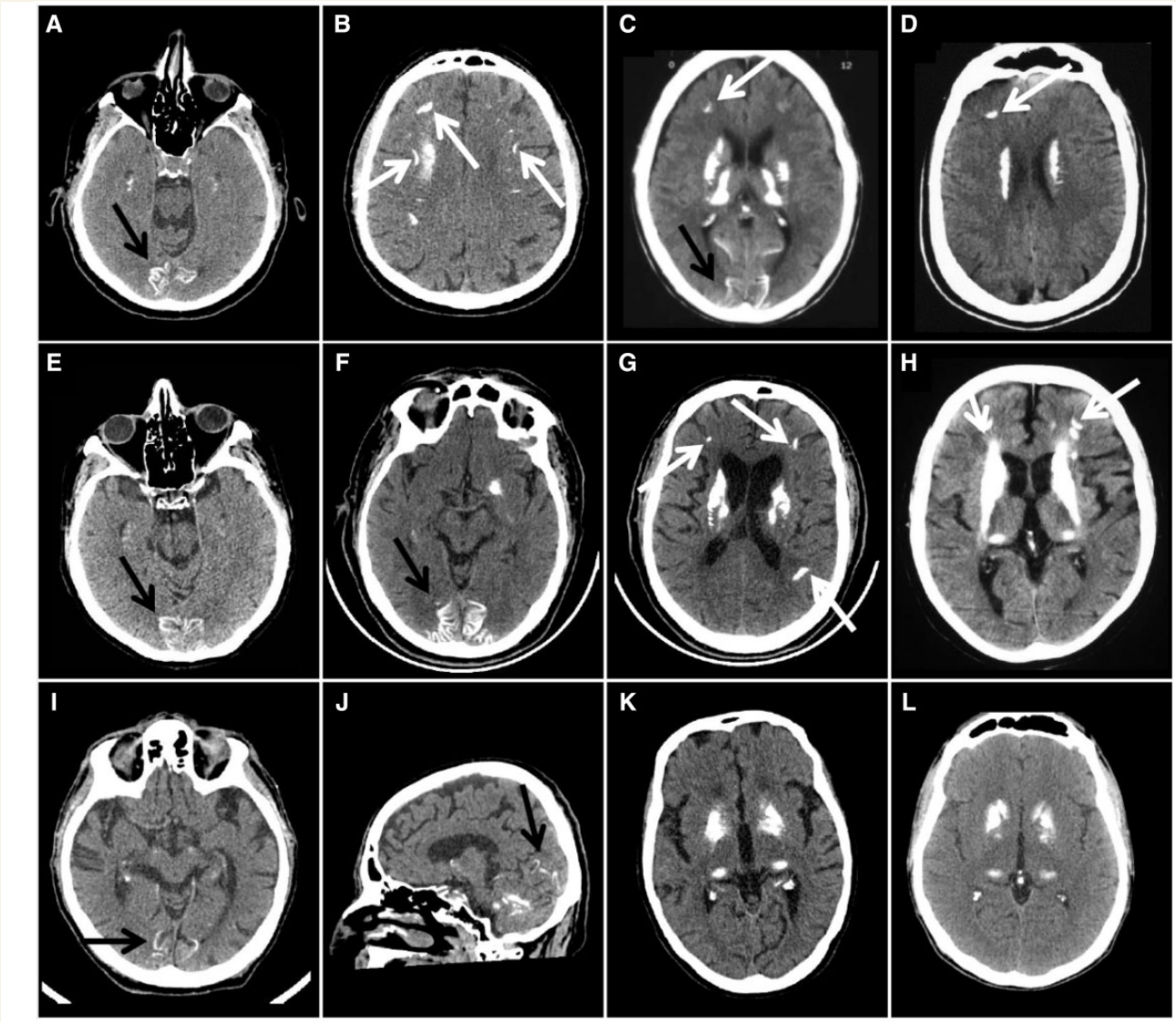

Figure 3 Cortical calcifications on the CT scans of the patients with *SLC20A2* mutations, mainly occipital (black arrows), and/or in the depth of the sulci (white arrows): Patient 1, Family 752 (**A** and **B**); Patient 2, Family 752 (**C**); Patient 1, Family 1038 (**D**); Case 1352 (**E**); Case 485 (**F** and **G**); Case 424 (**H**); Case 413 (**I** and **J**). Absence of cortical calcification in two *PDGFRB* patients (**K** and **L**, Family 870) despite severe calcification of the basal ganglia (severe calcification of both lenticular nuclei, confluent with severe calcification of both caudate nuclei (upstream of these slides), in **K** and **L**, and severe calcification of both thalami in **K**, faint in **L**).

These children carried a maternally-inherited missense variant, in addition to the paternally-inherited loss-of-function mutation, leading to the hypothesis that a biallelic contribution could explain a more severe and an earlier phenotype (Wang *et al.*, 2012). We found that one third of our patients who were affected by an *SLC20A2* mutation were asymptomatic. Publicly available data suggest great intra-familial and inter-familial diversity for *SLC20A2* mutations. For example, in the Family BJ-IBGC (Wang *et al.*, 2012), only one of nine mutation carrier exhibited one of the symptoms listed by Manyam *et al.* (2001), whereas the nine mutation carriers of the American Family F1 exhibited at least one symptom, particularly among the movement disorders (Geschwind *et al.*, 1999; Hsu *et al.*, 2013).

In the present study, few clinical differences appeared between *PDGFRB* and *SLC20A2* mutation carriers, which could be confirmed in further studies using larger populations. Few patients are reported with *PDGFRB* mutations. Because of that and of the high diversity in age of clinical onset in both genetic groups, they cannot systematically be compared in terms of ages. The proportion of symptomatic individuals in both groups may be interpreted carefully, since the majority of *PDGFRB* patients belonged to one family and the mean age of last evaluation of *PDGFRB* patients tended to be lower than *SLC20A2* patients.

Migraine or cephalalgia is sometimes reported in IBGC cases and family reports. Caution is needed concerning the exact prevalence of cephalagia among patients with IBGC, because this symptom

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was not systematically investigated in previous reports, and because of the large prevalence of migraine and cephalalgia in the general population. It was not reported in the previous case series (Manyam *et al.*, 2001), and it was present in four of the nine patients in the SD-IBGC family (*SCL20A2* mutation) (Dai *et al.*, 2010). Migraine was reported as a predominant sign in 15 of our patients. Nevertheless, in the family affected by a *PDGFRB* mutation, migraine did not segregate with IBGC. It would be necessary to perform a systematic case control study about cephalalgia in IBGC to address this issue.

Imaging

This study allows for the identification of differences in brain calcification between individuals with a *PDGFRB*, an *SLC20A2*, and no known mutation. In particular, the expected TCS was higher in *SLC20A2* mutation carriers, with respect to age. No patient with a *PDGFRB* mutation exhibited cortical or vermis calcifications, contrary to a striking majority of the *SLC20A2* mutation carriers. Expression data as given for example by the microarray or *in situ* hybridization analyses of human brains, the Allen brain atlas (Sunkin *et al.*, 2013), are useful tools in the corroboration of these findings (da Silva *et al.*, 2013). Expression data about these two genes in the occipital cortex are not consistent between the analysed brains and further studies would be necessary. Conversely, it seems that the cerebellar expression of *PDGFRB* is mainly restricted to the cerebellar nuclei, whereas the expression of *SLC20A2* seems to be more widespread in the cerebellum, including the vermis. This observation is in accordance with the location and the extension of the calcifications identified in our series. Brainstem calcifications are rare. To our knowledge, only one patient with an *SLC20A2* mutation and calcification in the brainstem was reported, but the precise location was not mentioned (Hsu *et al.*, 2013), and we only found one *SLC20A2* mutation carrier with midbrain calcification. Five patients with probable IBGC had impressive calcifications in the pons. The causative gene(s) of such a pattern of calcification is unknown. It (they) might have a different pattern of brain expression compared to *SLC20A2* and *PDGFRB*, which seem to have relatively weak expression in this area (Allen brain atlas). Another hypothesis would be that a causative gene might have a similar pattern of expression, but with biallelic mutations (recessive pattern of inheritance), which could explain a more severe imaging phenotype with calcifications in the pons.

The mean severity of the calcifications was higher in symptomatic compared to asymptomatic individuals. Interestingly, none of our *SLC20A2* mutation carriers exhibited cerebellar signs, despite the presence of severe cerebellar calcifications. Nonetheless, cerebellar syndrome has already been reported in *SCL20A2* mutations carriers, even in patients without cerebellar calcifications (Hsu *et al.*, 2013). Taken together, these data suggest that the severity of BGC, which are a mandatory criterion for IBGC diagnosis, helps predict the symptomatic status, but is not sufficient. Other biomarkers would be necessary to better correlate with the clinical diversity of IBGC.

The prevalence of IBGC is unknown. Three main reasons could explain this: the unknown clinical penetrance, the absence of epidemiological study with exploration of the other causes of BGC, and the absence of threshold-value to exclude calcification which may be related to normal aging. Our criteria of probable IBGC excluded the patients with a TCS under the age-specific threshold in order to exclude phenocopies. Such a criterion was stringent. Besides this, the age-specific radiological penetrance is unknown, and one 46-year-old patient with a *PDGFRB* mutation presented a TCS under the age-specific threshold. This finding may support an incomplete radiological penetrance for calcifications related to a *PDGFRB* mutation. Nevertheless, in this patient, calcification was seen in more than just the lenticular nuclei (cerebellum). As a consequence, this stringent threshold, designed to select the probands, must be used with care when studying the segregation of genetic variants of unknown penetrance in pedigrees.

As expected, though never before demonstrated, the TCS correlated positively with age in patients with IBGC, suggesting an increase of the severity of IBGC-related brain calcifications during aging. To our knowledge, this question has not yet been addressed with a longitudinal cohort study. Interestingly, an increase in the volume of lenticular calcifications during the follow-up period was recently observed in 35 of 49 patients with hypoparathyroidism (Goswami *et al.*, 2012). In previous reports of IBGC, brain calcifications were mostly described in terms of gross location and, rarely, quantified by volumetric studies, which might have been biased by brain atrophy (Manyam *et al.*, 2001). Moreover, volumetric studies did not take into account the density of the calcifications (Manyam *et al.*, 2001; Goswami *et al.*, 2012), which seems particularly problematic to us for faint calcification. Our visual rating scale is likely not to be affected by cerebral atrophy, is easily applicable and adaptable for the everyday use by radiologists and neurologists, reflects the visual severity of the calcifications and has satisfactory inter-rater agreement. Livingston *et al.* (2013) used a scale designed to classify calcifications seen in CT scans of patients with diverse disorders, among the following categories: spot, rock, blush and linear. This study provided for a noteworthy description of brain neuroimaging in these disorders (e.g. Aicardi-Goutieres syndrome), including neuroimaging features other than calcification alone. Among the patients with no known diagnosis, two families (seven patients) with calcification resembling dominant Fahr's disease were classified apart from the others in light of the calcification patterns. We chose to elaborate our own scale designed specifically to describe calcification related to IBGC, and using a semi-quantitative method.

We found that 15% of the control patients presented BGC, all but one >40 years of age, suggesting that the presence of BGC in an individual <40 years of age may not be considered as physiological. The first large studies, performed before the 1990s, had initially estimated the prevalence of BGC to be 0.6–0.8% (Murphy, 1979; Cohen *et al.*, 1980; Harrington *et al.*, 1981) to 1.1% (Konig, 1989). The thickness of the slices and the quality of the CT scans, not comparable with current technology, could explain why punctate lenticular calcifications would have been largely underestimated. More recent studies had contradictive results, probably due to diverse methods (Bordignon and Arruda, 2002; Eskandary *et al.*, 2005). Interestingly, a recent study found

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a BGC frequency of 20.5% in unselected patients from Japan, and the older group (>65 years) had a higher frequency than the younger (31% versus 13%) (Yamada *et al.*, 2013). In accordance with our results and the previously mentioned work, the studies focusing on older individuals showed that BGC is a common finding on systematic brain CT scans (Ostling *et al.*, 2003; Simoni *et al.*, 2008). Moreover, our study presents the first semi-quantitative data of brain calcification in an unselected series of controls, with interesting results concerning the prevalence and proposed age-specific thresholds, which have to be replicated in other studies.

Genetics

We studied more patients with a mutation in *PDGFRB* (due to the identification of a large family) than in *SLC20A2*. Nevertheless, the latter gene may be more frequently involved in familial IBGC. Indeed, a previous study reported *SLC20A2* mutations in 12 of 29 (41%) of families with IBGC (Hsu *et al.*, 2013). Considering only families with IBGC, we identified one family with a *PDGFRB* mutation, two with an *SCL20A2* mutation and 12 remained unexplained. If adding the cases with a sporadic presentation, we found three probands with a *PDGFRB* mutation and six with an *SLC20A2* mutation, where 29 did not have a genetic alteration in these two genes, underlining the need to pursue our effort to find further causative genes.

Some patients with a sporadic presentation were classified as having definite IBGC. It was not possible to ascertain the absence of heredity for all of them, because of the lack of brain imaging in their first-degree relatives. Indeed, the absence of neuropsychiatric signs in first-degree relatives by history is not sufficient to exclude IBGC, because of an incomplete clinical penetrance.

Conclusion

The diagnosis of IBGC is based first on neuroradiological findings and a significant proportion of individuals are asymptomatic. These observations, together with the high frequency of fortuitous discovery of BGC of unknown significance on brain CT scans, made IBGC, formerly called Fahr's disease, controversial as a distinct disorder with clinical significance. The ongoing identification of the molecular bases of IBGC demonstrates the high value of IBGC as a patent phenotypical marker. It also confirms the genetic heterogeneity of the disorder (Oliveira *et al.*, 2004). The number of affected individuals with definite IBGC and advanced age is still insufficient to determine the exact clinical penetrance of the disease. We show here that the mean severity of the calcifications is higher in symptomatic versus asymptomatic individuals, but the quantification of the calcifications seems insufficient to predict the clinical severity. A next step will be to investigate asymptomatic and symptomatic patients of advanced age with the same mutation to identify other biomarkers of the clinical severity of the disease. Moreover, the radiological penetrance, thought to be near 100% at the age of 50 (Sobrido *et al.*, 1993–2004) because of the neuroradiological definition of the disease, has to be reinvestigated in the future, as suggested by our data. As in many other neurogenetic disorders, the clinical penetrance is incomplete, rendering both genetic counseling and future therapeutic trials challenging.

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Supplementary material

Supplementary material is available at *Brain* online.

References

Baba Y, Broderick DF, Uitti RJ, Hutton ML, Wszolek ZK. Heredofamilial brain calcinosis syndrome. Mayo Clin Proc 2005; 80: 641–51.

Bordignon KC, Arruda WO. CT scan findings in mild head trauma: a series of 2,000 patients. Arq Neuropsiquiatr 2002; 60: 204–10.

Cohen CR, Duchesneau PM, Weinstein MA. Calcification of the basal ganglia as visualized by computed tomography. Radiology 1980; 134: 97–9.

da Silva RJ, Pereira IC, Oliveira JR. Analysis of gene expression pattern and neuroanatomical correlates for SLC20A2 (PiT-2) shows a molecular network with potential impact in idiopathic basal ganglia calcification ("Fahr's Disease"). J Mol Neurosci 2013; 50: 280–3.

Dai X, Gao Y, Xu Z, Cui X, Liu J, Li Y, et al. Identification of a novel genetic locus on chromosome 8p21.1-q11.23 for idiopathic basal ganglia calcification. Am J Med Genet B Neuropsychiatr Genet 2010; 153B: 1305–10.

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Delacour A. Ossification des capillaires du cerveau. Ann Med Psychol 1850; 2: 458–61.
Eskandary H, Sabba M, Khajehpour F, Eskandari M. Incidental findings in brain computed tomography scans of 3000 head trauma patients. Surg Neurol 2005; 63: 550–3. discussion 3.
Geschwind DH, Loginov M, Stern JM. Identification of a locus on chromosome 14q for idiopathic basal ganglia calcification (Fahr disease). Am J Hum Genet 1999; 65: 764–72.
Godefroy O, Azouvi P, Robert P, Roussel M, LeGall D, Meulemans T. Dysexecutive syndrome: diagnostic criteria and validation study. Ann Neurol 2010; 68: 855–64.
Godefroy O, Roussel M, Leclerc X, Leys D. Deficit of episodic memory: anatomy and related patterns in stroke patients. Eur Neurol 2009; 61: 223–9.
Goswami R, Sharma R, Sreenivas V, Gupta N, Ganapathy A, Das S. Prevalence and progression of basal ganglia calcification and its pathogenic mechanism in patients with idiopathic hypoparathyroidism. Clin Endocrinol (Oxf) 2012; 77: 200–6.
Harrington MG, Macpherson P, McIntosh WB, Allam BF, Bone I. The significance of the incidental finding of basal ganglia calcification on computed tomography. J Neurol Neurosurg Psychiatry 1981; 44: 1168–70.
Hsu SC, Sears RL, Lemos RR, Quintans B, Huang A, Spiteri E, et al. Mutations in SLC20A2 are a major cause of familial idiopathic basal ganglia calcification. Neurogenetics 2013; 14: 11–22.
Konig P. Psychopathological alterations in cases of symmetrical basal ganglia sclerosis. Biol Psychiatry 1989; 25: 459–68.
Kostic VS, Lukic-Jecmenica M, Novakovic I, Dobricic V, Brajkovic L, Krajinovic M, et al. Exclusion of linkage to chromosomes 14q, 2q37 and 8p21.1-q11.23 in a Serbian family with idiopathic basal ganglia calcification. J Neurol 2011; 258: 1637–42.
Le Ber I, Marie RM, Chabot B, Lalevee C, Defer GL. Neuropsychological and 18FDG-PET studies in a family with idiopathic basal ganglia calcifications. J Neurol Sci 2007; 258: 115–22.
Lemos R, Oliveira M, Oliveira J. Reporting a new mutation at the SLC20A2 gene in familial idiopathic basal ganglia calcification. Eur J Neurol 2013; 20: e43–4.
Liu X, Jian X, Boerwinkle E. dbNSFP: a lightweight database of human nonsynonymous SNPs and their functional predictions. Hum Mutat 2011; 32: 894–9.
Livingston JH, Stivaros S, van der Knaap MS, Crow YJ. Recognizable phenotypes associated with intracranial calcification. Dev Med Child Neurol 2013; 55: 46–57.
Lopez-Villegas D, Kulisevsky J, Deus J, Junque C, Pujol J, Guardia E, et al. Neuropsychological alterations in patients with computed tomography-detected basal ganglia calcification. Arch Neurol 1996; 53: 251–6.
Manyam BV. What is and what is not 'Fahr's disease'. Parkinsonism Relat Disord 2005; 11: 73–80.
Manyam BV, Walters AS, Narla KR. Bilateral striopallidodentate calcinosis: clinical characteristics of patients seen in a registry. Mov Disord 2001; 16: 258–64.
Murphy MJ. Clinical correlations of CT scan-detected calcifications of the basal ganglia. Ann Neurol 1979; 6: 507–11.
Nicolas G, Guillin O, Borden A, Bioux S, Lefaucheur R, Hannequin D. Psychosis revealing familial idiopathic basal ganglia calcification. Gen Hosp Psychiatry 2012, in press [Epub ahead of print].
Nicolas G, Pottier C, Maltete D, Coutant S, Rovelet-Lecrux A, Legallic S, et al. Mutation of the PDGFRB gene as a cause of idiopathic basal ganglia calcification. Neurology 2013; 80: 181–7.
Oliveira JR, Spiteri E, Sobrido MJ, Hopfer S, Klepper J, Voit T, et al. Genetic heterogeneity in familial idiopathic basal ganglia calcification (Fahr disease). Neurology 2004; 63: 2165–7.
Ostling S, Andreasson LA, Skoog I. Basal ganglia calcification and psychotic symptoms in the very old. Int J Geriatr Psychiatry 2003; 18: 983–7.
Sarazin M, Berr C, De Rotrou J, Fabrigoule C, Pasquier F, Legrain S, et al. Amnestic syndrome of the medial temporal type identifies prodromal AD: a longitudinal study. Neurology 2007; 69: 1859–67.
Schottlaender LV, Mencacci N, Koepp M, Hanna M, Hardy J, Lees AJ, et al. Interesting clinical features associated with mutations in the SLC20A2 gene (abstract). Eur J Neurol 2012; 19 (Suppl 1): 22–89.
Simoni M, Pantoni L, Pracucci G, Palmertz B, Guo X, Gustafson D, et al. Prevalence of CT-detected cerebral abnormalities in an elderly Swedish population sample. Acta Neurol Scand 2008; 118: 260–7.
Sobrido MJ, Hopfer S, Geschwind DH. Familial idiopathic basal ganglia calcification. In: Pagon RA, Bird TD, Dolan CR, editors. GeneReviews™ [Internet]. Seattle, WA: University of Washington; 1993–2004.
Sunkin SM, Ng L, Lau C, Dolbeare T, Gilbert TL, Thompson CL, et al. Allen Brain Atlas: an integrated spatio-temporal portal for exploring the central nervous system. Nucleic Acids Res 2013; 41: D996–1008.
Trautner RJ, Cummings JL, Read SL, Benson DF. Idiopathic basal ganglia calcification and organic mood disorder. Am J Psychiatry 1988; 145: 350–3.
Ver Hoef JM, Boveng PL. Quasi-Poisson vs. negative binomial regression: how should we model overdispersed count data? Ecology 2007; 88: 2766–72.
Wang C, Li Y, Shi L, Ren J, Patti M, Wang T, et al. Mutations in SLC20A2 link familial idiopathic basal ganglia calcification with phosphate homeostasis. Nat Genet 2012; 44: 254–6.
Yamada M, Asano T, Okamoto K, Hayashi Y, Kanematsu M, Hoshi H, et al. High frequency of calcification in basal ganglia on brain computed tomography images in Japanese older adults. Geriatr Gerontol Int 2013; 13: 706–10.
Yamada N, Hayashi T. Asymptomatic familial basal ganglia calcification with autosomal dominant inheritance: a family report [Article in Japanese]. No To Hattatsu 2000; 32: 515–9.
Zeileis A, Kleiber C, Jackman S. Regression models for count data in R. J Stat Softw 2008; 27: 1–25.
Zhang Y, Guo X, Wu A. Association between a novel mutation in SLC20A2 and familial idiopathic basal ganglia calcification. PLoS One 2013; 8: e57060.

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