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Iron accumulation in deep brain nuclei in migraine: A population-based Magnetic Resonance Imaging study

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Abstract

Background—A small MRI study showed increased iron depositions in the periaqueductal grey matter in migraineurs, suggestive of a disturbed central antinociceptive neuronal network.

Procedures—With 1.5T MRI, we assessed iron concentrations in seven deep brain nuclei in a large population-based cohort. We compared T2 values between migraineurs (n=138) and controls (n=75), with multivariate regression analysis. Analyses were conducted in age strata (<50, n=112; ≥50) because iron measures are increasingly influenced by non-iron related factors in the older group.

Findings—Overall, migraineurs and controls did not differ, nor did migraineurs with vs. without aura. In the younger migraineurs compared to controls, T2-values were lower in the putamen (p=0.02), globus pallidus (p=0.03) and red nucleus (p=0.03). Similarly, in these younger migraineurs, controlling for age, those with longer migraine-history had lower T2 values in the putamen (p=0.01), caudate (p=0.04) and red nucleus (p=0.001).

Conclusions—Repeated migraine attacks are associated with increased iron concentration / accumulation in multiple deep nuclei that are involved in central pain processing and migraine pathophysiology. It remains unclear whether iron accumulation in the antinociceptive network has a causative role in the development of (chronic) migraine headache.

Keywords

Migraine; Magnetic Resonance Imaging; Iron accumulation; Pain processing

Introduction

Migraine is a common, multifactorial neurovascular brain disorder, typically characterized by recurrent attacks of disabling headache and associated symptoms of autonomic nervous system dysfunction (migraine without aura; MO); in up to one-third of patients, attacks are also associated with transient focal neurological aura symptoms (migraine with aura; MA) (1). The median attack frequency is 1.5 per month (2); ten to twenty percent of patients show progression of the attack frequency up to daily or nearly daily attacks (chronification to chronic daily headache) (3;4). Episodic dysfunction of brain stem pathways, involved in

the control of pain and other sensory modalities, has been implicated in the pathogenesis of the migraine headache and associated autonomic symptoms (5;6).

Using magnetic resonance imaging (MRI) in two small studies, Welch and colleagues found increased iron levels in the periaqueductal gray matter (PAG) and red nucleus of patients who were attending a headache clinic for migraine or chronic daily headache (7). As the PAG is activated during migraine attacks (8–12), it was hypothesized that free-radical cell damage associated with hyperemia during repeated migraine attacks may lead to iron deposition. This would reflect a progressive dysfunction of the PAG (7) and other brainstem structures, leading to interruption of normal antinociceptive function, and thus contributing to migraine headaches (6). As the study was performed in a small and selected group of clinic based, and thus presumably severe migraine patients, it is unclear to what extent these findings reflect general processes in migraine or only changes occurring at the severe end of the clinical spectrum.

On MR images the age related accumulation of non-heme iron in the brain gives rise to decreased signal intensity in T2-weighted images in the globus pallidus, substantia nigra, red nucleus and putamen. The relationship between iron concentration and T2 relaxation times permits quantitative assessment of differences in iron concentration in the brain in vivo. In the present study we examined iron concentration in seven different deep brain nuclei with MRI in migraine cases and controls who participated in a population based study. Iron deposition was also correlated to migraine subtype, number of attacks experienced, and duration of migraine history.

Methods

Study population

The Dutch population-based Genetic Epidemiology of Migraine (GEM) study and its substudy, the Cerebral Abnormalities in Migraine, an Epidemiological Risk Analysis (CAMERA) MRI Study have been described previously (2;13). In brief, from a population-based survey of 6491 Dutch adults aged 20–60 years living in two representative Dutch municipalities (Maastricht and Doetinchem) 863 migraineurs and 5628 controls were identified according to the IHS-criteria (1). From those aged 30–60 yrs, we randomly selected for the MRI study 134 migraineurs without aura (MO), 161 with aura (MA) and 140 controls, frequency matched by sex, five year age strata and place of residence. Neither cases and controls, or responders (69%) and non-responders, differed by age, sex, and cardiovascular risk factors. All participants gave written informed consent and participated without any financial reimbursement. The study protocol was approved by the ethics committees of the cooperating institutions and included a structured telephone interview and a clinic visit for blood drawn, a standard physical and neurological examination and a brain MRI study. Since the Doetinchem participants (n=222) did not have the images available on Magneto Optical Disks, they were excluded from the current analyses, giving 213 Maastricht participants available for analysis (138 migraineurs, 75 controls).

Assessment of confounders, covariates and migraine characteristics

Sociodemographic, medical and migraine characteristics were assessed by interview. Education was categorized into low (primary school or lower vocational education) and high. Self-reported weight and height were used to calculate body mass index (weight in kilograms divided by the square of height in meters). Blood pressure was the mean of three measurements obtained at 1-minute intervals in the upper arm with an electronic oscillometric BP monitor (OMRON 711, Omron Healthcare Europe, Hoofddorp, the Netherlands). Hypertension was defined as a systolic blood pressure of 160 mm Hg and

higher or a diastolic blood pressure of 95 mm Hg and higher or current use of antihypertensive drugs. A measure of total cholesterol was available from the base-line examination (14). Smoking history was defined as never, former and current, and for ever smokers, pack-years of exposure was calculated. As previously detailed, migraine cases estimated headache and aura attack frequency, and frequency and amount of specific anti-migraine medication (ergotamines, triptans) they used in the years they had migraine attacks (13). There were no differences in demographic characteristics and cardiovascular risk factors between the included versus the excluded participants.

MRI

Brain MRI images were acquired on a 1.5-T unit (ACS-NT, Philips Medical Systems, Best, The Netherlands). Whole brain images were acquired with 48 contiguous 3 mm axial slices (field-of-view, 22 cm; matrix, 192–205 by 256). Pulse sequences included a combined proton density (PD) and T2-weighted fast spin-echo sequence (3000/27–120ms; relaxation time/echo time) and fluid-attenuated inversion-recovery sequence (8000/100/2000ms; relaxation time/echo time/inversion time). Methods used for rating of deep and periventricular white matter lesions and infarct-like lesions on hard copies have been described previously (13).

Quantitative T2 measurements

For the current quantitative analyses, digital MRI data was postprocessed on a commercially available workstation (EasyVision release 4.3, Philips Medical Systems, Best, The Netherlands). One observer (J.O.) measured signal intensities (SI), blinded to all clinical data. Region of Interest (ROI)-measurements of signal intensities were performed bilaterally within the following anatomical structures (Figure 1) by manually outlining the putamen, posterior putamen, globus pallidus and red nucleus, and by placing a standard-sized circle over the head of caudate nucleus (30 mm²) and substantia nigra (pars reticularis and pars compacta [10 mm²]). For the posterior part of the putamen, we used a standard area of 45 mm². All signal intensities were measured in the slice(s) that best visualized the structure. The putamen, caudate nucleus and globus pallidus were measured in two adjacent slides. ROI were drawn on the PD-images, copied and then pasted on the corresponding T2-images.

Mean signal intensity values of all ROI were noted. For each structure we calculated an average signal intensity by taking the means of measurements of both sides and/or adjacent slides was calculated. T2-values were then calculated using the expression $T2 = (TE_2 - TE_1) / [\ln(S_1/S_2)]$, where S_1 and S_2 are the signal intensities in the early- (=PD) and late-echo (=T2) images, with echo times TE_1 (=27ms) and TE_2 (=120ms) respectively. To determine the intra-observer variability, six weeks after the initial analysis was completed, the same observer reanalyzed 21 randomly selected MRI-examinations, using the same protocol. Intraclass Correlation Coefficients (ICC) were calculated for each anatomical structure and are shown in Table 1.

Statistics

χ^2 and t-tests were used to test for differences between cases and controls in sociodemographic and migraine characteristics, cardiovascular risk factors and prevalence of lesions on MRI. Linear regression analyses controlling for age were used to test for differences across groups in the distributions and means of measured T2 values among the study groups. From the literature it is known that T2 values of deep nuclei are significantly influenced by non-iron-related tissue changes after the age of 50 years (15;16). Therefore, both to eliminate the confounding effect of such changes, and to evaluate the changes of T2-values as a function of age, we split the total sample at the median age of 50 yrs, and performed sub analyses, grouping those <50 yrs of age (n=112) vs. those ≥50 yrs of age

(n=101). In addition, to assess the influence of migraine subtype, attack frequency and duration of migraine history on the reduction of T2-values, we analysed the migraineurs separately, and respectively split them in MO (n=80) vs. MA (n=58), in those with <1 attack/month (n=77) vs. those with ≥ 1 attack/month (n=61), and in those with a total migraine-history of <23 yrs (split at the median; n=68) vs. ≥ 23 yrs (n=70). All models controlled for age. Analyses were conducted with SPSS statistical software (version 12.0.1; SPSS Inc., Chicago, IL).

Role of the funding source

The sponsor of the study had no role in study design, data collection, data analysis, data interpretation, or writing of the report. The corresponding author had full access to all the data in the study and final responsibility for the decision to submit for publication.

Results

In the total sample, there were no differences in the variables between migraineurs and controls, except for a higher body mass index in the migraine subgroup (table 2). In those <50 years of age, migraineurs had lower systolic blood pressure (125.9 ± 1.6 vs. 133.5 ± 2.6 mm Hg; age adjusted $p=0.01$); in those >50 years of age, smoking was more prevalent in controls (21.4 ± 1.2 vs. 13.0 ± 2.1 pack years; age adjusted $p=0.03$). Regardless of age-group, prevalence of infarcts and prevalence of higher load of white matter lesions was similar in migraineurs and controls. Prevalence of higher vs. lower attack frequency did not differ between younger vs. older participants, nor between MO vs. MA.

In the whole sample, for all examined deep brain nuclei the mean T2-values did not differ between migraineurs and controls, both in the crude and in the multivariate analyses controlling for age (Table 3). Similarly, no differences in T2-values were found between male and female participants. A trend towards lower mean T2 values in the posterior part of the putamen was found in migraineurs compared to controls.

Among migraineurs, no differences were found between those with (MA; n=58) and those without aura (MO; n=80). In those with higher (n=61) vs. those with lower (n=77) mean attack frequency (<1 attack/month vs. ≥ 1 attack/month), mean T2-values were consistently slightly lower, but none of these differences reached statistical significance; the strongest effect was found in the red nucleus (77.7 ± 1.2 vs. 80.6 ± 1.3 ms; $p=0.1$). Among MA, this trend remained for the red nucleus (77.4 ± 1.7 vs. 81.0 ± 1.7 ms; $p=0.1$). Among MO, a trend towards lower T2-values was found in the posterior putamen in those with higher attack frequency (70.8 ± 0.6 vs. 72.1 ± 0.5 ms; $p=0.11$).

Among migraineurs, in the younger subjects compared to those ≥ 50 yrs of age, significantly lower T2 values were measured in the red nucleus (77.1 ± 8.0 vs. 81.6 ± 12.2 ms; $p=0.01$); for other structures no significant differences were found. Among controls, lower T2 values were measured in those ≥ 50 yrs of age in the putamen (74.8 ± 1.8 vs. 73.8 ± 2.5 ms; $p=0.04$), the head of the caudate nucleus (83.2 ± 2.4 vs. 81.5 ± 3.0 ms; $p=0.008$) and globus pallidus (73.7 ± 4.1 vs. 72.0 ± 3.9 ms; $p=0.07$, trend).

Among those <50 years of age, both in the crude and in the age-controlled analyses, significantly lower T2-values were measured in the posterior part of the putamen, the globus pallidus and the red nucleus of migraineurs compared to controls (Table 3). No differences were found between male and female participants <50 yrs of age. Among this subgroup of younger migraineurs, no differences were found between MO (n=33) and MA (n=36). In those with higher (n=31) vs. lower (n=38) mean attack rate, T2-values were consistently slightly lower for all nuclei, but not significantly different (again most pronounced in the red

nucleus [75.6 ± 0.8 vs. 78.6 ± 1.6 ms; $p=0.16$], and among MA with a slightly larger difference [75.3 ± 1.2 vs. 80.0 ± 2.7 ms; $p=0.16$]). Significantly lower T2-values were found in migraineurs <50 yrs of age with longer duration of migraine-history ($n=34$) vs. shorter duration, in the putamen, the head of the caudate and the red nucleus (Table 4).

In the older subgroup (age ≥ 50 years), no differences were found between controls and migraineurs, nor between subgroups of migraineurs.

Discussion

In this population based MRI study, we found evidence for increased local iron concentrations (reduced T2-values) in the putamen, globus pallidus and red nucleus of migraineurs <50 yrs. These data confirm and extend the clinic-based findings by Welch et al. (7). Controlling for age, we found significant differences between migraineurs and controls, and significant effects of (longer) duration of migraine history on T2 values. Consistent with the previous study on iron accumulation in migraineurs, we did not find differences between migraineurs with and without aura. These data suggest that not the aura pathophysiology, but merely pain processes during repetitive migraine attacks are likely involved.

Differences in methodology between the study by Welch et al. and ours have to be noted. First, they used T2 gradient echo imaging sequences on a 3.0 T scanner, whereas we applied turbo spin echo sequences on a 1.5 T scanner. Both our type of sequence and our lower field strength results in lower sensitivity to local susceptibility effects caused by iron (17); this suggests that our findings would probably have been even more clear using a more sensitive technique. Further we used regions of interest to measure signal intensities, whereas Welch et al. applied a semi automated brain tissue segmentation algorithm to identify separate types of tissue. Using that technique, they segmented brain regions, including the PAG, and measured T2 characteristics. We felt we were unable to select the PAG manually, without including the hyperintense signal from the CSF in the cerebral aqueduct itself, and therefore we could not confirm that part of the data directly. However, our data on other deep brain nuclei adds to the findings by Welch et al. and points to the possibility that impairment of iron homeostasis in migraineurs is not limited to the PAG. Although manually drawing ROI (by an observer blinded to the diagnosis) might introduce some observer-variability, we found for most structures the ICC values were fairly high. Only the ICC regarding the SNpr was low (0.63), likely to be explained by the very small size of the ROI, in an area with relatively many different signal intensities. In both studies, thin (3 mm) adjacent slices resulted in a sufficient number of slices including all structures and minimized partial volume effects.

Iron depositions – relevance to migraine pathophysiology

Iron is essential in oxygen carrier proteins and in many metabolic enzymes. Up to 25% of total oxygen consumption of the body is by brain metabolic processes, which implicates a relatively high utilization of iron (18;19). Non-heme iron is primarily stored inside the non-toxic ferritin protein, but can also be present in an unshielded, soluble form. This iron can become toxic since it can catalyze the formation of free radicals, that can lead to DNA, protein and neuronal cell damage (19).

In normal aging iron accumulates throughout the brain, and particularly in the basal ganglia (20). Several neurodegenerative diseases, including Hallervorden-Spatz syndrome, Parkinson's disease and Huntington's disease, are associated with errors in iron metabolism, transport and homeostasis (21), affecting more specific regions in the brain and basal ganglia, and likely affecting the disease process (22;23). Excessive iron accumulation can

directly cause basal ganglia dysfunction, eg. by damaging synapses or modulating protein synthesis, leading to either increase or decrease local levels of neurotransmitters. Indirectly, a mechanism of increased iron concentrations rendering the brain more vulnerable to oxidative stress has been suggested in several CNS diseases (18), including Alzheimer's disease and multiple sclerosis (22).

In line with these pathophysiological possibilities, the observed increased iron concentration in the PAG in the earlier studies was interpreted to be a possible result from an impaired iron homeostasis, possibly related to iron-catalyzed free radical injury during repeated episodes of hyperoxia during migraine attacks, and thus as a result ("burden") of migraine attacks (7). The episodic activation of the PAG has indeed been visualized with PET and functional MRI during spontaneous (8), triggered (9–11) and chronic (12) migraine, as well as in experiments of acute capsaicin-evoked pain in non-migraineurs (24). Similarly, activation (hyperemia) of the RN also occurs during migraine (25) as well as in non-migraine pain experiments (24). Also, activation in the lentiform nucleus (putamen and/or globus pallidus) was visualized during migraine (10) but is also described as part of normal central pain-processing (24). PAG, red nucleus, and lentiform nuclei are thus all involved in the normal, complex, central nociceptive network (sensory-motor integration) (24).

Our finding of increased iron accumulation in multiple nuclei involved in central pain processing, makes it less likely that the PAG is "the only" relevant deep grey matter and/or brainstem structure in the pathophysiology of migraine headache. The current findings, including the observation of higher concentration of iron in those with longest migraine history, favor the concept that repeated activation of several nuclei during migraine attacks (or "pain" in general) occurs, and is associated with increased iron accumulation or concentration. However, the findings from this and earlier studies are based on cross-sectional observation and cannot study temporality. Longitudinal data are needed to justify such conclusion, and the fact that migraine prevalence generally decreases with increasing age also argues against such mechanism. Similarly, it is speculative to state that an impaired iron homeostasis is a factor in the migraine pathophysiology; the increased concentrations might be just a normal physiological response or marker of increased cellular function, and could even be reversible.

If increased iron concentrations in a chain of deep grey nuclei would play a causative role in the migraine pathophysiology, and eg. by free-radical cell damage, secondary injury to these nuclei might occur, this could theoretically lead to progression of the disease. This might explain the observation that in a minority of migraine patients an increase in attack frequency from episodic to chronic (chronification) occurs.(3;4;26;27). This could then reflect a defective central pain processing system, rather than just one defective link in the chain (as the PAG earlier was suggested to be). The "sensory dysmodulation concept" of migraine pathophysiology (trigeminal somatosensory pathway), in which normal afferent stimuli are misperceived as being excessive (6), might then be explained by dysfunction of more than one deep nucleus. However, other explanations for this concept have been suggested, including a relationship with dysfunctional P/Q-type calcium channels in specific structures like the PAG (28;29). Although most of the above remains speculative, we know that structural changes in brainstem regions can act as a cause of migraine (eg. a bleed in a cavernoma (30), or hyperintense (eg. multiple sclerosis) changes (31;32)), and pathologic iron accumulation might act similarly.

MRI measurement of iron deposition

Although measurement of brain iron concentration is only possible post mortem, decreasing T2 values in MRI have been strongly correlated with age-related regional accumulation of iron (20). Age-dependent curves of T2 relaxation time in the extrapyramidal nuclei were

found to be very consistent with published curves of non-heme iron concentrations in post mortem histology (15;33). In this way, it is highly probable that changes in T2 relaxation time result to a large extent from non-heme iron deposition. Up to 90% of non-heme iron in the brain is in the iron-stage protein ferritin (20). Regions with high ferritin content produce a local magnetic field gradient and inhomogeneities of the magnetic field, which decreases the T2 relaxation time of tissue.

Age-dependent T2 relaxation curves differ for different nuclei. Already in childhood and early adulthood MRI showed (exponentially) increasing degrees of hypointensity in the substantia nigra and red nucleus up to the age of 30 years, and globus pallidus up to the age of 50 years, which was confirmed in histological studies (20;34;35). In the putamen the curve decreases slower (flatter), and a plateau is reached at age 50–60. Above age 50, T2 values of different structures deviate more widely with greater individual variation (15). Local structural changes such as atrophy, enlargement of Virchow-Robin spaces or microscopic lacunar infarcts result in higher local water content resulting in increased T2 values; microcalcification may decrease T2 values (16). Knowing that after the age of 50 years the T2 values are increasingly influenced by non-iron-related factors, we separately analyzed those below and above age 50. The effect of various age-related factors that can increase mean T2 values (“white areas in dark nuclei”) probably explains why we did not find significant differences after age 50 between migraineurs and controls. However, we can not exclude the possibility that after age 50, when prevalence of migraine attacks regularly decreases, non-heme iron concentration in deep grey nuclei decreases. If this is the case, it would plead against deposition of extracellular iron into tissue during active migraine, but rather imply that there could be a functional increase of cellular iron concentration, as long as there is regular (migraine induced) activation of involved nuclei.

The current population-based evidence of increased iron-concentration in multiple deep brain nuclei involved in central pain processing in migraine patients with and without aura, illustrates the complex nature of migraine as a disorder of sensory dysmodulation, in which not only the PAG is a factor. The higher concentration of iron in multiple nuclei in migraineurs compared to controls of similar age, is likely disease related. It remains unclear whether the increased iron concentration is just a physiological response induced by repeated activation of nuclei involved in central pain processing, or whether the increased iron concentration could also damage these structures secondarily due to oxidative stress, and could be related to chronification of the disease in some migraine patients. From earlier studies, there is increasing evidence that migraine is associated with structural brain changes on MRI, including hyperintense cerebral white matter lesions (13;36), cerebellar and brainstem hyperintensities (37), and clinical (38) and subclinical infarcts (13;39). The risk of such brain lesions increases with increasing attack frequency. A similar effect seems to play a role with respect to iron concentration in deep grey nuclei.

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Figure 1.

Proton Density images (3,000/27 [repetition time/echo time]), showing ROI locations at the level of the basal ganglia (left) and the brain stem (right). ROI were placed by outlining the putamen (left image, red), globus pallidus (left image, green), the posterior putamen (left image, orange filling in the putamen) and the red nucleus (right image, red), as well as placing standard sized circles in the head of the caudate nucleus (left image, blue) and the substantia nigra (pars compacta (right image, green) and pars reticularis (right image, yellow).

Table 1

Mean T2-values (ms) of two measurements, ICC and 95% Confidence Interval (CI) per structure

Structure	T2		Reliability	
	M1 [†]	M2 [†]	ICC	95% CI [‡]
Putamen	74.1 ± 2.7	74.3 ± 2.7	0.98	0.96–0.99
Posterior Putamen	71.2 ± 2.9	71.0 ± 3.1	0.86	0.69–0.94
Caudate Nucleus	82.5 ± 2.9	81.9 ± 3.3	0.81	0.60–0.92
Globus Pallidus	71.6 ± 3.8	70.8 ± 3.5	0.85	0.68–0.94
SN pars reticularis	70.8 ± 5.8	70.0 ± 5.0	0.63	0.28–0.83
SN pars compacta	81.4 ± 4.3	81.5 ± 4.5	0.80	0.58–0.91
Red Nucleus	75.8 ± 4.3	75.8 ± 5.1	0.92	0.81–0.97

* Data are estimated mean (ms) ± SD; SN = Substantia Nigra

[†] M1=1st measurement; M2= 2nd measurement

[‡] All p-values were < 0.001

Table 2Characteristics of Study Participants^{*}

Characteristic	All	
	Control (n=75)	Migraine (n=138)
Sociodemographic		
Age (years)	48.5 (0.9)	49.0 (0.7)
Female	68%	70%
Low education	49%	55%
Clinical		
Body mass index (kg/m ²)	24.3 (0.4)	25.7 (0.4) [†]
Bloodpressure		
Systolic (mmHg)	136.2 (2.1)	133.1 (1.5)
Diastolic (mmHg)	91.1 (1.1)	90.9 (0.9)
Hypertension	35%	40%
Cholesterol (mmol/l)	5.3 (0.1)	5.3 (0.1)
Diabetes	2.7%	0.7%
Smoking		
Never	33%	30%
Pack years	14.7 (2.0)	11.9 (1.2)
Migraine characteristics		
Average attack frequency (/yr)	-	17.5 (1.7)
Duration migraine history (yrs)	-	23.5 (1.0)
Other brain damage		
Any infarct	5%	8%
High PVWML-load	11%	12%
High DWML-load	15%	21%

^{*} Data are mean (SE) or percentage of subjects, based on the (unpaired) t-test for the comparison of means and Pearson's χ^2 test for the comparison of proportions. Unless stated otherwise, differences were not statistically significant. Low education = primary school or lower vocational education; High PVWML = high load of periventricular white matter lesions, ie. PVWML with score >2 out of max. 6; High DWML-load = High load of deep white matter lesions, ie. the upper 20th percentile of the total DWML volume distribution;.

[†] P < 0.05

Mean T2-values (ms $\pm$ SE) of subjects with and without migraine history (n=213)*

Structure	All participants			Age < 50 years			Age ≥ 50 years			
	Controls (n=75)	Migraine (n=138)	<i>p-value</i> <i>crude</i> <i>adjusted</i>	Controls (n=43)	Migraine (n=69)	<i>p-value</i> <i>crude</i> <i>adjusted</i>	Controls (N=32)	Migraine (N=69)	<i>p-value</i> <i>crude</i> <i>adjusted</i>	
Putamen	74.4 ± 2.2	74.1 ± 2.7	0.6	74.8 ± 1.8	74.0 ± 2.3	0.06	73.8 ± 2.5	74.3 ± 3.1	0.4	0.3
Posterior Putamen	72.2 ± 3.2	71.5 ± 3.6	0.14	72.4 ± 2.8	71.0 ± 2.8	0.02	72.0 ± 3.8	71.9 ± 4.2	0.9	0.9
Head of Caudate	82.4 ± 2.8	82.4 ± 2.5	0.9	83.2 ± 2.4	82.7 ± 2.5	0.4	81.5 ± 3.0	82.1 ± 2.6	0.3	0.3
Globus Pallidus	73.0 ± 4.1	72.3 ± 4.3	0.3	73.7 ± 4.1	71.9 ± 4.0	0.03	72.0 ± 3.9	72.7 ± 4.5	0.5	0.4
SN pars reticularis	74.8 ± 10.5	74.4 ± 10.6	0.8	74.8 ± 10.2	74.4 ± 11.1	0.8	74.6 ± 11.1	74.3 ± 10.3	0.9	1.0
SN pars compacta	81.9 ± 4.1	81.3 ± 4.2	0.3	81.6 ± 4.6	81.4 ± 4.0	0.8	82.4 ± 3.3	81.3 ± 4.4	0.2	0.2
Red Nucleus	81.4 ± 12.3	79.3 ± 10.5	0.2	81.1 ± 11.1	77.1 ± 8.0	0.03	81.7 ± 13.8	81.6 ± 12.2	0.9	1.0

* Data are estimated mean $\pm$ SE T2-values (ms). Crude p-values are from linear regression analysis controlling for age. SN = Substantia Nigra.

Table 4

Age-controlled mean T2-values (ms $\pm$ SE) of migraineurs with longer vs. shorter duration of migraine history (n=138)*

Structure	Age < 50 years				Age $\geq$ 50 years			
	<23 yrs (n=35)		$\geq$ 23yrs (n=34)		< 23yrs (n=33)		$\geq$ 23yrs (n=36)	
			<i>p-value</i>				<i>p-value</i>	
			<i>crude</i>	<i>adjusted</i>			<i>crude</i>	<i>adjusted</i>
Putamen	74.9 $\pm$ 1.9	73.1 $\pm$ 2.4	0.001	0.01	74.5 $\pm$ 3.0	74.0 $\pm$ 3.2	0.5	0.5
Posterior Putamen	71.8 $\pm$ 2.7	70.2 $\pm$ 2.8	0.02	0.1	72.7 $\pm$ 4.4	71.2 $\pm$ 3.9	0.2	0.12
Head of Caudate	83.5 $\pm$ 2.0	82.0 $\pm$ 2.8	0.02	0.04	81.9 $\pm$ 2.9	82.2 $\pm$ 2.3	0.7	0.7
Globus Pallidus	72.4 $\pm$ 3.6	71.5 $\pm$ 4.5	0.4	0.7	72.9 $\pm$ 4.5	72.5 $\pm$ 4.6	0.7	0.7
SN pars reticularis	75.8 $\pm$ 12.0	73.1 $\pm$ 10.0	0.3	0.3	75.4 $\pm$ 10.7	73.4 $\pm$ 10.0	0.4	0.4
SN pars compacta	81.9 $\pm$ 4.0	80.9 $\pm$ 4.0	0.3	0.5	81.3 $\pm$ 4.9	81.2 $\pm$ 4.0	0.9	0.9
Red Nucleus	79.7 $\pm$ 9.6	74.5 $\pm$ 5.0	0.006	0.001	82.2 $\pm$ 11.1	81.0 $\pm$ 13.3	0.7	0.6

* Data are estimated mean $\pm$ SE T2-values (ms). Crude p-values are from 2-tailed unpaired t-tests. Adjusted p-values are from linear regression analysis controlling for age. SN = Substantia Nigra..