1. Title:

Magnetic resonance spectroscopy in migraine: what have we learned so far ?

2. Authors:

Harmen Reyngoudt, PhD[1,2]
[1]Department of Radiology and Nuclear Medicine, Ghent University, Ghent, Belgium
[2]Ghent Institute for Functional and Metabolic Imaging, Ghent University, Ghent, Belgium

Eric Achten, MD, PhD[1,2]
[1]Department of Radiology and Nuclear Medicine, Ghent University, Ghent, Belgium
[2]Ghent Institute for Functional and Metabolic Imaging, Ghent University, Ghent, Belgium

Koen Paemeleire, MD, PhD[3,4]
[3]Department of Basic Medical Sciences, Ghent University, Ghent, Belgium
[4]Department of Neurology, Ghent University Hospital, Ghent, Belgium

3. Correspondence to:

Harmen Reyngoudt
Ghent University Hospital
MR-Department, -1K12B
De Pintelaan 185
B-9000 Ghent, Belgium

tel. +32 9 332 64 25
fax +32 9 332 49 69
E-mail: hreyngou@gmail.com

4. Running title:

MR spectroscopy in migraine

ABSTRACT:

Objective: To summarize and evaluate proton (^{1}H) and phosphorus (^{31}P) magnetic resonance spectroscopy (MRS) findings in migraine.

Methods: A thorough review of ^{1}H and/or ^{31}P-MRS studies in any form of migraine published until September 2011.

Results: Some findings were consistent in all studies, such as a lack of ictal/interictal brain pH change and a disturbed energy metabolism, the latter which is reflected in a drop in phosphocreatine (PCr) content, both in the resting brain and in muscle following exercise. In a recent interictal study adenosine triphosphate (ATP) was found to be significantly decreased in the occipital lobe of migraine with aura patients, reinforcing the concept of a mitochondrial component to the migraine threshold, at least in a subgroup of patients. In several studies a correlation between the extent of the energy disturbance and the clinical phenotype severity was apparent. Less consistent but still congruent with a disturbed energy metabolism is an observed lactate (Lac) increase in the occipital cortex of migraine with (prolonged) aura patients. No increases in brain glutamate (Glu) levels were found so far.

Conclusion: The combined abnormalities found in MRS studies imply a mitochondrial component in migraine neurobiology. This could be due to a primary mitochondrial dysfunction or be secondary to e.g. brain disexcitability. The extent of variation in the data can be attributed to both the variable clinical inclusion criteria used and the variation in applied methodology. Therefore it is necessary to continue to optimize MRS methodology to gain further insights, especially concerning Lac and Glu.

KEYWORDS: ^{31}P-MRS, ^{1}H-MRS, brain, migraine, migraine without aura, migraine with aura, migraine with prolonged aura, migrainous stroke, basilar-type migraine, familial hemiplegic migraine, sporadic hemiplegic migraine, brain metabolism, muscle metabolism, ATP, PCr, Lac, NAA, Glu

INTRODUCTION

Migraine is a complex neurovascular disorder, involving complex genetics, multiple neurotransmitter systems, and multiple cortical and subcortical regions of the brain (1). Experimental migraine models have been useful but incomplete in reflecting the full scope of the human condition (2). At present, no integrative model exists to explain this multifaceted syndrome. Over the past 25 years, several magnetic resonance spectroscopy (MRS) studies have been performed in migraine patients. Whereas in the first ten years these studies comprised mostly phosphorus MRS (^{31}P-MRS), a majority of proton MRS (^{1}H-MRS) studies were performed since 2003. Although a large number of data have been generated, the contribution to our understanding of migraine pathophysiology by these heterogeneous studies is not straightforward. Of these thirty-one MRS-studies, two handled multinuclear MRS (i.e. both ^{1}H-MRS and ^{31}P-MRS) and another three used visual stimulation to excite the brain. Thirty studies covered one or more brain areas, predominantly the occipital cortex, although measurements were also performed in other cortical regions and non-cortical regions such as the basal ganglia. Only nine studies covered ^{31}P-MRS of muscle tissue. Most studies emphasized on migraine with aura (MwA) and its rare subtype of hemiplegic migraine. To a lesser extent migraine without aura (MwoA) was covered, especially in combination with ^{1}H-MRS. The majority of studies were obtained interictally. ^{31}P-MRS results were mostly depicted as molar concentrations (i.e. absolute quantification), whereas ^{1}H-MRS studies used almost exclusively ratios (i.e. relative quantification) to represent the results.

A review of MRS data in migraine has not been performed since 2006 (3). We therefore embarked on an updated review of both ^{31}P-MRS and ^{1}H-MRS data . Besides discussing the objective MRS results, a critical review was made of patient and control inclusion criteria (i.e. migraine subtypes, regions of interest, age, gender, attack frequency, prophylaxis, ictal versus interictal) as well as methodological aspects (i.e. field strength, coil, MR-parameters, quantification method, assumptions concerning adenosine triphosphate or ATP) and their respective potential shortcomings. Eligibility criteria for papers to be included in this review were original articles in English on both ^{31}P-MRS and ^{1}H-MRS in muscle and brain of migraine patients, published up to September 2011 and available through MEDLINE using the search terms ‘Migraine’ and ‘Magnetic Resonance Spectroscopy’. This search

yielded 76 papers of which 30 original articles fulfilled eligibility criteria and were included. Two papers were excluded for language raison (one paper in Polish and one in Japanese), two more papers were excluded due to co-morbidities (lupus, during transient global amnesia). Among the 76 papers were 6 review papers on MRS in migraine, as well as one additional MRS study in a rat model of migraine. A broader MEDLINE search with 'Migraine' and 'Spectroscopy' did not yield additional papers, but using the search terms "MRS" and 'migraine' 1 additional eligible paper was identified, bringing the total of original articles to 31. Citation lists of all original articles and review papers were searched for additional references, but none were found. A Web of Science search using search terms 'Migraine' and 'Magnetic Resonance Spectroscopy' did not yield additional original articles.

^{31}P-MRS FINDINGS

^{31}P-MRS is a noninvasive tool to analyze energy metabolism of brain, muscle and other tissues (4). Under normal aerobic circumstances, ATP is produced through glycolysis and oxidative phosphorylation. ATP can also be generated through transfer of inorganic phosphate (P_i) from phosphocreatine (PCr) to adenosine diphosphate (ADP) via the creatine kinase reaction. Anaerobically, the brain switches to the significantly less efficient anaerobic glycolysis to produce ATP. The concentrations of ATP ([ATP]), PCr ([PCr]) and P_i ([P_i]), as well as phospholipids can be derived directly from the ^{31}P-spectrum (Fig. 1A). Additional parameters can be calculated, such as the PCr/P_i ratio (which reflects the energy status of the cell), the phosphorylation potential defined as ATP/(ADPxP_i), the ADP concentration ([ADP]) and the percentage of v/v_{max} (the ratio of the actual velocity of oxidative metabolism to the maximum oxydative capacity). The concentration of magnesium ([Mg^{2+}]), which is an important cofactor in the creatine kinase reaction, as well as the intracellular pH (pH_i), can be calculated from chemical shifts in the spectra. Table 1 gives a detailed overview of ^{31}P-MRS studies in the brain of migraine patients.

BRAIN

Ictal changes

Only the group of Welch et al. measured brain energy phosphate metabolism and pH_i during migraine attacks in both MwoA and MwA patients (5, 6). They observed a significant reduction of the PCr/Pi ratio in both anterior and posterior brain areas of MwA (but not in MwoA) patients, indicating a low availability of free cellular energy, but there was no alteration in brain pH_i during attacks (as well as interictally). Additionally, low magnesium levels were observed during migraine attacks without changes in pH_i (7). These early ^{31}P-MRS studies in migraine failed to support the prevailing concept at that time that migraine pain was caused by brain ischemic acidosis, but rather suggested a disordered energy metabolism during a migraine attack.

Interictal changes

Most ^{31}P-MRS studies were performed interictally, but the duration of the headache-free interval is rarely specified. Interictal phosphorus brain abnormalities in occipital lobes were common to different subtypes of migraine, such as MwoA (5, 6, 8-12), MwA (5, 6, 9-11, 13, 14), familial hemiplegic migraine (FHM) (11, 15), basilar-type migraine (10, 14), migraine with prolonged aura (9, 10, 16, 17) as well as in migrainous stroke (9, 10, 16, 18). A few studies report similar results in other brain regions, including the basal ganglia (19) and frontotemporal (11, 20) and parietal brain areas (20).

First of all, a significant decrease in [PCr] (8, 9, 12-15, 17) and an increase in [P_i] (13-15, 17) resulting in a reduced PCr/P_i ratio (5, 16, 17, 19, 20) were the most generalized observations, regardless of the brain region. One study, Boska et al., reported only a depletion of PCr in anterior brain regions of MwA and the occipital cortex of FHM patients (11). The calculated value of the phosphorylation potential was significantly decreased in migraine patients, as reported in a number of studies (8, 9, 12-15). The brain [ATP] was always assumed constant and equal to normal controls, being 3 mM or was not mentioned at all, except for Reyngoudt et al. who quantified [ATP] in the occipital lobe of MwoA patients and found a significant decrease compared to controls, at least in a subgroup of patients (12). A significantly increased [ADP], indicating that brain tissue is working at a higher metabolic rate and hence has a lower energy reserve, was also frequently observed (8, 9, 13-15, 18). The percentage of v/v_{max} was found to be significantly increased in migraine patients (8, 9, 14, 15).

pH_i was measured in all abovementioned studies and no significant pH_i-changes were observed in all but three studies (13, 14, 18). The pH_i-changes found in these studies should be regarded carefully, though, because of the presence of large standard deviations (13), borderline significance (14) or

migrainous stroke (18). The same remark holds true for Mg^{2+}, an essential cofactor for mitochondrial membrane stability and in oxidative phosphorylation, which was found significantly decreased in a minority of studies (7, 10, 11, 14).

The combined abovementioned abnormalities suggest a mitochondrial component in the neurobiology of migraine, at least in a subgroup of patients. Indeed, a similar cerebral ^{31}P-MRS pattern has been observed in some mitochondrial cytopathies (21, 22). However, a consistent pattern of mitochondrial abnormalities (e.g. from muscle biopsies or mitochondrial DNA analysis) in migraine has not emerged, and these steady state measurements reflect the balance of ATP demand and mitochondrial function/ATP production (4). Anyhow, the data suggests the brain of migraine patients is compromised to deal with metabolic stress.

It seems that the cerebral ^{31}P-MRS findings correlate with the severity of the clinical phenotype in several studies (10, 12, 16, 19) . Reyngoudt et al. observed a reduction in [ATP] in patients with the highest attack frequency and suggested that the existence of (a) subgroup(s) may explain apparent contradictions between previous studies as most were performed on small and/or heterogeneous patient groups (see further) (12).

MUSCLE

Several ^{31}P-MRS studies also reported a defective energy metabolism in skeletal muscle tissue (always in the gastrocnemius muscle except for two studies which did not specify which calf muscle), including MwoA, MwA, migraine with prolonged aura, FHM and migrainous stroke patients (8, 13-18, 23, 24). The measurement paradigm generally existed of a rest period, followed by an exercise period, and again followed by a rest period. The (isokinetic) exercise involved pressing a pedal (plantar flexion) connected with a pneumatic ergometer and the cycle rest-exercise-rest was repeated several times. Most ^{31}P-MRS muscle studies showed no significant differences in phosphorus metabolites between patients and controls at rest (8, 13-15, 17, 18). However, following exercise patient's muscles had a lower PCr content and a higher P_i content than controls showing a slower recovery of both PCr and P_i, which is reflected in an elevated recovery time constant. According to Uncini et al., the degree of muscle oxidative metabolism defect in FHM patients seemed to be correlated with the clinical phenotype (15). PCr and P_i concentrations at rest and during work and recovery were calculated

assuming an intramuscular ATP concentration of 8 mM (25). These ^{31}P-MRS data suggest a multisystem oxidative metabolism defect, including both muscle and brain tissue.

^{1}H-MRS FINDINGS

^{1}H-MRS allows the detection of a number of important neurotransmitters, such as glutamate (Glu) and γ-aminobutyric acid (GABA) and related compounds such as glutamine (Gln), as well as the endproduct of anaerobic glycolysis, lactate (Lac) (26). Glu is the major excitatory neurotransmitter in the central nervous system and constitutes with Gln the so-called Glu-Gln cycle. N-acetylaspartylglutamate is the most abundant peptide neurotransmitter in the mammalian central nervous system, consisting of NAA and glutamic acid coupled via a peptide bond, and is thought to play a role as a major osmolyte in the vertebrate brain (46). Other metabolites including N-acetylaspartate (NAA), total creatine (tCr, 'total' referring to the contribution of both creatine and phosphocreatine), choline (Cho) and myo-inositol (mIns) are also found abundantly in brain using ^{1}H-MRS (Fig. 1B). NAA is considered a neuronal marker of particular axonal integrity whereas mIns is assumed to be a glial marker. NAA is synthesized and located predominantly in neuronal mitochondria, and assumed to be involved in mitochondrial/cytosolic carbon transport. mIns is at the center of a complex metabolite pathway involving inositol phosphates and inositol phospholipids which have well established functions in signal transduction and in Ca^{2+} homeostasis in the central nervous system. tCr depicts the combined spectral resonance of both creatine and PCr, that together play a role in the creatine kinase reaction. Finally, Cho reflects membrane turnover. Table 2 gives a detailed overview of ^{1}H-MRS studies in the brain of migraine patients.

Lac

An early ^{1}H-MRS study showed elevated interictal levels of cerebral Lac in the occipital cortex of a heterogeneous group of migraine patients (i.e. three MwA patients, one basilar-type migraine patient, one migrainous stroke patient and one migraine with prolonged aura/ migrainous stroke patient) (27). A similar observation was made in the cerebellum of four patients with FHM2, a rare subtype of MwA (23). In a functional ^{1}H-MRS study, visual stimulation resulted in a Lac increase in the occipital visual cortex of MwA patients with both visual symptoms and paraesthesia, paresis and/or dysphasia, but not

in MwA patients with only visual symptoms, in which Lac was already higher than normal in the resting state (28). The authors of the abovementioned studies suggested that the accumulation of Lac was a marker for a disturbance in oxidative glycolysis, related to energy metabolism impairment, which is typical for mitochondrial diseases (29). Recent studies, however, performed by Reyngoudt et al. at higher field strength (3T) in MwoA patients did not demonstrate significant increases in Lac, both at rest (30) and following visual stimulation (31). It was postulated that the lack of stimulation-induced Lac increase in MwA, as reported in Sandor et al. (28), and also in patients with mitochondriopathies might be partially attributed to a saturation of Lac transporter systems (32). In the subgroup of MwA, with high resting-state Lac levels, it was hypothesized that the aura was limited to the visual cortex, whereas in the subgroup of migraine patients with both visual symptoms and paraesthesia, paresis and/or dysphasia, this was not the case. Experiments demonstrated that decreased pH_i, which would accompany increased extracellular Lac, is an inhibiting factor for cortical spreading depression (CSD), which is believed to be at the basis of the aura symptoms (33). However a decrease of pHi has never been observed, and an accurate detection, let alone accurate quantification of Lac, is cumbersome and will be discussed further on in this paper.

NAA

A functional ^{1}H-MRS study reported significantly reduced NAA levels in the occipital cortex of MwA patients but not in MwoA patients compared to controls, before and following visual stimulation (34). The authors hypothesized the presence of less efficient mitochondrial functioning in MwA patients compared to MwoA patients and controls (34), and that the reversible loss of NAA during visual stimulation is due to a redistribution of NAA from the intra to the extracellular space which would result in a variation in chemical environment that may alter its magnetic resonance visibility (35). The hypothesis of a mitochondrial deficiency was also put forward by a study in which decreased NAA was found in the (left) thalamus of MwoA patients (36). Finally, a reduced NAA concentration was also found at rest in the cerebellum of FHM1 patients (37), in the left temporoparietal cortex (site of migrainous stroke) of a sporadic hemiplegic migraine (SHM) patient (38) and in the occipital cortex of another SHM patient (39), which has been rather attributed to neuronal impairment or loss. The latter is obvious in migrainous stroke but may also fit with the frequently observed cerebellar atrophy in FHM (40).

Glu/Gln

Glu levels were found to be markedly reduced in the cerebellum of FHM1 patients in a ^{1}H-MRS study performed at 1.5 T (37). However, methodological aspects including a potential spectral overlap of Gln- and GABA-resonances at 1.5 T make it difficult to assess these changes. According to Dichgans et al., the reduction of Glu may in part reflect neuronal impairment as was already indicated by a concomitant NAA reduction (37). Another possibility to explain the decreased Glu levels is an impaired glutamatergic neurotransmission. The latter hypothesis, however, is in contrast with a prediction of enhanced glutamatergic neurotransmission in FHM1 patients, based on a gain of function of pre-synaptic Ca^{2+} channels by CACNA1A mutations (41). The concomitant decrease in Glu and NAA (described above) are therefore likely to be attributed to atrophy.

Recently susceptibility loci for migraine, both MwA and MwoA, have been described in genome-wide association studies, and some refer to Glu homeostasis (42-44)**.** Although no significant metabolite differences were observed in a study using two-dimensional ^{1}H-MRS, linear discriminant analysis revealed a separation between migraine patients and controls based on Gln and N-acetylaspartylglutamate in the anterior cingulate cortex (ACC) and insula, suggesting glutamatergic abnormalities in the brain of migraine patients compared to controls (45). At present, however, there is no direct proof of altered brain glutamate levels in MwA and MwoA.

mIns

Limited data are reported concerning mIns. A decreased mIns content was found in the left temporoparietal cortex of one SHM patient during prolonged hemiparetic migraine aura (38). Another study in FHM1 patients showed an increased interictal cerebellar mIns concentration (37). As mIns is considered an *in vivo* glial marker, an increase in this metabolite would signify a regional glial cell proliferation (46), which could be in line with an autopsy report that gave evidence of a proliferation of astrocytes and gliosis in the cerebellum of a CACNA1A mutation carrier (47).

Cho

A single study reports a decreased Cho level in the cerebellum of MwA patients, suggesting a variation in membrane turnover and/or composition (48).

METHODOLOGY

Not only have MRS studies been performed in a wide variety of migraine subtypes and brain regions (see Tables 1 and 2), they also have been performed using different strategies concerning the recruitment of patients and control subjects (Table 3) and there are substantial methodological MRS differences (Table 4). We will discuss these important methodological aspects, before we attempt to draw general conclusions from the body of MRS literature in migraine so far.

REGIONS OF INTEREST

Occipital

In most studies the occipital cortex was chosen as the region of interest (5-11, 13-18, 23, 27, 28, 30, 31, 34, 37, 39, 49). According to several studies this was due to the fact that aura, with most often visual symptoms, could be attributed to this area in MwA patients (13, 27, 37, 49). However, aura symptoms are experienced by 20-30% of migraine patients. Consequently, other reasons for performing measurements in the occipital lobes are more of a practical nature, including its ideal location for high signal-to-noise ratio when using surface coils, as stated in several early MRS studies (5-7), and the fact that it is easily stimulated (28, 31, 34).

Frontal regions, ACC, insula, cerebellum, hypothalami, thalami,…

Frontal regions were investigated in some earlier studies based on the hypothesis that CSD progressed anteriorly (5-7). Frontal regions, including the primary motor cortex, were also examined in a few more recent studies, including a study covering FHM and migrainous stroke (11, 20). Moreover, in the study of Prescot et al. (45) the ACC as well as the insula were under investigation for reasons that both brain areas are implicated in the perception of pain (50, 51). MRS was also performed in the cerebellum of MwA (48) and FHM patients (37). The cerebellum was investigated in FHM patients since a substantial proportion develop cerebellar degeneration (37). Subclinical cerebellar abnormalities have been described and are more pronounced in MwA than MwoA (52), but the cerebellar volume in MwA is unaltered (53). The thalamus and hypothalamus, subcortical structures involved in the pathophysiology of migraine attacks, were investigated in MwoA patients (1, 36) and

(probable) chronic migraine (54) respectively. Several other studies focused on larger brain regions, both grey and white matter, for different reasons (see Table 1 and 2 for details), including the hemisphere on the side of the headache (5-7), the site of MRI abnormalities (38), or to look at white matter specifically (19).

Muscle

^{31}P-MRS of muscle was always conducted in the right calf muscles (almost exclusively in gastrocnemius muscle except for two studies where the muscle type was not specified) (8, 13-18, 23, 24). Reasons for this are practical since surface coils can easily be positioned around the calf, gastrocnemius is a large enough muscle to perform localized spectroscopy and calf muscles can easily be fatigued by employing an ergometer.

PATIENTS AND CONTROLS

Age and gender

All but five studies (9, 18, 19, 37, 39) reported the age-ranges of both migraine patients and controls. Overall, most subjects were between 20 and 45 years of age, but there are limited data in children and adolescents (14, 17, 39, 49).

Migraine patients and controls were matched for both gender and age in merely four studies (10, 14, 23, 31, 45), for age specifically in approximately 50 % of the studies (8, 11, 12, 17, 19, 27, 28, 30, 34, 48, 54) and for gender specifically in one study (36).

Migraine subtypes and attack frequency

Patient groups did not always exist of a homogeneous migraine subtype making interpretations of metabolic alterations not always as straightforward. Montagna et al. performed ^{31}P-MRS in a large cohort of migraine patients including MwoA patients, MwA patients and a group of 'complicated' migraine patients, the latter comprising both migrainous stroke and migraine with prolonged aura (9). In Lodi et al., ^{31}P-MRS was performed in a migraine group consisting of MwoA, MwA, basilar-type migraine, migraine with prolonged aura and migrainous stroke as well as in a group of cluster headache patients (10). Both studies, Montagna et al. (9) and Lodi et al. (10), did not mention

migraine attack frequencies. Watanabe et al. reported elevated Lac levels in the occipital cortex of five (two with MwA, one with basilar-type migraine, one with migrainous stroke and one with migraine with prolonged aura/ migrainous stroke) out of six investigated migraine patients (27). The migraine patient group in the study of Sandor et al. was also rather heterogeneous, including patients suffering from MwA, migraine with prolonged aura and MwAplus, the latter consisting of patients who had visual aura associated with at least one of the following: paraesthesia, dysphasia or paresis (some perhaps should have been labeled hemiplegic migraine ?) (28). In Prescot et al., they mention patients who suffered from acute episodic migraine without verifying whether it concerned MwoA or MwA (45). In several studies the criteria for 'migraine attack frequency' were not very stringent (13, 19, 20, 34, 37) or were lacking altogether (5-7, 9-11, 15, 24, 48, 49).

Several studies, though, did cover homogeneous patient groups who experienced a well-defined number of attacks, as was the case for MwoA (8, 12, 13, 30, 31, 34), MwA (19, 34) and FHM1 (37).

Ictal versus interictal

In all but a few studies migraine patients were scanned interictally, mostly because of obvious practical reasons but also to explore the interictal character of the disorder rather than the attack itself. Attack-free intervals ranged from 48 hours (12, 27, 30, 31) to a few days (11, 28, 34, 49), from a week (5-7, 10, 14) to more than a week (17, 19, 20, 23, 39). In all other studies no information concerning the attack-free period is specified (8, 9, 13, 15, 16, 18, 24, 36, 37, 45, 48, 54).

The first ^{31}P-MRS studies in migraine, however, were studies in which both ictal and interictal measurements were performed, yet in two different fractions of the migraine patients (5-7). In these aforementioned studies not one patient underwent both an ictal and interictal measurement or had an aura at the time of the study. These ictal spectra were measured between 3 and 48 hours following the headache onset (5-7). Finally, Jacob et al. obtained an ictal ^{1}H-MR spectrum during a long-lasting migraine attack of a SHM patient (38).

Prophylaxis: yes or no?

Most studies described the lack of use of prophylactic medication in a certain (in most cases not defined) time period before as well as during the course of the MRS-experiments (8, 9, 11-14, 16, 18, 24, 30, 31, 34, 36, 39, 49). In quite a few other studies, however, no information about the medication

history of migraine patients is given (15, 17, 19, 20, 27, 28, 37, 48, 54). Furthermore, in the earliest MRS-studies (5-7) as well as in Prescot et al. (45), a small minority of patients studied ictally were taking medication for migraine prophylaxis.

MRS METHODOLOGY

Methodological differences may also explain the discrepancies found in MRS studies in migraine. Methodological aspects include the choice of magnetic field strength, coil, localization technique, repetition time, echo time, flip angle, (pre)processing steps, signal estimation, quantification procedure,... These aspects can be found in Table 4.

Magnetic field strength and coils

Only a minority of the studies was conducted at higher field strengths (i.e. ≥ 3 T) (11, 12, 30, 31, 37, 45), providing greater spectral resolution, which allows more accurate metabolite quantification and determination of spectral frequencies, which is also the basis of pH and [Mg^{2+}] calculations. A potential spectral overlap of Gln- and GABA-resonances was mentioned in a ^{1}H-MRS study performed at 1.5 T by Dichgans et al. (37). The variability in the MRS data reflects a combination of true variation between study subjects and experimental variation, which mainly arises from the signal-to-noise ratio of the acquired spectra. It is possible that the signal-to-noise ratio, in particular in ^{31}P-MRS with the inherent lower sensitivity of the ^{31}P-nucleus, did not allow to detect significant differences between controls and some patient subgroups. This holds true for ^{1}H-MRS as well and the detection of more subtle metabolic changes, such as a rise in Lac in healthy volunteers (28, 34), could be achieved with high-field MR systems and/or the use of surface coils.

Indeed, the detection of Lac has long been a burning issue in MRS. It should be emphasized that Lac is a low-concentration metabolite in healthy brain tissue (i.e. in the orders of 0.2-1 mM). And although early functional ^{1}H-MRS studies, performed at lower field strength (i.e. 1.5 T), found Lac increases of 60-150% (55-57), Mangia et al. (58-60) reported in several advanced ^{1}H-MRS studies, performed at 7 T, an increase in [Lac] of 0.1-0.2 mM following visual stimulation and corresponding to an increase of only 20%. A Lac increase of up to 0.2 mM is unlikely to reflect a switch to anaerobic glycolysis, according to Mangia et al. (59, 60). Considering these results and the fact there is very little

information about absolute Lac values in the migrainous brain, except the 3 T functional ^{1}H-MRS study of Reyngoudt et al. in which Lac concentrations of 0.5-0.6 mM were found in controls and MwoA (31), one should be very careful concerning Lac quantification as well as the interpretation of the results.

^{31}P-MRS studies were generally performed using surface coils because of a high achievable signal-to-noise ratio (5-10, 13-20, 23, 24). Volume coils were employed in most ^{1}H-MRS studies (19, 23, 28, 30, 31, 34, 36, 45, 48) compared to in only two studies performing ^{31}P-MRS (11, 12). In a few studies information concerning coil specification was lacking (37-39, 49).

MRS measurement parameters

Table 4 describes which MRS localization technique was used in each study. The majority of ^{31}P-MRS studies made use of the depth-resolved surface-coil spectroscopy (8-10, 13-18, 23, 24) whereas ^{1}H-MRS was mostly performed using point-resolved spectroscopy (19, 20, 27, 28, 30, 31, 34, 36-39, 54). The advantages of single voxel applications are a homogeneous magnetic field across the volume, good spectral resolution and the measurement of a well-defined volume, although the latter can simultaneously be seen as a main drawback since other important areas may be missed. This can be solved by multivoxel applications although this increases measurement times significantly. Moreover, magnetic field inhomogeneities across an entire object, inherent to multivoxel measurements, make quantitative spectroscopy quite challenging.

The choice of specific MRS measurement parameters such as echo time and repetition time may also have a substantial effect on the acquired spectra. Spectra acquired at short echo time often reveal a complex spectral pattern and severe overlap of broad resonances from macromolecules and lipids, further complicating the accurate assessment of changes in metabolites. Moreover, long echo times are often preferred for the detection of low-concentration resonances such as Lac (19, 20, 27, 28, 31, 34, 36, 39, 45).

Signal processing

Signal processing procedures differed extensively between studies over the years. The Welch group initially used basic triangulation to determine signal areas of ^{31}P-peaks (5-7), whereas the Bologna group quantified phosphorus data using a user-interactive curve fitting program (8-10, 13-18, 24). Later ^{31}P-MRS studies used scanner software (11, 19, 20, 34, 45), LCModel (23, 37, 45) or the

AMARES (30, 48) and/or QUEST (31) algorithm (jMRUI). Information regarding signal estimation was not reported in the remaining studies (27, 28, 38, 39, 49, 54).

Quantification

Relative quantification was performed by calculating the ratios between peaks and was conducted in approximately half of the studies (see Table 4). In ^{31}P-MRS studies this involved in most cases the PCr/P_i ratio while in ^{1}H-MRS studies metabolites were generally normalized to tCr, which is assumed to be relatively constant. However, changes in the tCr content have been observed in several conditions and one should be cautious when using tCr as an internal reference (see review in (46)).

In the remaining studies attempts were made to perform 'absolute' quantification. In almost all ^{31}P-MRS studies in migraine (8-10, 13-15, 17, 18) metabolites were assessed by assuming a constant cytosolic ATP concentration of 3 mM (61), since no absolute data on ATP concentration in human brain of migraine patients exist. In Reyngoudt el al. (12), however, [ATP] was quantified, based on the fact that β–ATP is proportional to the total cellular ATP content (62, 63). Similarly, muscle ATP concent was assumed to be a constant 8 mM (15, 23). Dichgans et al. (37) and the more recent studies of Reyngoudt et al. (12, 30, 31) are the only ^{1}H-MRS studies reporting 'absolute' metabolite values, and included also the incorporation of some additional correction factors (see Table 4).

Functional paradigm

Three different functional ^{1}H-MRS studies used three different functional paradigms, as can be seen in Table 4 (28, 31, 34, 64). Stimulation duration varied from around 13-15 min. (28, 31) to 25 min. (34). In comparison, the studies of Mangia et al. mentioned earlier (58-60), performing functional ^{1}H-MRS at 7 T using a radial red/black checkerboard (8 Hz), observed a significant increase in Lac and Glu of approximately 20 % and 3%, respectively, following a stimulation of only 2 min. These Lac increases were reduced over time, implying an adaptation of Lac changes following visual stimulation.

CONCLUSION

Clearly, MRS studies in migraine have yielded disparate information on brain metabolism. The data regarding ^{31}P-MRS in muscle is far more scarce, yet the results seem to be rather reproducible. The

variation found throughout these studies can be attributed to a number of reasons including heterogeneous migraine populations, migraine severity (attack frequency), medication history, time of measurement (ictal versus interictal), regions of interest, magnetic field strength, signal processing and/or quantification strategies. Sometimes important study details (attack frequency, age, gender, quantification steps,...) were not recorded and therefore it is not possible to determine the extent to which they may have affected the results.

Nevertheless, some findings were consistent overall regardless of abovementioned variability, such as a lack of acidosis (no pH changes) and a disturbed energy metabolism, at least in a subgroup of patients, implying a mitochondrial component in migraine pathophysiology. Moreover, in several studies a correlation between the extent of the energy disturbance and the intensity of the clinical phenotype was apparent. ATP was found to be reduced in the occipital lobe of MwoA patients, however, ATP remains to be measured in MwA patients. Additional evidence supporting a disturbed energy metabolism in migraine, comes from pharmacological studies showing the effects of metabolic enhancers, such as riboflavin and coenzyme Q10 (both with a well defined role in mitochondrial membrane generation of ATP), in migraine prophylaxis (65-68). It is worthy of note that the therapeutic response to riboflavin was associated with specific mitochondrial DNA haplogroups (69). Less consistent but still possibly congruent with the disturbed energy metabolism is an observed Lac increase in the occipital cortex of several migraine subtypes (i.e. MwA, migraine with prolonged aura), however, potential Lac changes remain to be confirmed in a larger homogeneous population of both MwA and MwoA patients, preferably at higher field strength (i.e. 7 T). An increased Glu content was not found at all, despite its potential correlation with well-defined missense mutations found in FHM patients causing a hyperexcitable cortex, and therefore Glu should also be investigated at higher field strengths. Table 5 displays the major interictal (occipital) MRS findings in common forms of migraine (MwoA and MwA) and hemiplegic migraine.

We would like to stress that the depletion of brain high-energy phosphates reflects an imbalance between ATP production and ATP use in migraine patients, but we do not know whether is due to a primary mitochondrial dysfunction or secondary to alterations in brain excitability. The 31P-MRS muscle studies support the former hypothesis. It is interesting to speculate that abnormalities are more

apparent in the brain because it is in fact under constant activation. Only when duly exercised, in fact, muscle energetics were similarly abnormal in migraine patients. In general, migraine can be considered a 'threshold disorder' or a 'biobehavioural disorder' (70), implying that an intrinsic metabolic defect may render the brain more susceptible to various factors triggering an attack. Triggering factors would act by increasing the metabolic demand or hampering ATP production, and when a certain threshold is reached in an organ which is rather near its maximum capabilities, such as the brain, they would then induce a metabolic crisis responsible for the headache attack.

Potential future research may comprise further robust, quantitative and multinuclear brain MRS studies in migraine, including patients with a single phenotype who experience a well-defined number of attacks and are not using prophylaxis, compared to a gender- and age-matched control group. Another possible next step could be the absolute quantification of muscle phosphorus metabolites, especially ATP, in migraine patients, since [ATP] was assumed to be constant (i.e. 8 mM) in the muscle of migraine patients and controls in previous ^{31}P-MRS studies. Finally, ^{13}carbon-MRS could provide further insight in possible Lac and Glu dynamics in migraine pathophysiology.

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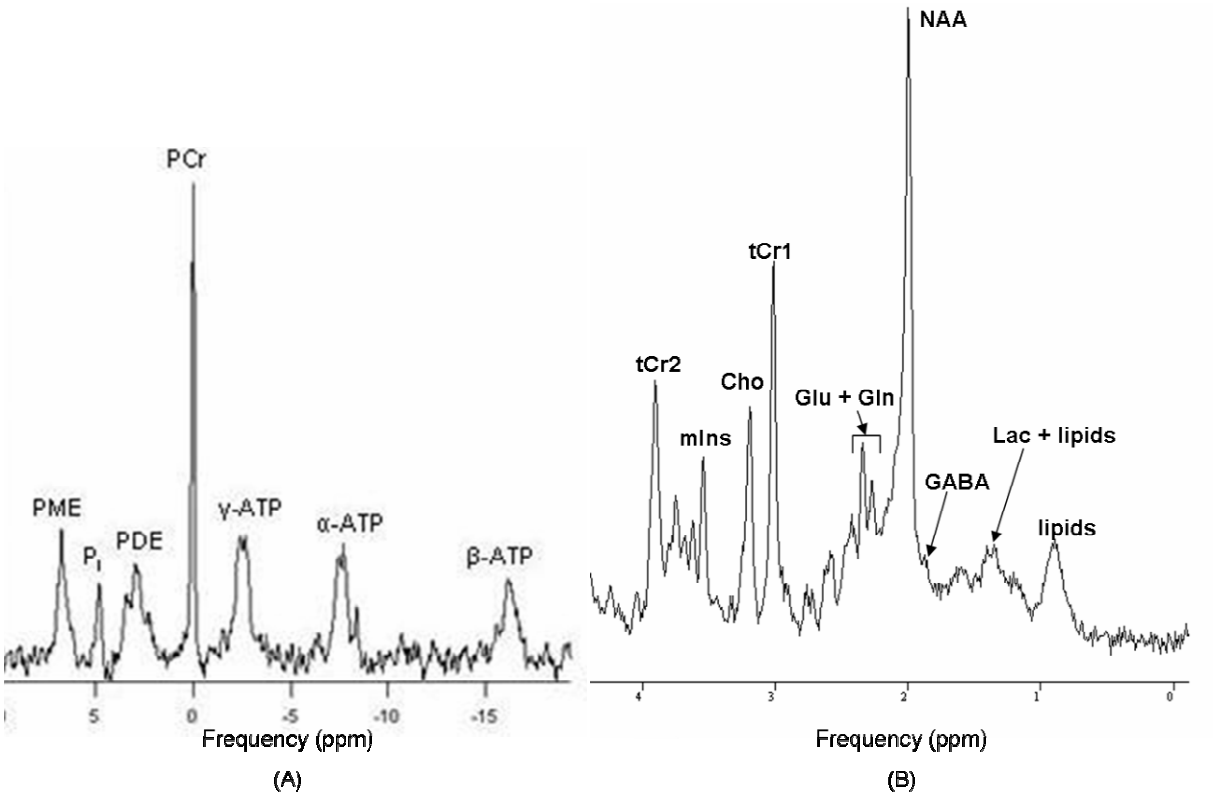

Figure 1: Brain ^{31}P (A) and ^{1}H -spectrum (B). Several ^{1}H-metabolites are difficult to quantify, e.g. Glu + Gln (difficult to differentiate, small peaks, close to NAA), GABA (very small peak right to NAA), Lac (low concentration, resonates at same frequency as lipids such as subcutaneous fat). From the ^{31}P-spectrum, pH_i can be calculated from the frequency difference between P_i and PCr, and $[Mg^{2+}]$ can be calculated from the frequency difference between PCr and β-ATP. Some metabolites show several resonance peaks in the spectrum originating from different proton or phosphorus nuclei, such as tCr (i.e. tCr1 and tCr2) and ATP (i.e. α-ATP, β-ATP and γ-ATP). (PME = phosphomonoesters, PDE = phosphodiesters).

Study (reference number)	Migraine type (sample size)	Brain region(s)	Results[a]
Welch et al. 1988 (6)[b,c]	MwoA (12), MwA (8)	fronto(temporal), (parieto)occipital	no changes
Welch et al. 1989 (5)[b]	MwoA (12), MwA (8)	fronto(temporal), (parieto)occipital	$PCr/P_i \downarrow$, $P_i/TP \downarrow$
Ramadan et al. 1989 (7)[b]	MwoA (11), MwA (8)	fronto(temporal), (parieto)occipital	$[Mg^{2+}] \downarrow$
Barbiroli et al. 1990 (16)	MwpA (4), MS (4)	occipital	$PCr/P_i \downarrow$, $PCr/ATP \downarrow$
Bresolin et al. 1991 (18)	MS (1)	occipital	$PCr/ATP \downarrow$, $P_i/ATP \uparrow$, $[ADP] \uparrow$, $pH \uparrow$
Sacquegna et al. 1992 (17)	MwpA (1)	occipital	$PCr/P_i \downarrow$, $[PCr] \downarrow$, $PCr/ATP \downarrow$, $[P_i] \uparrow$
Barbiroli et al. 1992 (13)	MwA (12)[d]	occipital	$[PCr] \downarrow$, $[P_i] \uparrow$, $pH \downarrow$, $[ADP] \uparrow$, $PP \downarrow$, $v/v_{max} \uparrow$
Montagna et al. 1994 (8)	MwoA (22)	occipital	$[PCr] \downarrow$, $[ADP] \uparrow$, $PP \downarrow$, $v/v_{max} \uparrow$
Montagna et al. 1995 (9)	MwoA (22), MwA (18), other (15)	occipital	$[PCr] \downarrow$, $[ADP] \uparrow$, $PP \downarrow$, $v/v_{max} \uparrow$
Uncini et al. 1995 (15)	FHM (2), MwoA (3)	occipital	$[PCr] \downarrow$, $[P_i] \uparrow$, $[ADP] \uparrow$, $PP \downarrow$, $v/v_{max} \uparrow$
Lodi et al. 1997 (14)	MwA (7), MwpA (3), BM (5)	occipital	$[PCr] \downarrow$, $[P_i] \uparrow$, $pH \downarrow$, $[ADP] \uparrow$, $PP \downarrow$, $v/v_{max} \uparrow$, $[Mg^{2+}] \downarrow$
Lodi et al. 2001 (10)	MwoA (21), MwA[e] (37), MwpA (13), MS (7)	occipital	$[Mg^{2+}] \downarrow$
Boska et al. 2002 (11)	MwoA (19), MwA (19), FHM (8)	anterior, occipital	$[PCr] \downarrow$, $[Mg^{2+}] \downarrow$, $[PDE] \uparrow$
Schulz et al. 2007 (19)	MwA (21)	temporoparietal	$PCr/P_i \downarrow$, $P_i/ATP \uparrow$
Schulz et al. 2009 (20)	MwpA (9), MS (5)	frontotemporal, parietal	$PCr/P_i \downarrow$
Reyngoudt et al. 2011 (12)	MwoA (19)	occipital	$[PCr] \downarrow$, $[ATP] \downarrow$, $PP \downarrow$

MwoA, migraine without aura; MwA, migraine with aura; MwpA, migraine with prolonged aura; MS; migrainous stroke; BM, basilar-type migraine; FHM, familial hemiplegic migraine; other: MS and MwpA; PCr, phosphocreatine; P_i, inorganic phosphate; ATP, adenosine triphosphate; ADP, adenosine diphosphate; PP, phosphorylation potential; v/v_{max}, rate of ATP synthesis; TP, total phosphorus signal; PDE, phosphodiesters; Mg^{2+}, magnesium; ↑, increased; ↓, decreased.

[a] $P < 0.05$

[b] This study was performed both ictally and interictally.

[c] Only pH was measured in this study.

[d] 4/12 patients suffered from MwoA as well.

[e] MwA patients included typical aura and BM.

Table 1: Overview of brain ^{31}P-MRS studies in migraine patients: migraine types, brain regions and results.

Study (reference number)	Migraine type (sample size)	Brain region(s)	Results[a]
Watanabe et al. 1996 (27)	MwA, other (6)	occipital	Lac/NAA ↑
Macri et al. 2003 [(48)	MwA (8)	cerebellum	mIns/tCr ↓, Cho/tCr ↓
Sandor et al. 2005 (28)[b]	MwA (5), MwAplus (5)	occipital	Lac/NAA ↑
Dichgans et al. 2005 (37)	FHM (15)	cerebellum, occipital, parietal	Glu ↓, NAA ↓, mIns ↑
Sarchielli et al. 2005 (34)[c]	MwoA (22), MwA (22)	occipital	NAA/tCr ↓
Jacob et al. 2006 (38)	SHM (1)	temporoparietal	NAA/tCr ↓, mIns/tCr ↓
Wang et al. 2006 (54)	CM (16)	hypothalamus	no changes
Schulz et al. 2007 (19)	MwA (21)	basal ganglia	no changes
Gu et al. 2008 (36)	MwoA (20)	thalamus	NAA/tCr ↓
Prescot et al. 2009 (45)	Mw(o)A (10)	ACC, insula	no changes
Grimaldi et. 2010 (23)	FHM2 (4)	(parieto-)occipital, ventricles	Lac ↑
Chawalparit et al. 2010 (49)	MwpA[d] (1)	occipital	no changes
Toldo et al. 2010 (39)	SHM (1)	occipital	NAA/tCr ↓
Reyngoudt et al. 2010 (30)	MwoA (22)	occipital	no changes
Reyngoudt et al. 2011a (31)[e]	MwoA (20)	occipital	no changes

MwoA, migraine without aura; MwA, migraine with aura; MwpA, migraine with prolonged aura; MS; migrainous stroke; FHM(2), familial hemiplegic migraine (type 2); SHM, sporadic hemiplegic migraine; CM, chronic migraine; other: MS and MwpA; MwAplus: MwA with symptoms of paresthesia/paresis/dysphasia; Lac, lactate; NAA, N-acetylaspartate; mIns, myo-inositol; Cho, choline; tCr, total creatine; Glu, glutamate; ↑, increased; ↓, decreased.

[a] $P < 0.05$

[b] Visual stimulation was performed by projection of a blue/yellow flickering checkerboard (8 Hz).

[c] Visual stimulation was performed by projection of a flashing red light (14 Hz).

[d] This patient suffered actually from migrainous headache with a series of long-lasting visual aura.

[e] Visual stimulation was performed by projection of a black/white flickering checkerboard (8 Hz).

Table 2: Overview of brain ^{1}H-MRS studies in migraine patients: migraine types, brain regions and results.

Study (reference number)	Patients						Controls		
	Migraine type (sample size)	F/M	Age (mean ± standard deviation)	Attack frequency	Pφ	(inter)ictal	n	F/M	Age
Welch et al. 1988 (6), Welch et al. 1989 (5)	MwoA (12), MwA (8)	12/0, 6/2	37.2 ± 12.0, 37.1 ± 13.2	-	Y	ictal (11/20), inter (9/20)	27	18/9	45.1 ± 17.6
Ramadan et al. 1989 (7)	MwoA (11), MwA (8)	11/0, 6/2	34.3 ± 9.7, 37.1 ± 13.2	-	Y	ictal (10/19), inter (9/19)	25	15/10	43.4 ± 18.2
Barbiroli et al. 1990 (16)	MwpA (4), MS (4)	-, -	31.3 ± 15.6, 34.5 ± 4.0	2 ± 1 (/month)	N	interictal	15	15/0	18-44
Bresolin et al. 1991(18)	MS (1)	1/0	40	1.5 (/month)	N	interictal	14	-	-
Sacquegna et al. 1992 (17)	MwpA (1)	0/1	17	2-4 (/year)	-	interictal	8	0/8	17-21
Barbiroli et al. 1992 (13)	MwA (12)	10/2	30.6 ± 9.0	1-24 (/year)[a]	N	interictal	14	14/0	18-50
Montagna et al. 1994 (8)	MwoA (22)	19/3	33.7 ± 9.8	3.6 ± 1.8 (/month)	N	interictal	18	-	33.7 ± 9.8
Montagna et al. 1995 (9)	Mw/oA (55)	-	-	-	N	interictal	50	-	-
Uncini et al. 1995 (15)	FHM,MwoA (5)	3/2	26.0 ± 12.7	-	-	interictal	50	28/22	16-50
Watanabe et al. 1996 (27)	MwA,other (6)	5/1	31.0 ± 9.5	1-12 (/year)[a]	-	interictal	6	4/2	30.0 ± 4.7
Lodi et al. 1997 (14)	MwA (7), MwpA (3), BM (5)	5/2, 0/3, 1/4	13.1 ± 2.2, 13.6 ± 1.3, 12.4 ± 2.4	3.8 ± 1.7 (/month)	N	interictal	12[b], 15[b]	5/7, 6/9	12.9 ± 1.9, 13.1 ± 2.1
Lodi et al. 1997 (24)	MwoA (23), MwA (22), MwpA (11), MS (7)	-	16 to 61	-	N	interictal	49	-	16 to 63
Lodi et al. 2001(10)	MwoA (21), MwA (37), MwpA (13), MS (7)	19/2, 21/16, 8/5, 7/0	32.0 ± 2.0, 23.0 ± 2.0, 25.0 ± 3.0, 38.0 ± 3.0	-	N	interictal	36	-	36.0 ± 3.0
Boska et al. 2002 (11)	MwoA (19), MwA (19), FHM (8)	17/2, 15/4, 5/3	35.7 ± 9.5, 40.9 ± 8.4, 39.3 ± 4.1	-	N	interictal	40	27/13	37.5 ± 11.3
Macri et al. 2003 (48)	MwA (8)	6/2	28.0 ± 11.0	-	-	interictal	7	3/4	28.0 ± 9.0
Sandor et al. 2005 (28)[b]	MwA (5), MwAplus (5)	4/1, 5/0	37.5 ± 11.4, 26.5 ± 14.0	3.5 ± 2.1 (/month)	-	interictal	11	5/6	29.9 ± 6.6
Dichgans et al. 2005 (37)	FHM (15)	9/6	37.1 ± 12.4	1-72 (/year)[a]	-	interictal	17	9/8	22-54
Sarchielli et al. 2005 (34)[c]	MwoA (22), 22 (22)	15/7, 14/8	37.2 ± 5.1, 34.5 ± 4.9	1-40 (/year)[a]	N	interictal	10	5/5	33.3 ± 6.4
Jacob et al. 2006 (38)	SHM (1)	1/0	33	-	N	ictal	0	n/a	n/a
Wang et al. 2006 (54)	CM (16)	12/4	42.3 ± 10.9	-	-	-	21	8/13	36.1 ± 9.0
Schulz et al. 2007 (19)	MwA (21)	12/9	42.0 ± 11.0	1->12 (/year)[a]	-	interictal	16	8/8	39.0 ± 15.0
Gu et al. 2008 (36)	MwoA (20)	11/9	37.3 ± 12.3	2.3 ± 1.6 (/month)	N	interictal	14	8/6	20-62
Prescot et al. 2009 (45)	Mw/oA (10)	7/3	43.3 ± 3.3	3.3 ± 1.2 (/month)	Y	interictal	8	-	-
Schulz et al. 2009 (20)	MwpA (9), MS (5)	5/4, 2/3	41.2 ± 9.5, 46.6 ± 16.2	1->12 (/year)[a]	-	interictal	14	-	-
Grimaldi et. 2010 (23)	FHM2 (4)	4/0	35.0 ± 15.0	2.5 ± 0.7 (/year)	-	interictal	10	10/0	34.0 ± 10.0
Chawalparit et al. 2010 (49)	MwpA (1)[d]	1/0	17	-	N	interictal	0	n/a	n/a
Toldo et al. 2010 (39)	SHM (1)	1/0	8	1 (/year)	N	interictal	0	n/a	n/a

Reyngoudt et al. 2010 (30)	MwoA (22)	21/1	33.3 ± 12.2	3.4 ± 1.1 (/month)	N	interictal	26	15/10	27.6 ± 12.0
Reyngoudt et al. 2011 (12)	MwoA (19)	18/1	32.3 ± 12.1	3.6 ± 1.1 (/month)	N	interictal	26	15/11	27.6 ± 10.9
Reyngoudt et al. 2011a (31)	MwoA (20)	17/3	31.9 ± 9.1	3.6 ± 1.1 (/month)	N	interictal	20	17/3	31.9 ± 10.3

MwoA, migraine without aura; MwA, migraine with aura; MwpA, migraine with prolonged aura; MS; migrainous stroke; BM, basilar-type migraine; FHM(2), familial hemiplegic migraine (type 2); SHM, sporadic hemiplegic migraine; CM, chronic migraine; MwAplus: MwA with symptoms of paresthesia/paresis/dysphasia; other: MS and MwpA; n, number of volunteers; F/M, female/male ratio; Pφ, prophylaxis; Y, yes, N, no; '-', information is lacking; n/a, not applicable. [a] Heterogeneous data: both MwA and MwoA attacks; [b] 12 subjects for brain, 15 subjects for muscle.

Table 3: Demographic and patient information of MRS studies in migraine.

Study	B_0 (vendor)	coil	MR-sequence	TR/TE (s/ms)	Quantification	ATP (mM)[a]	corrections
Welch et al. 1988 (6), Welch et al. 1989 (5)	1.89 (Bruker)	surface	-	-	relative	-	-
Ramadan et al. 1989 (7)	1.89 (Bruker)	surface	DRESS	-	absolute	-	-
Barbiroli et al. 1990 (16)	1.5 (GE)	surface	DRESS	5/-	relative	-	-
Bresolin et al. 1991 (18)	1.5 (GE)	surface	DRESS	5/-	relative	3.0	-
Sacquegna et al. 1992 (17)	1.5 (GE)	surface	DRESS	5/-	relative, absolute	3.0	-
Barbiroli et al. 1992 (13)	1.5 (GE)	surface	DRESS	5/-	absolute	3.0	-
Montagna et al. 1994 (8)	1.5 (GE)	surface	DRESS	5/-	absolute	3.0	-
Montagna et al. 1995 (9)	1.5 (GE)	surface	DRESS	5/-	absolute	3.0	-
Uncini et al. 1995 (15)	1.5 (GE)	surface	DRESS	5/-	absolute	3.0/8.0	-
Watanabe et al. 1996 (27)	1.5 (GE)	surface	PRESS	1.5/270	relative	n/a	-
Lodi et al. 1997 (14)	1.5 (Siemens)	surface	DRESS	5/-	absolute	3.0	-
Lodi et al. 1997 (24)	1.5 (GE)	surface	DRESS	5/-	relative, absolute	8.2	-
Lodi et al. 2001 (10)	1.5 (GE)	surface	DRESS	5/-	absolute	3.0	-
Boska et al. 2002 (11)	3 (Magnex)	volume	MRSI	1/-	absolute	3.0	-
Macri et al. 2003 (48)	1.5 (GE)	volume	PRESS	1.5/30	relative	n/a	-
Sandor et al. 2005 (28)[b]	1.5 (Philips)	volume	MRSI	1.5/288	relative	n/a	-
Dichgans et al. 2005 (37)	1.5 (GE)	-	PRESS	2/35	absolute (LCModel)	n/a	segment.
Sarchielli et al. 2005 (34)[c]	1.5 (GE)	volume	PRESS	2/144	relative	n/a	-
Jacob et al. 2006 (38)	1.5 (GE)	-	PRESS	1.5/35	relative	n/a	-
Wang et al. 2006 (54)	1.5 (GE)	surface	PRESS	1.5/144	relative	n/a	-
Schulz et al. 2007 (19)	2 (Bruker)	volume, surface	PRESS, MRSI	1.5/135 (^{1}H), 2.5/- (^{31}P)	relative (jMRUI)	-	-
Gu et al. 2008 (36)	3 (GE)	volume	MRSI	1/144	relative	n/a	segment.
Prescot et al. 2009 (45)	4 (Varian)	volume	2D-PRESS	2/30-260	relative (LCModel)	n/a	-
Schulz et al. 2009 (20)	2 (Bruker)	surface	PRESS	1.5/135	relative (jMRUI)	-	-
Grimaldi et. 2010 (23)	1.5 (GE)	volume, surface	PRESS, DRESS	4/35 (^{1}H), 5/- (^{31}P)	relative (LCModel, jMRUI)	8.0	segment.
Chawalparit et al. 2010 (49)	1.5 (Philips)	-	svs	-	-	n/a	-
Toldo et al. 2010 (39)	1.5 (Philips)	-	PRESS	2/299	relative	n/a	-
Reyngoudt et al. 2010 (30)	3 (Siemens)	volume	PRESS	2/30	relative, absolute (jMRUI)	-	CSF,…
Reyngoudt et al. 2011 (12)	3 (Siemens)	volume	CSI-FID	4/2.3	absolute (Siemens)	meas.	
Reyngoudt et al. 2011a (31)	3 (Siemens)	volume	PRESS	2/288	relative, absolute (jMRUI)	n/a	CSF,…

B_0, magnetic field strength (in T); TR, repetition time (in s); TE, echo time (in ms); DRESS, depth-resolved surface coil spectroscopy; (2D-)PRESS, (2D-) point-resolved spectroscopy; svs, undefined single voxel spectroscopy measurement; MRSI, magnetic resonance spectroscopy imaging; CSI-FID, chemical shift imaging free induction decay; ATP, adenosine triphosphate; '-', information is lacking; n/a, not applicable; segment., brain segmentation; meas., measured.
[a] Assumed ATP concentrations: in brain: 3.0 mM; in muscle: 8.0 mM.

Table 4: Methodological parameters of MRS studies in migraine.

Migraine subtype	ATP	PCr	PP	Lac
MwoA	↓ (12)	↓ (5, 8, 9, 11, 12)	↓ (8, 12)	= (30, 31)
MwA	?	↓ (5, 9, 11, 13, 14)	↓ (9, 13, 14)	↑ (27, 28)
S/FHM	?	↓ (14, 15)	↓ (15)	?

MwoA, migraine without aura; MwA, migraine with aura; S/FHM, sporadic and familial hemiplegic migraine; ATP, adenosine triphosphate; PCr, phosphocreatine; PP, phosphorylation potential; Lac, lactate

Table 5: Synthesis of major interictal brain MRS findings in migraine.

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