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Research Submission

MMP-9 Immunoreactivity in Acute Migraine

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Objective.—To examine the role of matrix metalloproteinase 9 (MMP-9) in migraine during headache and asymptomatic periods.

Methods.—Thirty-four patients with migraine with and without aura (according to International Headache Society criteria) were studied. Clinical characteristic of headache were recorded. Blood samples for measurement of MMP-9 were drawn during headache attacks and during asymptomatic periods in all patients and in 10 healthy controls.

Results.—We found higher plasma MMP-9 levels in migraine patients than in control group (129.3 [78.0–258.9] vs. 49.6 [39.1-64.3] ng/mL; $P < .001$). Migrainous patients showed higher MMP-9 plasma levels during headache attacks than in asymptomatic periods, both in migraine without aura (338.4 [275.1–396.2] vs. 118.2 [75.3–137.5] ng/mL; $P < .0001$), and migraine with aura (389.3 [273.4–487.1] vs. 139.3 [107.3–191.4] ng/mL; $P < .0001$).

Conclusions.—Our study showed an increased production of MMP-9 during migraine attacks. These data suggest a possible role of inflammation or blood–brain barrier disruption during the migraine attack.

Key words: migraine, metalloproteinase, inflammation, blood–brain barrier

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The pathophysiology of migraine is complex, and not completely understood. A variety of factors are implicated in the development of migraine aura or headache such as genetic factors, environmental predisposition, as well as neurophysiologic, biochemical, and vascular mechanisms. The 2 main symptoms are aura, probably caused by cortical spreading depression and headache, as a result of pial and extracerebral vasodillation and neuroinflammation.[1] Although a number of pathophysiological mechanisms are involved, one important mechanism that remains to be explored is the role of the blood–brain barrier (BBB).

Few references can be found in the literature regarding the function of the BBB in the pathophysiology of the migraine attack. For example, there has been a suggestion of the possible role of the alteration of BBB integrity in migraine attack.[2] Additionally, a mild but significant left-hemispheric BBB opening limited to the cortex and preceding cortical edema was revealed by early gadolinium-enhanced MRI in a patient with a severe attack of familiar hemiplegic migraine type II.[3] Experimental studies showed that sumatriptan does not pass the BBB easily under normal circumstances, and it is only effective on trigeminal neurons to alter their function after disruption of the BBB.[4]

The basic component of the BBB, endothelial basal lamina, can be degraded by a group of proteolityc zinc-dependent enzymes known as matrix

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metalloproteinases (MMPs).[5] MMPs are produced by all cell types in the brain, including neurons, glia, and invading leukocytes and macrophages. MMPs have many roles in CNS ranging from remodelling tissues during development to a critical role in multiple phases of neuroinflammation. Loss of control and increased expression and activity of MMP have been implicated in various neurological and nonneurological disorders. MMPs have been implicated in processes such as activation of cytokines and cytokines receptors,[6] invasion of neural tissue by blood derived immune cells,[7] and interaction between cells and their matrix allowing movement and shape changes,[8] and formation of edema.[9]

One of these proteolityic enzymes, MMP-9, has been shown to induce a high breakdown capacity, especially in the arteriolar basement membrane, leading to cerebral edema and secondary hemorrhage.[10,11] In animal models of cerebral ischemia, expression of MMP-9 was significantly increased and related to BBB disruption, edema formation, and hemorrhagic transformation.[12,13] Recently, it was shown in an animal model of cortical spreading depression (CSD) that intense neuronal and glial depolarization initiates a cascade that disrupts the BBB via an MMP-9-dependent mechanism.[14]

The aim of the present study was to investigate the role of MMP-9 in migraine during headache and asymptomatic periods.

PATIENTS AND METHODS

We investigated prospectively 34 migraine patients with and without aura according to IHS criteria.[15] Twenty patients suffered from migraine with aura (72% women, 40.9 ± 10 years), and 14 suffered from migraine without aura (76% women; 39.1 ± 12 years). A control group consisting of 10 healthy subjects (68% women; 49.1 ± 11 years) was also included.

The main variables related to the migraine attacks were recorded: frequency (1 = less than 3 attacks/month; 2 = 3–6 attacks/month; 3 = higher than 6 attacks/month), intensity (1 = mild; 2 = moderate; 3 = severe), and duration (1 = less than 12 hours; 2 = between 12 and 24 hours; 3 = higher than 24 hours). None of the subjects had taken preventive treatment in the previous 3 months. Patients were instructed not to take symptomatic treatment before blood extraction. Blood plasma samples were collected during headache attacks in the first 3–6 hours from onset and during pain free periods. The study was approved by the Ethics Committee at the University of Santiago de Compostela.

Laboratory Test.—Blood samples were collected in glass test tubes containing potassium edetate. Suspensions of plasma were centrifuged at 3000 g for 5 minutes and immediately frozen and stored at –80°C. Serum total MMP-9 (92 kDa pro- and 82 kDa active forms) was measured with commercially available quantitative sandwich enzyme-linked immunoabsorbent assay (ELISA) kits obtained by Biotrak (Amersham Pharmacia, UK). Amershan RPN2614 is a kit specific for free pro-MMP-9 + pro-MMP-9 complexed to TIMP-1; no cross-reactivity with pro-MMP1, pro-MMP-2, pro-MMP-3, TIM-1, and TIM-2; range: 1–32 ng/mL; sensitivity: 0.6 ng/mL. All ELISAs were performed according to manufacturer's instructions by researchers blinded to clinical data.

Statistical Analysis.—The 2 migraine groups were compared by use of the χ^2 test. Given that MMP-9 levels were not normally distributed, they were expressed as median (quartiles). Both migraine groups were compared using the Mann–Whitney test. The relation between intensity and duration of attacks and MMP-9 plasma levels were evaluated by the Kruskall Wallis test.

RESULTS

Plasma MMP-9 levels were higher in the migraine groups than in controls (129.3 [78.0–258.9] vs. 49.6 [39.1–64.3] ng/mL; $P < .001$). Moreover, plasma MMP-9 levels were significantly higher during migraine attacks than during pain-free periods (366.3 [269.3–483.0] vs. 129.3 [78.0–258.9] ng/mL; $P < .0001$).

The increase of MMP-9 during migraine attacks was similar in both groups: migraine patients without aura (338.4 [275.1–396.2] vs. 118.2 [75.3–137.5] ng/mL; $P < .0001$), and migraine patients with aura (389.3 [273.4–487.1] vs. 139.3 [107.3–191.4] ng/mL; $P < .0001$) (Fig. 1).

Neither intensity nor duration of attacks presented any correlation with MMP-9 plasma levels

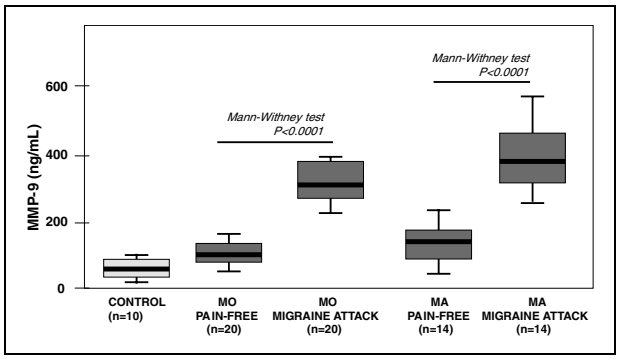

Fig 1.—Plasma MMP-9 levels in migraine patients with and without aura during migraine attacks and asymptomatic periods. Both types of migraine showed higher plasma levels of MMP-9 during migraine attacks.

(Fig. 2). No significant differences ($P = .492$) in plasma MMP-9 levels were found with respect to intensity of migraine attacks: mild (234.7 ng/mL [94.5–403.9]), moderate (126.2 ng/mL [67.1–287.8]), and severe headache (287 ng/mL [91.3–380.8]). Similarly, no differences ($P = .378$) were observed in plasma MMP-9 levels with respect to migraine duration: less than 6 hours (210.5 ng/mL [87.5–396.5], between 6 and 12 hours (189.4 ng/mL [73.4–269.5], and migraine attacks of more than 12 hours (297 ng/mL [113.5–402.7]).

COMMENTS

This study shows that plasma levels of MMP-9 are markedly increased during migraine attacks, both in migraine with and without aura. The relevance of this finding is that MMP-9 is a potential molecular marker of neuroinflammation, and BBB disruption. This finding also may have implications in the identification of new potential targets for migraine therapy.

In an experimental animal model, Gursoy-Ozdemir et al[14] showed that cortical spreading depression activates and upregulates MMPs and promotes sustained changes in vascular permeability. These authors suggest that intense neuronal and glial depolarization initiates a cascade that disrupts BBB via an MMP-9 dependent mechanism. The link between migraine and MMP-9, however, has not yet been demonstrated.

Certain evidence supports the association between CSD and migraine. In 1981, Olesen et al[16] provided the first report of slowly spreading cortical hypoperfusion in human patients during migraine attacks incompatible with spasm of a major artery but fully compatible with the occurrence of CSD. Later studies of regional cerebral blood flow support these observations.[17] A detailed magnetic resonance study of some patients gave a particularly graphic demonstration of a slowly spreading phenomenon in the occipital lobe during a migraine with aura attack.[18]

The presence of high plasma MMP-9 during migraine attacks suggests that neuroinflammatory mechanisms or blood–brain barrier disruption might be involved in the pathophysiology of migraine attacks. Nevertheless, a relation with CSD cannot yet be demonstrated in humans.

High plasma levels of MMP-9 were observed during pain-free periods in migraine patients with aura.

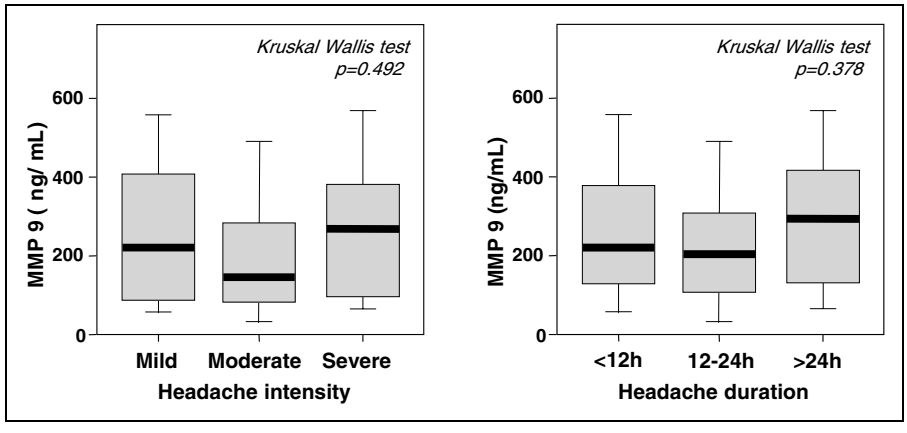

Fig. 2.—Relation between intensity or duration of migraine attacks and plasma MMP-9 levels.

This may be because biochemical abnormalities (interictal hypersensivity to nitroglycerine, calcitonin gene-related peptide (CGRP), or magnesium) in migraine sufferers outside the attacks may be genetic or environmental, but the former is most likely when changes are present weeks after the last attack and when they persist for years.[1] In this study, however, the time from last migraine attack to the current attack was not recorded, and, thus, no conclusions can be drawn.

This study has several limitations. MMPs are produced by all cell types in the brain, including neurons, glia, and invading leukocytes as well as macrophages. For the moment, we cannot establish the relative importance of these pools to plasma levels. Furthermore, it was not possible to analyze the importance of responses to stress or pain in the increase of MMP-9 plasma levels, because the control group consisted of healthy subjects who did not present any symptoms. In addition, this study did not register the time from migraine attack to pain-free period, or the time from the last migraine attack to the current attack. These variables would be important in order to interpret the results of MMP-9 plasma levels during pain-free periods.

CONCLUSION

The present study demonstrates that high plasma levels of MMP-9 are associated with migraine attacks with and without aura, suggesting a possible neuroinflammatory mechanism or BBB disruption. Further studies are needed to analyze the temporary profile of MMP-9 during the migraine attack in order to better understand the role of this molecular marker in the pathophysiology of migraine.

Conflict of Interest: None

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