
The Biology of Serotonin Receptors: Focus on Migraine Pathophysiology and Treatment

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ABSTRACT: Serotonin receptors are highly heterogeneous and they have been regrouped within seven different families (5-HT₁ - 5-HT₇). With the exception of the 5-HT₃ which is a ligand-gated ion channel, all others are G-protein coupled receptors with each family sharing structural, pharmacological and transductional characteristics. 5-HT receptors have been implicated in the regulation of several psychiatric and neurological disorders related to serotonergic neurotransmission, and specific receptor subtypes have recently been associated with either the pathogenesis or the treatment of migraine headache. In this respect, activation of vascular 5-HT_{2B} and/or 5-HT₇ receptors, possibly as a consequence of the sudden rise in 5-HT levels reported at the onset of a migraine attack, would hypothetically result in dilation of cerebral blood vessels and concomitant activation of sensory trigeminovascular afferents, hence initiating the manifestation of head pain. At this stage in the migraine process, activation of specific subtypes of 5-HT₁ receptors has proven clinically effective in relieving migraine pain. Neural 5-HT_{1D} and/or 5-HT_{1F} receptors localized pre-junctionally on trigeminovascular afferents appear to mediate the triptan-induced inhibition of the neurogenic inflammatory response, with possible additional sites of action for brain penetrant 5-HT₁ receptor agonists in inhibiting the transmission of pain centrally. In contrast, activation of vascular 5-HT_{1B} receptors would constrict meningeal vessels hence recovering their pre-migraine diameter. The recent availability of subtype selective 5-HT_{1D} and 5-HT_{1F} receptor agonists should allow a further test of the neural/vascular hypothesis and could possibly lead to antimigraine drugs with a safer cardiovascular profile.

RÉSUMÉ: Biologie des récepteurs sérotoninergiques. Les récepteurs sérotoninergiques sont très hétérogènes. On les a regroupés en 7 familles différentes (5-HT₁ - 5-HT₇). À l'exception de 5HT₃ qui appartient à la famille des récepteurs canaux, tous les autres appartiennent à la famille des récepteurs couplés à la protéine G et chaque famille a des caractéristiques structurales, pharmacologiques et transductionnelles communes. Les récepteurs 5-HT ont été impliqués dans la régulation de plusieurs maladies psychiatriques et neurologiques reliées à la neurotransmission sérotoninergique et des sous-types de récepteurs spécifiques ont été associés récemment à la pathogenèse ou au traitement de la migraine. À ce sujet, l'activation de récepteurs vasculaires 5-HT_{2b} et/ou 5-HT₇, possiblement comme conséquence de la hausse soudaine des niveaux de 5-HT rapportés au début d'un accès de migraine, provoquerait hypothétiquement une dilatation des vaisseaux sanguins cérébraux et une activation concomitante des afférents trigéminovasculaires sensitifs, amorçant ainsi la manifestation de la céphalée. À ce stade dans le processus migraineux, l'efficacité de l'activation de sous-types spécifiques de récepteurs 5-HT₁ pour soulager la douleur migraineuse a été démontrée en clinique. Les récepteurs neuronaux 5-HT_{1D} et/ou 5-HT_{1F} localisés en présynapse sur les afférents trigéminovasculaires semblent médier l'inhibition de la réponse inflammatoire neurogène induite par le triptan. Il est également possible qu'il existe des sites additionnels pour les agonistes des récepteurs 5-HT₁ qui pénètrent dans le cerveau et qui inhibent la transmission centrale de la douleur. Par ailleurs, l'activation des récepteurs vasculaires 5-HT_{1B} provoquerait une vasoconstriction des vaisseaux méningés qui reprendraient ainsi leur diamètre pré-migraine. L'arrivée d'agonistes sélectifs pour les sous-types de récepteurs 5-HT_{1D} et 5-HT_{1F} devrait permettre de tester l'hypothèse nerveuse/vasculaire et pourrait peut-être mener au développement de médicaments antimigraineux ayant un profil cardiovasculaire plus sûr.

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Serotonin (5-hydroxytryptamine, 5-HT) is a widespread neuromodulator which performs a variety of functions in the central and peripheral nervous systems, as well as in tissues such as the blood vessels. Serotonin is involved in several psychiatric and neurological disorders^{1,2,3} and, more specifically, in the pathogenesis of migraine headache.⁴ These

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conditions may result from inadequate neurotransmission, and hence in dysfunctional interactions between 5-HT and its pre- and/or post-synaptic receptors. 5-HT receptors are highly heterogeneous and the advent of molecular biology has allowed the cloning of several pharmacologically characterized receptors but also of new, so far unsuspected 5-HT receptors. Together with the availability of new pharmacological tools, these developments have provided new insights into the role of specific types of 5-HT receptors in the neurobiology of migraine headache. The present paper will briefly consider the different families of 5-HT receptors and will then focus more specifically on those that have attracted attention either in the aetiology or in the treatment of migraine headache. For a more detailed description of different aspects of 5-HT receptors, in terms of molecular, functional and pharmacological properties, extensive reviews are available.^{2,3,5}

HETEROGENEITY OF 5-HT RECEPTORS

Based on the recommended International Union of Pharmacology (IUPHAR), 5-HT receptors have been classified into seven different families (5-HT₁ to 5-HT₇).^{1,5} Except for the 5-HT₃ receptor which is a ligand-gated ion channel, all other receptors belong to the superfamily of G-protein-coupled receptors with seven transmembrane domains. Receptors within a single family share high homology in their amino acid sequence identity, they couple to the same second messenger systems and, although they display an overall typical pharmacological profile, individual receptors can be discriminated if subtype-selective pharmacological tools are available. Based on the IUPHAR nomenclature, receptors are written in upper case letter when they fulfill all molecular, pharmacological and functional criteria in biological tissues but those still lacking a biological role are listed in lower case letters.

5-HT₁ receptors represent the largest subfamily of 5-HT receptors and they typically exhibit a high affinity for 5-HT. They are subdivided into 5-HT_{1A}, 5-HT_{1B}, 5-HT_{1D}, 5-HT_{1E} and 5-HT_{1F} receptors, all of which are coupled to inhibition of adenylate cyclase activity.^{3,4} More specifically, 5-HT_{1A} receptors have been implicated in the regulation of many functions, including feeding behaviour, body temperature, and in disorders such as depression and anxiety.^{1,2} At present, there is no strong evidence to suggest that this receptor subtype would be selectively involved in migraine headache. However, it should be kept in mind that 5-HT₁ receptor agonists such as sumatriptan, the new triptan derivatives and other compounds such as the ergotamine and dihydroergotamine exhibit a relatively high affinity at 5-HT_{1A} receptors via which they may regulate nausea and vomiting responses.⁴

When first identified, 5-HT_{1B} and 5-HT_{1D} receptor subtypes were strictly impossible to discriminate on a pharmacological basis, except for a few compounds such as ketanserin and ritanserin which exert antagonistic activity at 5-HT_{1D} but not 5-HT_{1B} receptors.⁶ The cloning of two distinct genes for the 5-HT_{1B} and 5-HT_{1D} receptors then allowed for a better insight into their distribution and expression in different tissues.^{1,3,5} However, localization of their mRNA expression within brain tissues has shown several areas of overlap and it is still quite difficult to ascertain their respective roles. Together, these 5-HT₁

receptors have been implicated in depression, anxiety, movement disorders, and food intake.^{1,2} More recently, the role of 5-HT_{1B/1D} receptors in aborting migraine headache has been quite well elucidated (see below).

The cloning of 5-HT_{1E} and 5-HT_{1F} receptors has revealed their high sequence homology, and expression of mRNA for these two receptors has been found in the peripheral and/or central nervous system. Their coupling to adenylate cyclase activity appears to be less well defined than that of the other 5-HT₁ receptors and they may also couple to other signalling pathways.^{3,5} Although no definite function has been ascribed for these two receptor subtypes, recent evidence suggest that the 5-HT_{1F} receptor may be able to modulate the release of neurotransmitter from the trigeminovascular system, and as such, it has been implicated in the pathophysiology/treatment of migraine headache.⁷

5-HT₂ receptors have been involved in a plethora of functions spanning from smooth muscle contraction, transmitter release, sexual activity, appetite, sleep, locomotion, regulation of cerebrospinal fluid exchange, neurological and psychiatric disorders, and also migraine.² Three subtypes constitute this receptor subfamily, namely 5-HT_{2A} (the classical 5-HT₂ receptor), 5-HT_{2B} (highly expressed in stomach fundus and originally referred to as 5-HT_{2F}), and the 5-HT_{2C} (former 5-HT_{1C}) receptors. These receptors characteristically exhibit a lower affinity for 5-HT than 5-HT₁ receptors, and they positively couple to phospholipase C. 5-HT_{2A} receptors exist at high levels in many brain areas, while 5-HT_{2B} receptors are reportedly expressed at low levels in human brain and 5-HT_{2C} receptors are primarily enriched in the choroid plexus although they also overlap in their distribution with 5-HT_{2A} receptors.^{2,3,5} As will be described below, the originally suspected role for 5-HT_{2A} and 5-HT_{2C} receptors in migraine pathogenesis has recently been superseded by a most probable implication of 5-HT_{2B} receptors in the disease process.

The 5-HT₃ receptor represents the only ligand-gated ion channel 5-HT receptor, its activation leads to rapid increase in Na⁺/K⁺ conductance and a subsequent influx of extracellular Ca²⁺ and, in brain, it appears to be primarily composed of the heteromeric assembly of 5-HT_{3A} and 5-HT_{3B} subunits.⁸ 5-HT₃ receptors are located in the peripheral and central nervous system where high levels are found in the nucleus tractus solitarius and the area postrema, area of the brain emetic center. This localization most probably accounts for the original interest for 5-HT₃ receptor antagonists in the treatment of migraine headache. 5-HT₃ receptor antagonists indeed behave as antiemetic agents and are commonly used in cancer patients undergoing chemotherapy, however, they have not sustained a great interest in the treatment of migraine-associated symptoms over the recent years. A role for 5-HT₃ receptors in the modulation of cranial nociception has also been suggested.^{4,9} The cloning of a new 5-HT₃ receptor subunit⁸ further suggests that additional work is needed to clarify if 5-HT₃ receptors may still represent an interesting target in the management of migraine headache.

5-HT₄ 5-HT₆ and 5-HT₇ receptors have been cloned in human tissues and found in peripheral organs and/or in brain, however, very little is known about the roles of these 5-HT receptors (for more details, see ^{3,5}). 5-HT₄ and 5-HT₆ receptors seem to both

increase adenylate cyclase activity whereas a definite coupling system has not yet been identified for the 5-HT₅ receptors. Two isoforms of 5-HT₄ receptors have been reported,¹⁰ two different subtypes of 5-HT₅ receptors (5-HT_{5A} and 5-HT_{5B}) have been cloned,⁴ thus suggesting that much remains to be done in order to better understand the biology of these specific types of 5-HT receptors. 5-HT₆ receptors exhibit good affinity for many antipsychotic and antidepressants and they may be involved in such functions.³ No data exist at this point to suggest that either of these receptors could play a role in migraine biology.

5-HT₇ receptors, like the 5-HT₄ and 5-HT₆ receptors are positively coupled to adenylate cyclase activity, exhibit high affinity for a variety of antipsychotic drugs, and they may exist in multiple isoforms,¹¹ a finding which will deserve further investigation of their putative respective roles. 5-HT₇ receptors are discretely distributed in brain, mainly in the hippocampus, thalamic and limbic structures as well as in many other areas such as the spinal trigeminal nucleus.^{3,5,11} They are also present in many peripheral and cerebral blood vessels where they mediate an endothelium-independent dilation.^{11,12} Roles in affective behaviour, circadian rhythm and, possibly, migraine headache¹³ have been recently suggested.

5-HT RECEPTORS: RELEVANCE TO MIGRAINE PATHOGENESIS AND TREATMENT

5-HT₁ receptors in migraine headache

It is undeniable that the cloning of 5-HT_{1B} and 5-HT_{1D} receptors and the development of the 5-HT_{1B/1D/1F} receptor full agonist sumatriptan have revolutionized not only our ability to offer adequate treatment to more migraine sufferers but also our understanding of the neurobiology of migraine headache. Sumatriptan, originally developed to induce contraction of the putatively distended blood vessels during a migraine attack, and then found to potently inhibit the neurogenic inflammatory response resulting from activation of the trigeminovascular system,¹⁴ has led the impetus towards the identification of these respective vascular and neuronal 5-HT₁ sites of action. Moreover, recently developed brain penetrant triptan derivatives may offer an additional central locus of action through interaction with 5-HT₁ receptors able to interrupt the transmission of vascular pain centrally.^{5,15,16} The relative importance of the different 5-HT₁ receptor subtypes in the treatment of migraine headache may be appreciated when considering the meningeal neurogenic inflammation model proposed by Lance, Moskowitz and Fozard¹⁷ and characterized at length by Moskowitz and colleagues. Although this model has not been proven in man, reports of increased levels of calcitonin-gene related peptide (CGRP) in the jugular venous outflow during a migraine attack and their normalization after treatment with sumatriptan concomitantly with the relief of migraine symptoms,¹⁸ has provided arguments for activation of the trigeminovascular system in migraine sufferers.

Vascular 5-HT_{1B} receptors

Current pharmacological, molecular and morphological evidence indicate that the 5-HT₁ receptor which mediates the sumatriptan-induced contraction of the meningeal blood vessels

corresponds primarily, if not exclusively, to a 5-HT_{1B} receptor localized on the smooth muscle cells of brain vessels.^{19,20,21} However, in contrast to the original claim that sumatriptan could act on receptors that are unique to cerebral blood vessels, it has now been clearly demonstrated that vasocontractile 5-HT_{1B} receptors are also present in peripheral blood vessels such as the human coronary artery.²² Such findings indicate that all 5-HT₁ receptor agonists with affinity at the 5-HT_{1B} subtype share a similar potential risk of undesirable cardiovascular effects, a property which might be a contraindication in some patients.

Neuronal 5-HT_{1D} and 5-HT_{1F} sites of action

As mentioned above, activation of trigeminovascular afferents has been suggested as an important initiator of head pain. Once activated, these nerve fibers have the ability to release neuropeptides such as CGRP, substance P and neurokinin A¹⁴ and to promote the neurogenic inflammatory response thought to be important in the sensitization of the sensory afferents, as well as in the generation and transmission of head pain centrally.⁴ The ability of sumatriptan and other related compounds to act on pre-junctional 5-HT₁ receptors that inhibit neurogenic inflammation (characterized in animal models by protein extravasation and dilation of meningeal blood vessels) has been attributed to interaction with a 5-HT_{1D} and, possibly, a 5-HT_{1F} receptor.^{6,21} In support of these conclusions are the findings that mRNA and/or proteins for 5-HT_{1D}^{20,21} and 5-HT_{1F}²⁰ receptors are expressed in human trigeminal ganglia. Interestingly, although 5-HT_{1B} receptor mRNA and protein are present in human trigeminal ganglion neurons,²⁰ immunocytochemical evidence suggests that this receptor is not transported to terminals either peripherally on trigeminovascular afferents or centrally in the nucleus trigeminalis caudalis. Interestingly, the recent report that sumatriptan can directly repress CGRP gene transcription from sensory trigeminal neurons²³ may offer additional cellular targets to explain its antimigraine efficacy. More studies will be needed to clearly identify this trigeminal 5-HT₁ receptor as well as those localized in the brainstem in order to assess the respective role, if any, of the various 5-HT₁ receptors in neurogenic inflammation and central pain transmission. It should also be noted that 5-HT_{1B/1D} and 5-HT_{1F} receptors are present in multiple locations within the human brain²⁴ and their possible activation by brain penetrant 5-HT₁ receptor agonists deserves further investigation.

5-HT_{2B} and/or 5-HT₇ receptors in migraine pathogenesis

A landmark characteristic of commonly used compounds in migraine prophylaxis (e.g. pizotifen, methysergide, cyproheptadine, amitriptyline) is reportedly their antagonistic property at 5-HT_{2B}, but not 5-HT_{2A} and 5-HT_{2C} receptors.²⁵ In this respect, the similarity between the pharmacological profile of these antimigraine prophylactic agents and that of an endothelial 5-HT receptor mediating a nitric oxide (NO)-dependent dilation led to the hypothesis that activation of endothelial 5-HT_{2B} receptors would result not only in dilation of the cerebral blood vessels but also in the activation of the trigeminovascular afferents.²⁶ Clearly this aspect of migraine pathogenesis is reductionist as it again targets the pathway of vascular head pain and ignores all neuronal events that may also be altered in patients receiving prophylactically 5-HT₂ receptor antagonists. Whether specific

populations of 5-HT₂ receptors have been differentially regulated by such chronic treatments still remains unknown.

Despite this possible drawback, support for a role of vascular 5-HT_{2B} receptors in triggering migraine headache includes the selective expression of 5-HT_{2B} receptor message in human brain vessels,^{27,28} and endothelial cells,²⁸ and their reported ability, albeit in the pig, to induce an NO-dependent cerebral dilation.²⁷ Although more studies will be required to confirm such a role in human brain vessels, it is interesting that activation of these receptors in porcine cerebral vessels seems to promote the synthesis and release of NO,²⁷ a molecule known as an effective activator of trigeminovascular afferents.²⁹ In this respect, the recently reported efficacy of NO synthase inhibitors in the treatment of migraine headache³⁰ brings support for NO contributing to the pathogenesis of migraine headache. However, the constitutive nature of the NOS involved as well as the endothelial and/or neuronal origin of NO remain to be identified in order for this hypothesis to be proven true in the human situation.

Recently, a similar argument has been made with the 5-HT₇ receptors. Indeed, as for the 5-HT_{2B} receptors, a highly significant correlation has been found between the effective clinical doses of several prophylactic antimigraine compounds and their antagonistic affinity at 5-HT₇ receptors.^{12,13} These receptors can induce cerebral dilation through a direct action on the smooth muscle,^{11,12} they are expressed and functional in human brain vessels and vascular smooth muscle cells³¹ and they could thus play a role in regulating cerebrovascular tone. It is possible that during a migraine headache, the rise in serotonergic tone could result in a 5-HT₇-mediated increase in vessel diameter that could excite sensory trigeminovascular nerve terminals, which upon activation would initiate the neurogenic trigeminovascular inflammatory response. Additional experiments are needed to clarify this possibility as well as that of a direct role for pre-junctional 5-HT₇ receptors on trigeminovascular afferents.

CONCLUSION

Specific populations of 5-HT receptors seem to be involved in the pathogenesis and treatment of migraine headache. Most interesting is the fact that while 5-HT_{2B} and/or 5-HT₇ receptors are putatively implicated in the initiation of migraine pain, 5-HT_{1B/1D} and/or 5-HT_{1F} receptors appear as potent inhibitors of this symptom. These findings again underscore the complexity of serotonergic mechanisms and how different receptor populations can exert distinct functional responses. It is clear that changes in serotonergic status alone cannot explain the full array of the symptoms associated with migraine, but it is also impressive to realize how much the discovery of the different 5-HT₁ receptor subtypes has improved the treatment of migraine headache. It is hoped that the advent of subtype-selective 5-HT_{1D} and 5-HT_{1F} receptor agonists will similarly expand our understanding of the basic mechanisms that underlie these symptoms, and possibly lead to antimigraine drugs with better cardiovascular safety. Further studies will be needed to prove the clinical efficacy of such 5-HT₁ selective compounds, as well as the involvement of neuronal and/or vascular 5-HT₂ receptors in the initiation of the vascular head pain.

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