Pathophysiological role of Calcitonin Gene Related Peptide (CGRP) in Migraine

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Abstract

Our understanding regarding the pathophysiology of migaraine has changed a lot in past few years and now it is considered as a neuro-inflammatory disorder where calcitonin gene related peptide (CGRP) plays a major role. This peptide is found in the trigemino-vascular system and its role in migraine is confirmed by a number of different evidences including- higher serum concentration during spontaneous as well as experimental migraine, decrement in serum CGRP concentration after migraine abortive therapy and termination of acute attack of migraine with CGRP antagonists. This review examines all the evidences. Migraine and tension type headache are two types of headaches that are widely prevalent. Their prevalence varies from 5% to 70% across various studies depending upon the methodological differences.[1] World Health Organization recognizes migraine among the top ten disabling conditions.[2] Despite a huge burden imparted by this illness and wide prevalence, we are still not able to find out the exact patho-physiological mechanisms.

Introduction

In the past twenty years, our knowledge regarding the patho-physiological mechanisms of migraine has taken a totally new dimension and the vascular theory was replaced by concept of trigemino-vascular inflammation.[3-5] The calcitonin gene related peptide is found in the body and has caught maximum attention especially in cases of headache. This peptide has proven to be the key molecule for the headache as now-a-days its antagonists are being developed which have proven efficacy in management of acute migraine attacks. In this article we will review its chemistry and possible patho-physiological role in migraine.

Chemistry and distribution

CGRP is a 37 amino acid peptide and it was identified originally from structural analysis of the expression products of the calcitonin - gene. Its expression is dependent upon the tissue specific alternative mRNA processing. CGRP is generated mainly in the neural tissue while calcitonin is formed in the thyroid C-cells.[6] In humans, CGRP is found in both alpha and ß forms and their distribution in neural tissue also varies.[7] Alpha- CGRP is predominantly found in trigeminal ganglion, where sensory fibers are gathered, whereas Beta form is localized in occulomotor, trochlear and motor nucleus of trigeminal nerve.[8] Possibly, it is synthesized in the peripheral part of the nerves and then moves centripetally. This is evidenced by the fact that after experimental compression of the sensory neurons in dogs, CGRP accumulates distal to compression, and dorsal root ganglia cell positivity for CGRP decreases.[9]

CGRP-immuno-reactivity is present in perikarya in the trigeminal ganglia, trigeminal nucleus and in the cerebral vasculature of all species examined, including man.[10-15] It must be noted here that besides autonomic nerve supply, cerebral vasculature also has sensory supply and these nerves originate in trigeminal ganglion. It is further confirmed by immunochemical studies that have shown that cerebral arteries in animals have a moderate supply of CGRP reactive fibers that terminate in adventitia or adventitia-media border and similar fibers have been shown in human cerebral vasculature.[10-13]

In trigeminal ganglion, CGRP is frequently co-localized with SP in trigeminal cell bodies,

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although the number of CGRP-containing cells exceeds the number of SP-containing cells.16 Recently, CGRP has been found to be co-localized with 5HT1D receptors in human spinal trigeminal tract, although the co-localization was not seen in all fibers. Similar to the ganglia, few fibers were found to co-localize CGRP and substance P at this location also.[15]

Besides immunochemical studies, clinical evidences also favor the presence of CGRP in trigeminal sensory neurons. In a patient with facial pain, cutaneous stimulation was found to increase the blood levels of CGRP, to the extent that it was measurable in the cubital venous blood.[17] This shows that CGRP was released following cutaneous stimulation. These sensory nerves are capsaicin sensitive and are unique in a manner that impulses in these nerves travel both orthodromically as well as antidromically, causing release of mediators at both ends.[18]

CGRP is not limited to the neural tissue and CGRP is also found in adipose tissue where it is released after inflammation and has autocrine and paracrine effects.[19] Thus a possibility remains that headache is either more common or refractory to treatment in obese individuals or subjects with higher adipose tissue.[20]

Actions of CGRP

It is most potent vasodilator in the human body, causing 71% to 92% increase in cerebral artery caliber. In human, its infusion decreases the systolic as well as diastolic blood pressure which can be corrected after a short period of cessation of infusion.21 Whether endothelium is important for the action of CGRP or not is a debatable issue, since few studies suggest that CGRP induces eNOS in the endothelium and thereby causes vasodilatation.[21,22] Whereas other studies have suggested that endothelium is not important for the action of CGRP as it does not act when injected intra-luminally in a vessel but induces dilatation when applied abluminally.[23] Moreover, CGRP antagonists introduced in the lumen could not reverse its action while those applied abluminally reversed it. This data confirms that CGRP receptors are located in the perivascular spaces, most likely perivascular nerves or that CGRP acts through the smooth muscles of the arterioles and endothelium acts as a perfect barrier which does not allow these molecules to reach smooth muscles.[23,24] Besides vasodilatation, it also has inflammatory properties and it acts throughout the inflammatory process as it action lasts longer even than that of nerve growth factor (NGF), IL-beta and TNF-Alpha. [25]

In cranial vasculature, dural nerve fibers containing CGRP are in close approximation with mast cells. CGRP degranulates the mast cells and releases histamine which can be blocked by CGRP antagonist.[26,27] It shows that these mast cells contain receptors for CGRP while such receptors are not found in endothelium. It may be one reason why intra-luminal CGRP does not cause vasodilatation. Released histamine in turn causes opening of tight junctions in dural arterioles and plasma extravasation thus resulting in sterile neurogenic inflammation. It is also observed that application of histamine to peripheral nerve endings of trigeminal nerves cause SP and CGRP release from the nerves.[28] Hence, it is possible that both the molecules are engaged in a vicious cycle to enhance each other's concentration. It also causes release of nitric oxide and TNF which take part in inflammation.

Interactions of CGRP with other molecules

Actions of CGRP are dependent upon other molecules like histamine, nitric oxide and adenosine A1 receptors etc. Nitric oxide is intimately linked to the CGRP at least in the trigeminal nucleus. Intravenous injection of nitric oxide donors are known to induce c-fos immunoreactivity in trigeminal nucleus caudalis and it was shown to be inhibited by the NOS inhibitor.[29,30] Adenosine is a purine analogue and plays important role in blocking nociception via adenosine A1 receptors. Its agonist GR79236 can undo the hemodynamic changes in carotid artery observed after capsaicin injection without affecting the concentration of plasma CGRP, indicating the action on postsynaptic receptors.[31] This molecule also acts on pre-synaptic receptors as it has been shown to decrease plasma CGRP level after electrical stimulation of sensory nerves without concomitant vasoconstriction.[32]

Role of CGRP in Migraine

Studies demonstrating the role of CGRP in the migraine headache have used one of the

following four methods: (i) studies that examined the CGRP during spontaneous headache; (ii) studies that demonstrated higher concentration of this peptide during experimental induced headaches; (iii) other had demonstrated that anti-migraine drugs decrease the concentration of CGRP in blood and, lastly (iv) that CGRP antagonists reverses the headache.

(a) CGRP during spontaneous headache

During spontaneous migraine with and without aura episodes, CGRP was found to be increased in the cubital venous blood. Its role in the pathogenesis of pain was based on the evidences that its levels were not different between patients and controls during inter-ictal period levels. Moreover, its concentration also varies during a single attack as levels are significantly higher when sample are taken closest to headache onset i.e., samples collected within two hours of start of headache had higher concentrations than those collected after 3 or 4 hours, even when samples were taken in different subjects.33 Not only the presence of migraine, but also its subtype may affect its concentration. Subjects with migraine with aura (MA) were found to have slightly higher level than migraine without aura (MO) patients despite the fact the MA subject had less frequent attacks than the counterparts.[33,34] Since cubital vein sample does not provide any evidence regarding the exact source of the peptide, it was proposed that a more appropriate method would be to measure the concentration of CGRP in the blood coming from the head and jugular venous samples were chosen for the assessment. It must be recalled here that trigeminal sensory; however during attacks patients had significantly higher levels than the inter-ictal nerves are seen along the cerebral and facial vasculature, and since activation of trigemino-vascular system is thought to play a role in migraine, a rise in peptide in jugular venous blood was expected. Furthermore, internal jugular vein contains blood coming from the cranial cavity including brain while external jugular vein drains blood from facial structures. A significant difference in the concentration of CGRP between migraine without aura and control subjects was seen during headache in the blood sample collected from external jugular vein.[34] Sarcheilli et. al.[35,36] collected internal jugular venous blood during migraine attack and found that levels of CGRP reach maximum up to one hour and then decrease despite persistence of symptoms and fall below baseline value after cessation of symptoms. This increase was corresponding to the levels of nitrite, c-GMP and c-AMP in blood. They suggested that the pain in migraine in not of extra-cranial origin and cerebral arteries dilate during the episode.

Since salivary glands have rich sensory innervations and are close to the site of pain, it was thought that saliva should contain CGRP during the headache.[37] Nicolodi and Del Bianco[37] recovered the CGRP from the saliva during spontaneous migraine without aura attacks and found that during basal conditions, migraineur's saliva contain lower level of CGRP than the controls. However, this difference was not observed during the headache phase. This suggests that CGRP level is though, lower in migraineurs under basal conditions, but does not differ markedly during headache phase from controls. It must be noted here that saliva was collected through the floor of the mouth and not from the ipsilateral side to the headache. Even then it does not explain why levels were lower during asymptomatic stage.

This peptide is elevated not only during attacks, but also outside attacks in migraineurs suggesting that trigemino-vascular system remains hyperactive even during the inter-ictal period and slightest provocation is sufficient to induce headache in migraineurs. It can be one reason why not all people develop migraine in the stressful situations. Studies have demonstrated that this increased concentration can be measured in cubital as well as internal jugular blood.[38,39]

However, these findings could not be successfully replicated across all studies.[34,40-42] Many reasons may account for differential results- its concentration in cubital vain may go below measurable level due to dilution effect or the short half life of the peptide may affect the yield as it requires meticulous sampling.

(b) CGRP in experimental migraine

Experimental migraine is a good tool to understand the patho-physiological mechanisms

of migraine. Migraine like headache was induced by a variety of methods- intravenous injections of histamine, intravenous and oral nitrates and lastly intravenous human Alpha-CGRP. During nitroglycerin induced headache, CGRP increased by 30% in the ante-cubital vein as compared to baseline and this concentration correlated with the intensity of headache.[43] In another study, migraine with and without aura was induced by cerebral angiography in otherwise spontaneous migraineurs. They reported that CGRP was not increased in the internal jugular vein during the aura phase of the attack and headache developed only after the investigation was over. Moreover, these samples did not have significantly different concentrations of peptide as compared to the samples collected from internal carotid artery. This suggests that these peptides are not involved in at least, triggering of the events and generation of aura. A more likely explanation is that intracranial structures are not involved in the genesis of migraine and the pain originates in extracranial structures.40 But this finding contradicts with the observation found in spontaneous headaches and described above. Infusion of human Alpha-CGRP in migraine without aura sufferers who had co-morbid TTH induced headache. In this experiment, initial headache which appeared after forty minutes and delayed headache also, did not match the International Headache Society symptoms of migraine without aura, except in 33% subjects. However, it did not change the cerebral blood flow and probably for the same reason MA could not be induced.[21] When these findings are seen in the light of previous literature, they support the notion that CGRP has a role in the scalp pain only and not in the pathogenesis of migraine.[40] Secondly, it goes with the previous findings that intact endothelium is a strong barrier to the action of CGRP and it can not exert its action intra-luminally.[23] Another study, where h-Alpha-CGRP was infused in healthy volunteers shows that it induces a headache that is characterized by sensation of fullness in head. This absence of migraine like headache has been thought to be related to the threshold phenomenon.[44]

Thus, experimental studies are not able to provide any conclusive evidence regarding role of CGRP in migraine development. Since migraine is a heterogeneous disorder, possibly, experimental conditions are unable to invoke the complete cascade (if any). Secondly, it is possible that CGRP is not causative in migraine but is just an epiphenomenon. However, these issues are still unresolved and require more probing.

(c) Effect of anti-migraine drugs on CGRP level

Drugs used for termination of acute migraine e.g., sumatriptan decreases the plasma CGRP concentration in the ante-cubital vein during migraine attacks elicited by nitroglycerin administration.[45] This decrease parallels clinical improvement of headache and is not seen in subjects that do not respond to these drugs. Furthermore, plasma CGRP significantly correlates with headache scores after sumatriptan treatment, as also seen in spontaneous headache previously.[33,43] These results underline the close relationship of CGRP release and migraine headache. This lowering is dependent upon the 5HT1B/1D receptors that are present pre-synaptically on the sensory nerve terminals.34,46 Although recent studies suggest that this action can be mediated through the 5HT1D receptors found on spinal trigeminal tract. In essence, these studies suggest though, does not prove the causal effect of CGRP, yet prove that pain is associated with CGRP levels and drugs affect both of them.

(d) Effect of CGRP-antagonist on migraine

With the development of biotechnology, CGRP antagonists have been developed that can terminate the migraine headache. One such molecule BIBN4096BS has been found to be as efficacious as oral sumatriptan in terminating acute attack of migraine.47 This molecule is also efficacious in terminating the attack induced by Alpha-CGRP as well as extra cerebral artery dilation.[44]

CGRP related to migraine symptoms

Few clinical symptoms of migraine were ascribed to the CGRP. These symptoms include scalp pain[23] and probably lacrimation. Intravenous infusion of CGRP induces vasodilatation in lacrimal glands which may cause tearing.[48] In addition, capsaicin selectively depletes CGRP-SP containing sensory fibers and rats treated with capsaicin show decreased

tearing on Shimmer's test.[49] Thus, these facts suggest that probably, cranial autonomic symptoms are related with the CGRP and hence, CGRP levels must be higher in migraineurs with cranial autonomic symptoms.

Unsolved issues

However, a number of questions remain to be answered; in particular, what causes the activation of the trigemino-vascular system, and how does the pain generates subsequent to its activation? Why the drug induced headache show deficit in CGRP during the attack as compared to the spontaneous headaches?

More importantly, it is not yet known whether the migraine is truly an episodic disorder or the pathology persists in between attacks? In this regard, we would like to mention at least one study that found persistently higher levels of CGRP in migraineurs, even between episodes of pain.[38] Similarly, which symptoms of migraine determine the plasma CGRP, except lacrimation, is an area for further studies.

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