Calcitonin Gene-Related Peptide(CGRP): Insights into Its *Physiological Functions* and *Pathological Implications.*

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Acknowledgement

I owe a gratitude to the concerned authorities for enabling me to compile this interesting and informative booklet titled- **Calcitonin Gene-Related Peptide(CGRP): Insights into Its *Physiological Functions* and *Pathological Implications*.**, for the enthusiastic and the busy Medicos.

It is strived in this E-Booklet to *discuss* and *describe* calcitonin gene-related peptide (CGRP), a 37-amino acid neuropeptide, focusing on its *Synthesis Regulation, Structure, Central And Peripheral Distribution,* and *Physiological Actions.*

Although care has been taken while compiling and checking information given in this Booklet to ensure that it is accurate, and none shall be responsible, in any way, liable for contemporaneousness of this information or for any errors, omissions or inaccuracies in this publication.

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Calcitonin Gene-Related Peptide(CGRP): Insights into Its *Physiological Functions* and *Pathological Implications*.

1. Introduction.

Calcitonin gene-related peptide (CGRP) is a 37-amino acid peptide found primarily in C and Aδ sensory fibers, which are widely distributed throughout the body, especially around *blood vessels*.

It has dual functions, acting in *both sensory perception* and as an *effector in various processes*.

While its role in *non-neuronal tissues* is less understood, it is known for its potent vasodilatory effects, particularly when administered in very low doses,

suggesting a *protective role* in cardiovascular health.

Initially identified for mediating sympathetic outflow from the *brain*, CGRP's *significant* cardiovascular activities and its *involvement* in pain signaling have been explored extensively.

The development of ***CGRP antagonists*** has highlighted its crucial role in *migraines*, prompting interest in *therapeutic applications* for cardiovascular diseases, although progress has been slow.

CGRP arises from the *calcitonin gene* through tissue-specific splicing, with two major *isoforms* that have similar *structures* but different *genes*.

❖ **Studies have shown that CGRP is released in response to stimuli like capsaicin, which activates *pain pathways* and is linked to sensory nerve function.**

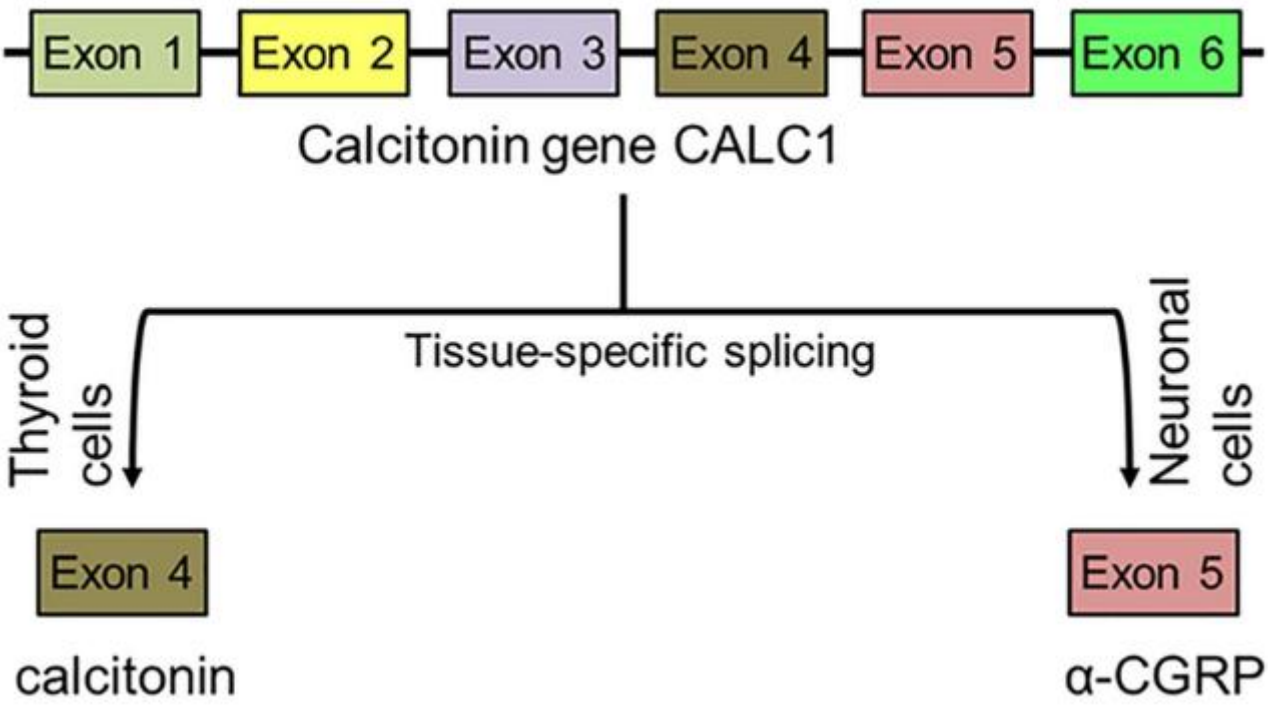
- **Its interactions with the *sympathetic nervous system* and its debated role in inflammation (either pro- or anti-inflammatory depending on context) are also *notable*.**

2. Synthesis of CGRP.

It exists in two isoforms:

1. **αCGRP and**
2. **βCGRP, derived from distinct *genes* located on chromosome 11.**

- **αCGRP is produced through alternative *splicing* of the CALC I gene, while**



Synthesis of peptide α-CGRP.

After tissue-specific alternative splicing of calcitonin gene CALC1, α-CGRP is synthesized in the neuronal cells while calcitonin is formed in the thyroid cells.

- **βCGRP is transcribed from the *separate* CALC II gene.**

Despite their differences—only three *amino acids* apart—both isoforms *share* over *90% homology*

and *exhibit* similar ***biological functions.***

- αCGRP is primarily found in the *central* and *peripheral nervous systems,* whereas
- βCGRP is predominantly located in the *enteric nervous system.*

❖ To produce *calcitonin* from the *CALC I gene*, it is necessary for *exon 4* within the *gene* to be *expressed* in the *mature protein.*

- This is the situation that occurs predominantly in the *thyroid* where *calcitonin* is the major *gene product*.
- Conversely, *expression* of *exons 5* and *6* results in production of *αCGRP mRNA*, which is first translated into a 121-amino acid pro-hormone, and then

subsequently cleaved to create the mature 37-amino acid peptide.

- *αCGRP* is predominantly expressed throughout the *central and peripheral nervous system.*
- However, the *mechanism* that determines this *alternate splicing* remains unclear.

3. Structure of CGRP.

The structure of human αCGRP has been revealed to contain four clear domains, *similar* to that of βCGRP.

The first seven residues of the *NH2 terminus* make up the first domain and form a *ringlike structure*, held together by a *disulfide bridge*.

The *peptide CGRP antagonist, CGRP8–37*, is formed from the removal of this first domain.

Residues 8–18 make up domain 2, which consists of an α helix, deletion of which causes a 50- to 100-fold decrease in affinity.

In particular, residues 11 and 18 within the *hydrophilic face* of the α-helix play a crucial role in promoting *high-affinity binding.*

Residues 19–27 make up domain 3 of CGRP and form either a *β- or γ-twist*.

The final fourth domain contains the COOH terminus and the remaining residues from 28–37 and has two turn regions that are thought to form a *binding epitope*.

The species *differences* and *structure-*activity relationships for CGRP have been extensively investigated, and key amino acids playing pivotal roles in receptor binding and activation have been identified.

4. Regulation of Synthesis and Release of CGRP

The *regulation* of Calcitonin Gene-Related Peptide (CGRP) synthesis is complex and not fully understood.

- ❖ It is known to *increase* in response to *nerve damage*, such as *peripheral axotomy*, and during *inflammatory processes.*

- This upregulation is believed to be linked to local release of nerve growth factor (NGF) from cells like *macrophages* and *keratinocytes*.

❖ NGF plays a critical role in the *growth* and *maintenance* of sensory nerves.

- After *sensory neuropeptides* are depleted due to *treatments* like capsaicin, NGF is essential for *synthesizing* new CGRP.

❖ *NGF* has been shown to enhance *CGRP production* in *dorsal root ganglia (DRG)* and is implicated in *cardiovascular* dysfunction.

❖ Elevated NGF and CGRP levels have been observed in *plasma* and *saliva* of migraine patients,

indicating a potential role in *migraine pathology*.

❖ Additionally, *brain-derived neurotrophic factor (BDNF)* might also influence CGRP release and activity.

5. Synthesis and Storage.

It is *synthesized and stored* in large, dense-core vesicles in *sensory nerve terminals*.

Its *release* occurs via *calcium-dependent exocytosis*, mediated by SNARE proteins following *neuronal depolarization*.

- **Capsaicin**, a **TRPV1 agonist**, was pivotal in **demonstrating CGRP release**, which can also be triggered by **noxious *heat*** and **low *ph*.**

The roles of **various endogenous agonists** in this process are still being explored, with substances like **rutaecarpine (a TRPV1 agonist)** showing **potential** in **promoting *CGRP release*** and having ***cardiovascular protective effects***.

- **Anandamide**, an **endocannabinoid**, has also been implicated in ***CGRP release***, **contributing** to ***vasodilation*** in various tissues.
- **Interestingly**, **estrogen** appears to **regulate this pathway**, with evidence showing that ***plasma CGRP*** levels ***decline*** in **postmenopausal women** but can be **restored** with ***hormone replacement therapy***.

6. Mechanisms of Release

Apart from *TRPV1*, other *TRP channels* like *TRPA1* are involved in *CGRP release*.

- *TRPA1* is often co-expressed with *TRPV1*, and stimulation of *TRPA1* can enhance *blood flow* through *CGRP release*, although its impact on *blood pressure* remains unclear.
- Additionally, hydrogen sulfide is noted to facilitate *TRPA1*-mediated *CGRP release*.
- CGRP is also released in response to *pressor agents* such as *angiotensin II (ANG II)* and *norepinephrine*.
  - *Norepinephrine* can inhibit sensory nerve activation via α2-adrenoceptors, with *antagonists* restoring NGF-induced CGRP release in DRG, indicating its relevance to *hypertension*.

- ❖ The compound Angeli's salt generates nitroxyl anion (HNO), which acts through *CGRP* to mediate positive *ionotropic effects* on the *heart* and *vasodilation*.
  - The *clinical implications* of this mechanism remain to be fully determined, though *new HNO donors* are being investigated for potential therapeutic benefits.

7. CGRP Metabolism

CGRP metabolism is another aspect that needs exploration to understand its overall role and regulation in *physiological and pathological states*.

Research continues to uncover the intricacies of *CGRP synthesis, release,*

and metabolism, providing *insight* into its *function* in both the *sensory and sympathetic nervous system*.

8. Central and peripheral distribution of CGRP

It is distributed throughout the *central and peripheral nervous systems*, primarily associated with *sensory neurons* like:

- Unmyelinated C fibers and
- Thinly myelinated Aδ fibers.

❖ It is often co-expressed with substance P (SP) and acetylcholine in *motor neurons*, possibly influencing *acetylcholine receptor synthesis*.

- ❖ It plays a significant role in *cardiovascular regulation*, as it is found in *perivascular nerves* that *innervate blood vessels*, with higher concentrations in arterial tissues.
- ❖ *Plasma levels* of CGRP are typically low but can *increase* under certain conditions, such as *during* pregnancy or migraine attacks.
- ❖ The two isoforms of *CGRP, α-CGRP and β-CGRP*, have distinct distributions, with:
  - α-CGRP being more prevalent in the nervous system and
  - β-CGRP is primarily found in the intestine, although both can be expressed in *various contexts.*
- ❖ Additionally, CGRP is produced not only by neurons but also by non-

neuronal cells, including *endothelial cells* and *immune cells*.

- ❖ Recent studies indicate CGRP's presence in *keratinocytes*, highlighting its broader biological roles.
- ❖ Overall, *CGRP* is *essential* for *local signaling* in vascular systems and various physiological responses.

9. Receptors

Discovery of calcitonin receptor-like receptor (CLR)

The discovery of *CGRP receptors* began with early *radioligand binding* studies that identified two receptor types, *CGRP1 and CGRP2*, based on their ability to bind *CGRP*.

Specific peptide analogs exhibited distinct *agonistic* and *antagonistic* properties across different tissues.

Over time, it became clear that only one true CGRP receptor exists, often associated with low-affinity binding to other *receptors*, such as those for adrenomedullin and intermedin.

The CGRP receptor, initially termed CLR (calcitonin receptor-like receptor), is composed of two subunits and is characterized by seven *transmembrane domains.*

Research in the 1990s revealed that CLR could stimulate significant cAMP production in human embryonic kidney cells when co-expressed with a previously unknown protein known as a *receptor activity modifying protein (RAMP).*

- ❖ **Three RAMPs (RAMP1, RAMP2, and RAMP3) are recognized, and their co-expression with CLR is essential for proper *receptor function* and *localization*.**
  - **The combination of CLR and RAMP1 forms a high-affinity *CGRP receptor*, while CLR with RAMP2 and RAMP3 creates receptors that are responsive to adrenomedullin and have varying selectivity for CGRP.**

This understanding emphasizes the importance of *receptor heterodimerization* and the role of RAMPs in receptor functionality.

10. Physiological Actions Of CGRP.

A. CGRP as a Potent Microvascular Vasodilator.

CGRP (Calcitonin Gene-Related Peptide) is recognized as the most potent *microvascular vasodilator*, with effects approximately 10-fold stronger than the most potent *prostaglandins* and 10–100 times greater than other *vasodilators* like *acetylcholine (ACh)* and *substance P (SP).*

- **Its *injections* can induce significant *reddening* of the skin due to increased blood flow, with effects lasting 5–6 hours even at *picomole* doses.**
- **It *selectively* dilates blood vessels in *cerebral, coronary*, and *kidney***

regions, with its activity inhibited by CGRP receptor antagonists.

- In humans, CGRP acts as a vasodilator and can produce *positive inotropic* and *chronotropic effects,* likely as a compensatory response to *hypotension*.
  - However, when administered directly to the *brain's ventricular system*, it can induce *hypertension* by activating *sympathetic nerves*.
  - Despite its effects, it does not play a *critical role* in regulating *systemic blood pressure* in healthy individuals, as *CGRP receptor antagonists* have *shown no significant impact* on resting *heart rate* or *blood pressure*.

- This *characteristic* makes *CGRP antagonists* promising for *migraine treatment.*

❖ CGRP-dependent vasorelaxation involves multiple *pathways,* primarily through an *endothelium-independent mechanism* that stimulates *cAMP* production in *smooth muscle cells.*

- This *pathway* leads to the activation of protein kinase A and the opening of ATP-sensitive potassium channels, resulting in *relaxation* of the vascular smooth muscle.

Overall, CGRP's robust *vasodilatory effects* and *mechanisms* highlight its potential *therapeutic implications,* especially in conditions like migraine.

- ❖ The most common pathway involves a rise in *cyclic AMP (cAMP)* after CGRP administration, indicating that CGRP directly stimulates adenylate cyclase in *smooth muscle cells* to produce *cAMP,* enabling *relaxation* even without *endothelial cells*.
- ❖ While CGRP increases *cAMP* in both *human endothelial cells* and *vascular smooth muscle cells,* it does not directly stimulate *cyclic GMP (cGMP)* production.
  - CGRP remains *effective* in *arteries lacking an endothelium,* such as those in *cat brain vessels* and *human intracranial arteries.*
- ❖ The rise in cAMP activates *protein kinase A,* which leads to the *phosphorylation* and opening of *ATP-sensitive potassium (K+) channels,* promoting relaxation.

- **Blocking these channels with *glibenclamide* inhibits CGRP-induced relaxation by *preventing* arterial smooth muscle hyperpolarization.**

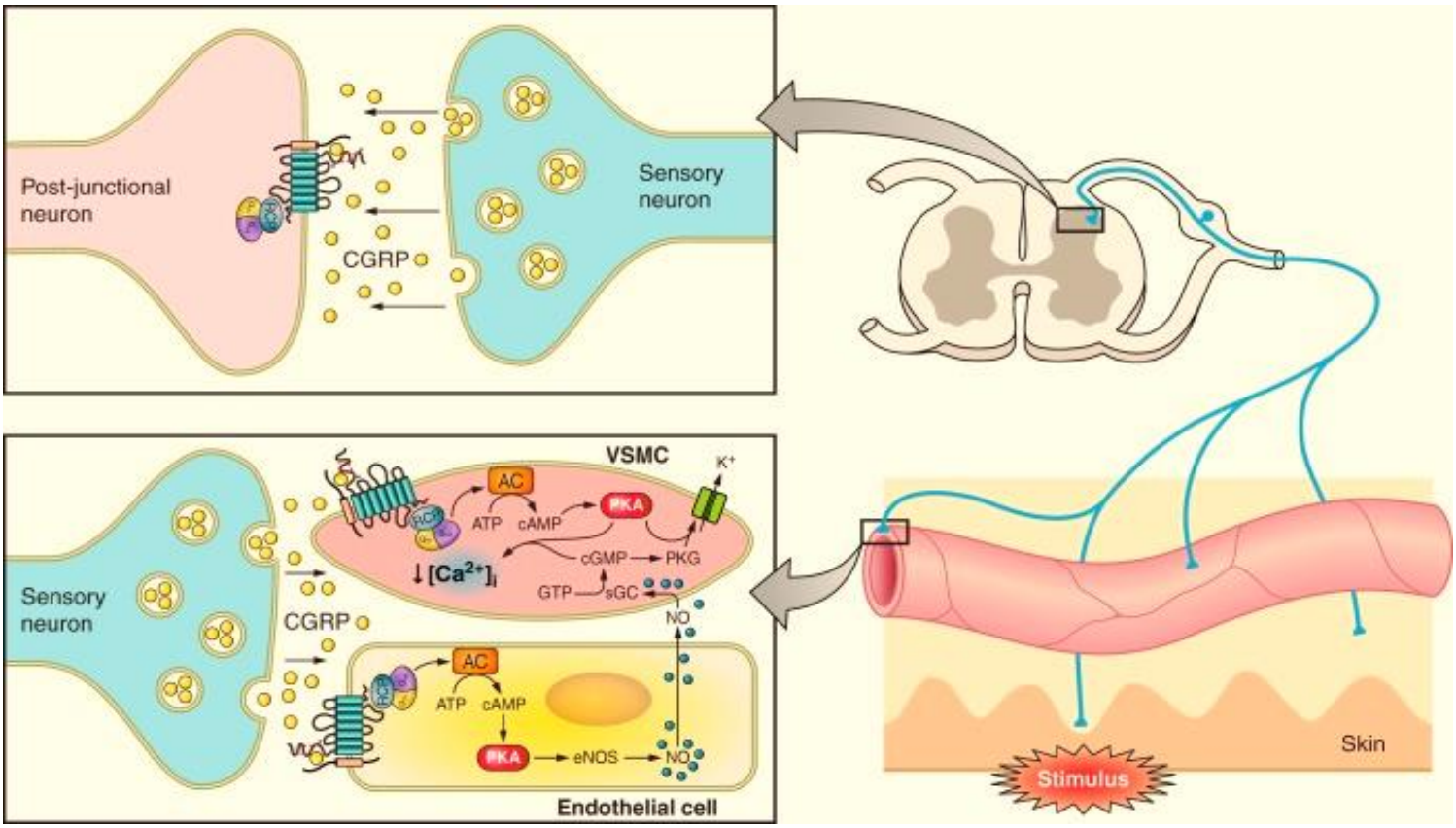


Fig.1. ***A diagram representing the interactions between the sensory nerves, skin, and arterioles.***

11. Involvement in cardiovascular disease

A. CGRP and Hypertension.

Evidence suggests that CGRP is crucial for counteracting the onset of *hypertension,* though its effectiveness varies depending on the *specific experimental model* used.

- For instance, in certain *kidney-related models*, such as the
  - Doca,
  - Two-kidney one-clip, and
  - Phenol models, CGRP shows increased *synthesis* and *release* as a compensatory or protective response.
- In contrast, reduced CGRP activity is associated with worsened *cardiovascular* outcomes in

models like the spontaneously hypertensive rat (SHR) and CGRP knockout mice.

Overall, these findings highlight the complex and context-dependent role of CGRP in *blood pressure* regulation.

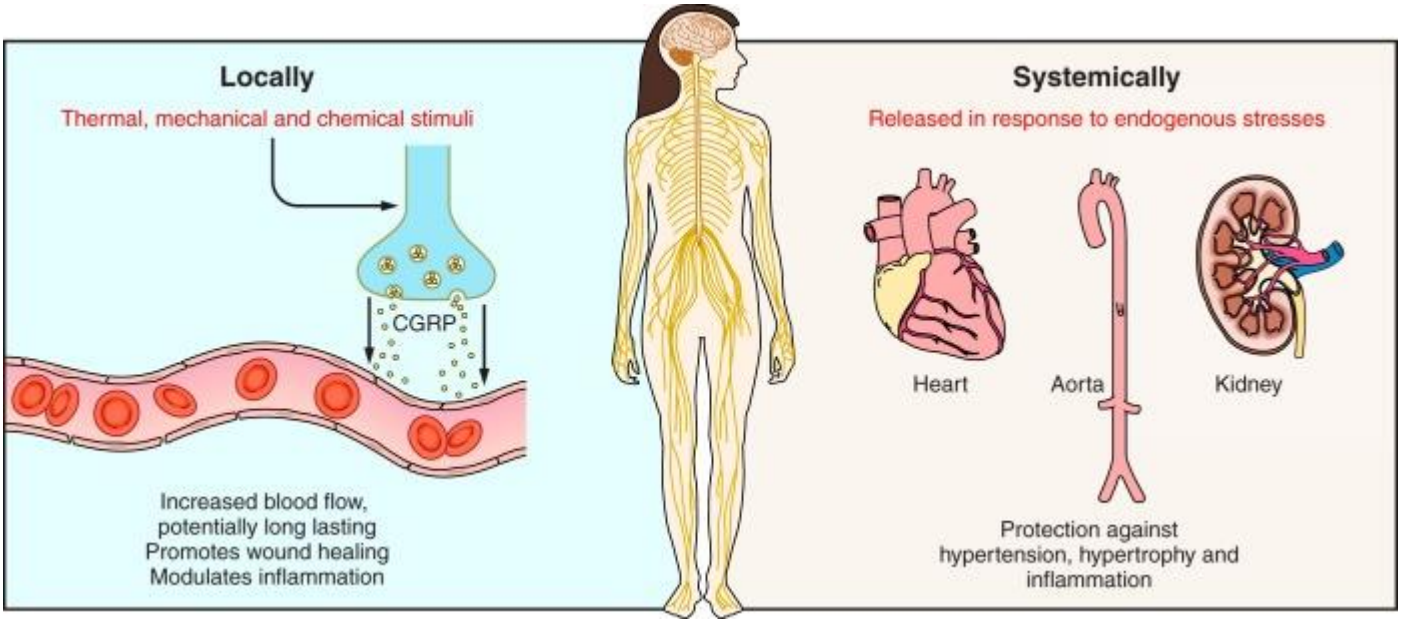


Fig.2. *Local and systemic mechanisms involving CGRP in cardiovascular regulation.*

B. CGRP and the sympathetic system

A subpressor dose of CGRP can counteract the effect of *norepinephrine* as well as *ANG II*, in short-term 6-day

studies, leading to the suggestion that CGRP acts via an effect on *peripheral resistance*.

- ❖ Kawasaki et al. showed that the sympathetic nervous system signals to the sensory CGRP system via *α2 adrenergic receptors*.
  - The α2 receptor has been suggested to mediate the *inhibition* of NGF-stimulated CGRP release in the DRG.
  - This has been further investigated in a phenol model of *hypertension,* where intrarenal injection of phenol is associated with *hypertension,* decreased sensory nerve activation, and, in turn, reduced CGRP.
- ❖ While an *α1 receptor antagonist* was found to reduce the

hypertension, only an *α2 receptor antagonist* was able to restore CGRP levels (via a mechanism involving *NGF*), but the *α2 antagonist* was not able to reduce BP.

This suggests that even in the *hypertensive state* when CGRP levels are *raised* sufficiently to reach plasma, *CGRP* does not *influence BP regulation* in this hypertensive model.

C. CGRP and Pulmonary Hypertension

Research on *CGRP (Calcitonin Gene-Related Peptide)* in pulmonary hypertension is significant due to the *limitations* and *side effects* of current *treatments*.

- αCGRP is highly present in the *lungs,* particularly in *nerve fibers* of

the airway mucosa and epithelium. CGRP can *dilate precontracted* pulmonary arteries in vitro and protect against hypoxia-induced tissue remodeling in pulmonary hypertension.

- ❖ **Adenovirus-mediated delivery of CGRP** to mouse lungs prior to *chronic hypoxia* demonstrated *protective effects, r*educing *pulmonary vascular resistance* and remodeling through *cAMP-induced vasodilation* and suppression of *endothelin-1 (ET-1)* and *angiotensin II (ANG II) release.*
- ❖ Furthermore, the loss of *capsaicin-sensitive nerves,* which *release* CGRP via TRPV1 activation, may worsen *pulmonary hypertension*.

- In studies where endogenous CGRP was depleted, *hypoxia-induced pulmonary hypertension* was exacerbated, but exogenous CGRP application *mitigated* pulmonary arterial remodeling by *inhibiting* the ERK1/2 pathway.
- Injecting endothelial progenitor cells (EPCs) engineered to express CGRP also reduced established *pulmonary hypertension* and *vascular remodeling*.

Overall, these findings suggest that CGRP and CGRP-overexpressing EPCs could be promising therapies for *pulmonary hypertension.*

D. CGRP and Heart Failure.

CGRP (Calcitonin Gene-Related Peptide) is found throughout the *heart's*

vasculature, particularly around *coronary arteries* and important *cardiac nodes,* suggesting its role in *sensory* and *efferent* functions.

- Studies indicate that CGRP can promote *positive effects* on *heart function,* including *vasodilation* and protective responses during *cardiac stress.*
- In models of *heart failure*, such as those *induced* by isoprenaline, CGRP release has been linked to *reduced* hypertrophy and *improved* cardiac function.
- Increased CGRP levels have been observed in *genetically predisposed individuals* with heart failure, and CGRP infusion has shown *beneficial effects* on circulation in heart disease.

- ❖ **Research using knockout (KO) mice lacking CGRP revealed *worse cardiac remodeling* and *survival rates* compared to wild-type (WT) mice, highlighting *CGRP's protective role* against inflammation and fibrosis.**
- ❖ **CGRP is also recognized as a *proangiogenic factor*, promoting *microvascular growth* and protecting against *cell death in cardiac tissues*.**
  - ***Activation* of protective kinases by CGRP is thought to inhibit *apoptosis in cardiomyocytes*, with studies showing that CGRP can *counteract* the apoptotic effects of *norepinephrine and oxidative stress*.**

Overall, CGRP appears *crucial* for maintaining *cardiac health* and

mitigating damage in various *stress conditions.*

E. Ischemia.

CGRP (Calcitonin Gene-Related Peptide) plays a *critical role* in *cardiac protection* during ischemic events, acting as a potent *endogenous mediator* of preconditioning—where prior exposure to a *stressor* reduces subsequent *ischemic damage*.

- In humans, plasma levels of CGRP increase following *acute myocardial infarction,* indicating its release in response to *ischemic stress*.
- CGRP's protective effects extend to *various organs*, including the gut, kidney, and brain.
  - While CGRP levels are reduced in patients with *coronary artery*

disease (CAD), its exogenous administration has shown mixed results, with some studies indicating it can *prolong exercise tolerance.*

❖ The role of CGRP in *preconditioning* is especially relevant in diabetic patients, where *ischemic protection* is significantly diminished but can be *restored* by *CGRP gene transfection.*

- Age and diabetes appear to *impair* CGRP-mediated cardio protection, likely due to *sensory nerve degeneration.*

Overall, CGRP's protective role in ischemia suggests it may be a *potential therapeutic target,* especially in vulnerable populations such as the *elderly a*nd those with *diabetes.*

F. CGRP, Atherosclerosis, and Vessel Remodeling.

- ❖ **CGRP (Calcitonin Gene-Related Peptide) has a *protective role* in *cardiovascular diseases* such as:**
  - **Atherosclerosis,**
  - **Hypertension, and**
  - **Vessel remodeling.**

- **These conditions are *associated* with:**
  - **Increased blood pressure (BP),**
  - **Vascular inflammation,**
  - **Endothelial dysfunction, and**
  - **Smooth muscle cell (VSMC) hypertrophy and proliferation.**

It has been shown to *counteract* some of these processes, offering:

- **Protection against vessel injury,**
- **Remodeling, and**
- **Plaque formation.**

- While CGRP's specific role in *atherosclerosis* is less well-documented, it has been shown to *inhibit* cell proliferation in response to *vascular injury* by *modulating* growth factors, and it is protective in *models* of *neointimal hyperplasia (thickening of the vessel wall)* and *endothelial dysfunction.*
- In genetic models, CGRP knockout (KO) mice show *increased neointimal formation,* further supporting CGRP's *Vaso protective* role.
- Overexpression of CGRP receptors (RAMP1) has been shown to protect against *ANG II-induced endothelial dysfunction* by enhancing vasodilation.
- CGRP also exhibits *anti-inflammatory properties,* reducing the

migration of *immune cells (neutrophils and monocytes)* to the *endothelium* and suppressing *inflammatory markers* like MCP-1 and VCAM-1, which are associated with *vascular inflammation* and *atherosclerosis.*

- CGRP is also involved in vascular endothelial repair, particularly in *endothelial progenitor cells (EPCs).*
  - These cells, which help repair *damaged blood vessels*, produce CGRP that protects against *senescence* and *VSMC hypertrophy* in hypertension.

CGRP therapy has been shown to reduce vascular resistance and inhibit *vessel wall thickening* in models of *pulmonary hypertension*.

Overall, CGRP appears to play a protective role in *cardiovascular diseases* by reducing *vascular inflammation*, inhibiting VSMC proliferation, and promoting endothelial repair.

G. Sepsis.

CGRP (Calcitonin Gene-Related Peptide) *plasma levels* increase in patients with *sepsis* and are thought to contribute to *hypotension*.

In contrast, CGRP may also have *protective effects*, such as *reducing* the production of certain *proinflammatory cytokines* and protecting against *fatal endotoxic shock* in mice.

RAMP-1-deficient mice lose *immunosuppression,* while TRPV1

deletion is linked to *worsened* sepsis due to *reduced* CGRP release.

- ❖ Some studies indicate that CGRP is *released* early during *infection*, acting anti-inflammatory by promoting IL-10 *production* among other effects.
- ❖ However, in mixed-bacterial infections, excessive CGRP may lead to i*mmunosuppression* and compromise *host defense*.
  - This area remains complex, with varying findings on CGRP's role in *inflammatory responses* during sepsis.

12.CGRP and other Pathophysiological conditions

A. CGRP in Neurogenic Inflammation and Pain.

Sensory nerve fibers are branched in a *collateral manner*; it is thought that when a *peripheral terminal* of a *sensory nerve* is activated, action potentials are not only transmitted to the *dorsal horn,* but are also transmitted *antidromically* down the *branches*.

- This results in *neuropeptide release* from the *peripheral branches* (Fig.1 above).
- It is this *axon reflex (a reflex in a single neuron)* that is thought to account for the *flare* seen after *intradermal injection* of histamine into *human skin* which can extend

for several *centimeters* away from the injection site.

- ❖ *Neurogenic inflammation* is the term given to the *microvascular effects* caused by the release of *neuropeptides* from the nerve terminals (Figure 1).
- ❖ The two main *proinflammatory neuropeptides* are CGRP and substance P (SP).
  - SP acting on *NK1 receptors* is a potent mediator of i*ncreased microvascular permeability* and, CGRP is an extremely potent vasodilator, thus release of these *peptides* results in:
    - Oedema formation,
    - Increased blood flow, and
    - Recruitment of inflammatory cells to the local area.

The role of CGRP in inflammation has been reviewed.

In general, CGRP has been suggested to mediate a host of *immune regulatory responses*, relevant to *host defense* and *inflammation*, but few have been fully supported by strong subsequent in vivo studies.

However, without doubt, depending on the situation, CGRP is *able to promote inflammation*, secondary to vasodilatation, and inhibit *inflammation/mediator release* through its ability to increase cAMP.

Thus, to a certain extent, it depends on when and how CGRP is released as to its net effect.

- There is a strong indication that CGRP has the ability to inhibit

lymphocyte differentiation and proliferation.

- CGRP has also been suggested to influence the activity of other inflammatory cells, including Langerhans cells and macrophages.
- CGRP can inhibit IL-1β-induced reactive oxygen species generation in alveolar epithelial cells.
- Overall, it is considered that CGRP can inhibit allergic conditions such as irritant dermatitis.
- CGRP, via increases in cAMP, was able to inhibit ovalbumin-induced airway inflammation in the mouse.
    - In keeping with this, CGRP deletion is associated with an inhibition of airway hyperresponsiveness .

- **CGRP has been proposed to influence neutrophil-endothelial cell interactions in a positive manner, and also a negative manner.**
  - **The *positive effect* has been suggested to be secondary to microvascular dilation.**
    - **Monneret et al.have shown that CGRP can inhibit cytokine production from LPS- and fMLP-stimulated blood cells, secondary to increasing cAMP.**
    - **An inhibition of chemokines (CXCL1, CXCL8, and CCL2) from LPS-stimulated cultured human microvascular endothelial cells *in vitro* was observed, and this correlated with the ability of local and systemic CGRP to inhibit monocytes and**

neutrophils in response to LPS in the mouse peritoneal cavity.

- This inhibition of recruitment may be secondary to *systemic hypotension* in this model.
- In vivo, *deletion* of RAMP1 is associated with *hypertension and proinflammatory cytokine* production, providing further evidence for an inhibitory role of CGRP.

❖ In the kidney, CGRP has been shown to be *proinflammatory* in a model of obstructive nephritis, and this may in part be related to the potentiating vasodilator activity, as inflammatory markers, as well as resulting fibrosis were obvious.

❖ It has long been known that capsaicin-sensitive neurons are

important in neurogenic inflammation, since the *hypersensitivity* and *flare* produced from intradermal injection of capsaicin can be prevented by denervation or by prolonged exposure to capsaicin that produces sensory nerve desensitization.

- ❖ In humans, the role of CGRP has been examined in a model of increased dermal blood flow induced by topical application of *capsaicin* .
  - Pretreatment with the CGRP antagonist telcagepant inhibited this increased blood flow after capsaicin, demonstrating a key role for CGRP in *neurogenic vasodilatation* in humans .
- ❖ Hyperalgesia is the heightened sensitivity to painful stimuli and

occurs under conditions that lead to *sensitization* of peripheral nerve terminals (peripheral sensitization), or *sensitization* of the spinal cord dorsal horn (central sensitization).

- Perhaps more focus has been given to the tachykinin neuropeptide SP, acting on the NK1 receptor on the dorsal horn after high-intensity stimuli and contributing to central sensitization.

❖ The injection of CGRP into human skin is not associated with *axon-reflex flare* initiation or increased nociception.

❖ Mechanical hyperalgesia caused by *intrathecal CGRP* has been observed, and electrophysiological data show *CGRP-induced*

sensitization of spinal dorsal horn neurons.

- ❖ It may be that the role of CGRP in pain becomes more important under conditions of abnormal pain processing such that occurs during inflammatory or neuropathic pain states.
  - Additionally, expression of CGRP mRNA was upregulated bilaterally in *trigeminal neurons* in a chronic inflammatory rodent model of periodontitis induced by LPS.Reduced pain thresholds are not observed in this model, or in patients with periodontitis; thus it is proposed that the upregulation of CGRP is involved with the *inflammatory process* and not *nociception*.

The discrepancy in results may be due to the *different models, experimental conditions,* and the many different ways of measuring pain.

However, an intriguing report by Mogil et al. provides evidence that different mouse strains differ widely in their expression of the CGRP gene and this correlates with their variable thermal sensitivity.

- In mouse strains that expressed low CGRP levels of CGRP, peripheral injection of CGRP into the hindpaw caused a decrease in thermal nociceptive threshold.
    - In comparison, CGRP-rich strains, which include C57/BL6 mice, were insensitive to CGRP injection.

- Thus the it is suggested that unknowingly using these CGRP-rich strains would lead to the failure to demonstrate CGRP effects.
- Perhaps as a matter of course all CGRP studies should look at gene expression and levels of CGRP release to be aware of this factor.

B. CGRP and Migraine

Migraine is a complex *neurovascular disorder* affecting a significant portion of the population, *particularly women*, and is characterized by debilitating *headaches* and a range of symptoms including:

- Light and sound sensitivity,
- Nausea and
- Dizziness.

The exact cause of *migraines* remains unclear, but *calcitonin gene-related peptide (CGRP)* is recognized as a key player in their *pathophysiology*.

- CGRP is released from *trigeminal nerve fibers* that innervate the *meningeal blood vessels*, especially during *migraine attacks*.
  - Studies have shown that *intravenous CGRP* can *trigger* migraine-like attacks in *susceptible individuals*, and elevated plasma CGRP levels have been observed during *migraines*.
  - Notably, these *levels* can *decrease* with t*reatment* using triptans, which are *effective anti-migraine agents*.

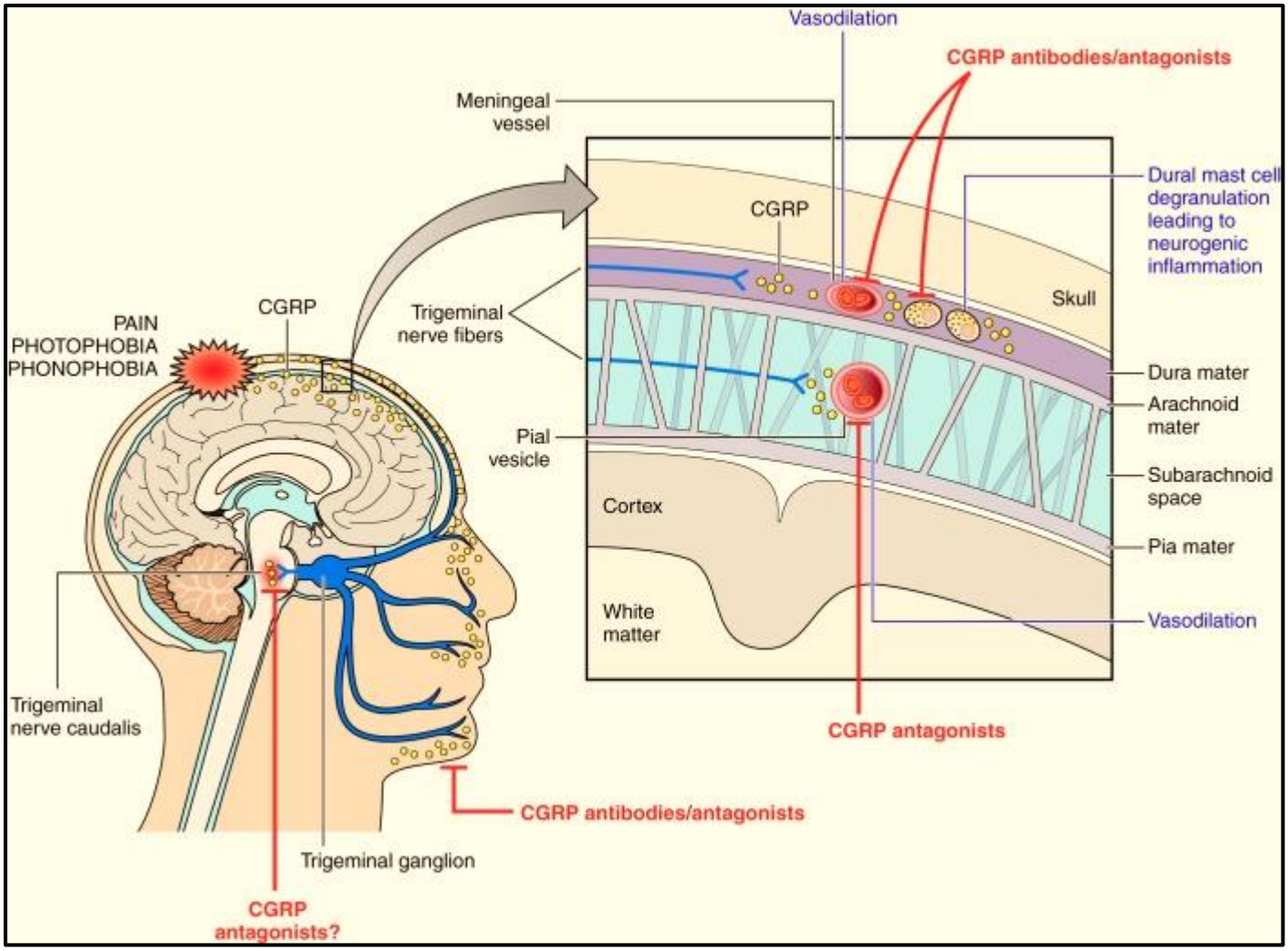


Fig.3.*Involvement of GRP in the pathophysiology of migraine.*

However, the relationship between CGRP and migraines is not entirely straightforward.

- **Some studies have reported no significant increase in *CGRP levels* in certain migraine patients, suggesting that *individual* genetic factors may**

influence CGRP's role in *migraine susceptibility*.

Given the significant role of calcitonin gene-related peptide (CGRP) in migraines, there has been *extensive research* to develop CGRP antagonists as potential treatments.

Current migraine medications, such as triptans, have limitations, including cardiovascular side effects and issues with medication overuse.

- ❖ The first selective nonpeptide CGRP antagonist, BIBN4096BS (Olcegepant), was developed by Boehringer Ingelheim in 2000.
  - This drug effectively blocked *trigeminal nerve-induced vasodilation* without affecting cardiovascular parameters.

❖ In clinical trials, BIBN4096BS showed a 60% response rate in treating migraines, comparable to *triptans*.

However, its high molecular weight necessitated *intravenous administration*, making it less practical for acute migraine treatment.

❖ In response, researchers focused on developing an orally available CGRP antagonist.

- Merck's MK-0974 (telcagepant) emerged in 2007, demonstrating strong efficacy and good oral bioavailability in phase II studies. A
- A larger *phase III trial* confirmed telcagepant's effectiveness in relieving *migraine pain* and *associated symptoms*, with fewer

adverse effects compared to *triptans*.

- Further studies suggested that combining telcagepant with other analgesics like *ibuprofen or acetaminophen* could enhance treatment efficacy, though results varied between groups.

Overall, *CGRP antagonists* represent a *promising advance* in migraine management.

1) Telcagepant and CGRP Antagonists in Migraine Treatment

A CGRP antagonist was evaluated in a *large phase III trial* that demonstrated its effectiveness in treating migraine symptoms across multiple attacks.

- Both 140 mg and 280 mg doses showed significant efficacy compared to placebo, with minimal

adverse effects primarily limited to drowsiness and vomiting.

- However, concerns arose during a *prophylactic trial*, where *elevated liver transaminase levels* led to its early termination.

A long-term study involving 19,820 migraine attacks treated with *telcagepant* indicated it had fewer side effects than the triptan rizatriptan, maintaining consistent efficacy without tolerance development.

❖ In trials involving patients with stable *coronary artery disease*, *telcagepant* was well tolerated with no serious *cardiovascular side effects.*

- Despite these promising results, Merck discontinued telcagepant's

development in 2011 due to ongoing liver toxicity concerns.

2) A second CGRP antagonist, MK-3207.

It showed *higher potency and efficacy* in *phase II trials*, but it too faced liver toxicity issues, leading to its *discontinuation*.

3) Another candidate, BI 44370 TA from Boehringer Ingelheim, demonstrated significant *pain relief* in early trials without major adverse events, though larger studies are needed for *confirmation*.

4) Bristol-Myers Squibb has been developing two CGRP antagonists:

1. BMS-927711 and
2. BMS-742413.

1. BMS-927711

This showed *good efficacy* and *tolerability* in early trials.

2. BMS-742413.

Designed for *intranasal delivery,* exhibited no vasoconstrictor effects on *coronary arteries.*

- ❖ In contrast to small molecule antagonists, *monoclonal antibodies* targeting CGRP are *emerging* as a promising alternative.

- Several candidates are in trials, including:
  - LY2951742,
  - ALD403, and
  - AMG 334, with early results indicating effectiveness without significant liver toxicity.

Despite the potential of these treatments, the exact mechanisms underlying migraine pain remain unclear, and further research is needed to fully

understand the role of CGRP and other mediators in migraine pathology.

Ultimately, while CGRP antagonists and monoclonal antibodies show promise, ongoing challenges such as *safety, administration methods*, and *long-term effects* need to be addressed.

C. CGRP and Arthritis

Chronic inflammatory joint disorders, which affect about 20% of adults in the Western world, are associated with *elevated levels* of calcitonin gene-related peptide (CGRP) in both plasma and synovial fluid of arthritic patients.

- Research indicates that *CGRP* may act as *an early mediator* in *arthritis*, with *increased* mRNA levels detected shortly after *inflammatory triggers* in *rodent* models.

- Modulating *CGRP activity* has shown to influence key aspects of *arthritis,* such as *cytokine production* and the *proliferation* of *synovial cells* in rheumatoid arthritis (RA) and osteoarthritis (OA).
- CGRP antagonists, like **CGRP8–37,** can inhibit *inflammatory responses* and the production of *pro-inflammatory cytokines* and *matrix metalloproteinases.*
- Interventions such as *capsaicin application* and *surgical denervation* have been found to lower CGRP levels and mitigate *joint inflammation* in animal studies.
- Moreover, increased *sensory innervation* and *CGRP-positive nerve fibers* are prominent in arthritic joints, contributing to:

- Neurogenic inflammation and
- Angiogenesis.

- *Arthritis* can be classified into:
  - Degenerative types like OA and
  - Inflammatory types like RA.

While CGRP has been linked more closely with inflammatory arthritis, it also plays a *role* in OA, *challenging* the notion that OA is solely a degenerative condition.

- Studies have shown *elevated CGRP expression* in OA models and in patients with hip OA, suggesting its *involvement* in *pain and inflammation* in OA as well.
- Recent findings highlight the potential *of CGRP-targeting therapies* for *arthritis pain* management.

- For instance, the neutralizing CGRP antibody LY2951742 demonstrated *pain relief* in OA models through mechanisms distinct from traditional NSAIDs, indicating its broader *therapeutic potential* beyond *migraine treatment*.

D. CGRP in Skin.

CGRP plays a significant role in *skin physiology,* primarily through *its potent vasodilatory effects,* which lead to increased *blood flow*.

❖ It is released from:

- Sensory neurons,
- Keratinocytes, and possibly
- Immune cells, affecting local blood flow and potentially mediating skin reflexes such as *flushing.*

- While CGRP can enhance oedema and leukocyte accumulation in response to *inflammatory mediators*, evidence also suggests it can inhibit certain *endothelial functions*, complicating its role in *inflammation*.
- Scratching has been shown to increase CGRP levels, possibly linked to nerve growth.
- CGRP may *influence* immune responses, particularly in conditions like *atopic dermatitis*, by enhancing IL-13 production, which is associated with worsening *symptoms*.
    - Its *antagonist*, CGRP8–37, has been noted to reduce itch responses.
- Furthermore, *studies* indicate *CGRP supports* keratinocyte proliferation and wound healing

processes, with CGRP *depletion* leading to *impaired* tissue repair.

E. CGRP in Wound Healing.

Research shows *CGRP facilitates* tissue repair and angiogenesis, essential for *effective* wound healing.

- *Depletion* of CGRP through capsaicin leads to *poor healing* outcomes in animal models.
- Conversely, *administering* CGRP improves *blood flow* and *enhances healing* in various wound models.

❖ Genetic studies indicate that *absence* of CGRP results in delayed *wound closure* and *angiogenesis*.

F. CGRP in Diabetes and Obesity.

- ❖ **CGRP is involved in insulin regulation and is localized in pancreatic β-cells.**
  - **It typically reduces *glucose-stimulated insulin release* and is linked to *insulin resistance.***
- ❖ **Studies using *CGRP knockout mice* suggest that the absence of CGRP may protect against *diet-induced obesity* and improve *metabolic parameters*, indicating potential *therapeutic targets* for obesity treatments.**
- ❖ **CGRP levels are *often altered* in diabetes, with potential associations between decreased CGRP and adverse cardiovascular outcomes in *diabetic patients*.**

- ❖ Investigations suggest that CGRP might play a role in both the neuropathy seen in *diabetes* and the overall *metabolic dysregulation* associated with the condition.

G. CGRP and Aging.

- ❖ Aging negatively impacts *sensory neurons,* potentially diminishing *CGRP activity*.
- ❖ Aging also affects *CGRP's role* in cardiovascular protection, with older individuals showing *reduced responsiveness* to CGRP.

13. Conclusions.

CGRP is **increasingly *recognized*** for its **multifaceted roles** in:

- **Pain,**
- **Inflammation,**
- **Wound healing,**
- **Metabolic regulation, and**
- **Vascular health.**

Despite significant *research*, many fundamental *questions* remain regarding its ***mechanisms*** and ***therapeutic potential,*** especially in **aging and chronic conditions** like ***diabetes and obesity***.

The **ongoing development** of **CGRP-targeted therapies** may further elucidate its ***functions*** and open avenues for **clinical applications**.

Additionally, ***CGRP replacement therapy,*** such as with **stabilized CGRP**

agonists, could become a potential treatment option.

Modulating CGRP *synthesis* and *release*, either chemically or electrically (e.g., through transcutaneous electrical stimulation for bladder dysfunction), is another avenue of investigation.

Despite decades of study, *significant questions* about CGRP's functional relevance and its therapeutic potential, especially regarding *diabetes*, remain unresolved.

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