REVIEW

Neurogenic Vasodilatation and Plasma Leakage in the Skin

Peter Holzer

UNIVERSITY OF GRAZ, DEPARTMENT OF EXPERIMENTAL AND CLINICAL PHARMACOLOGY, UNIVERSITATSPLATZ 4, A-8010 GRAZ, AUSTRIA
[TEL: [+43] (316) 380-4500; FAX: [+43] (316) 380-9645; E-MAIL: peter.holzer@kfunigraz.ac.at]

ABSTRACT. 1. Primary afferent nerve fibers control cutaneous blood flow and vascular permeability by releasing vasoactive peptides. These vascular reactions and the additional recruitment of leukocytes are commonly embodied in the term neurogenic inflammation.

2. Calcitonin gene-related peptide (CGRP) acting via CGRP<sub>1</sub> receptors is the principal transmitter of neurogenic dilatation of arterioles whereas substance P (SP) and neurokinin A (NKA) acting via NK<sub>1</sub> receptors mediate the increase in venular permeability.

3. Neurogenic vasodilatation and plasma protein leakage play a role in inflammation because many inflammatory and immune mediators including interleukin-1β, nitric oxide, prostanoids, protons, bradykinin, histamine, and 5-hydroxytryptamine can stimulate peptidergic afferent nerve fibers or enhance their excitability.

4. Neurogenic inflammatory reactions can be suppressed by α<sub>2</sub>-adrenoceptor agonists, histamine acting via H<sub>1</sub> receptors, 5-hydroxytryptamine acting via 5-HT<sub>1B</sub> receptors, opioid peptides, and somato- statin through prejunctional inhibition of peptide release from vasoactive afferent nerve fibers. CGRP, SP, and NKA receptor antagonists are powerful pharmacological tools to inhibit neurogenic inflammation at the postjunctional level.

5. Imbalance between the facilitatory and inhibitory influences on afferent nerve activity has a bearing on chronic inflammatory disease. Impaired nerve function represents a deficit in skin homeostasis while neuronal overactivity is a factor in allergic and hyperreactive disorders of the skin.

KEY WORDS. Afferent neurons, neurogenic inflammation, vasodilatation, plasma protein extravasation, leukocyte emigration, calcitonin gene-related peptide, substance P, nitric oxide, immune system, allergy, chronic inflammation

NEUROGENIC VASODILATATION AND INFLAMMATION

It has long been known that primary afferent neurons innervating the skin can function as vasodilator neurons (Geppetti and Holzer, 1996). Their ability to regulate cutaneous blood flow has directly been proved by the phenomenon of antidromic vasodilatation, which is due to the release of vasoactive transmitters from the peripheral varicosities of afferent neurons. Antidromic vasodilatation is tightly interwoven with another phenomenon, the axon reflex flare of Thomas Lewis' triple response to injury (Brain, 1996; Holzer, 1992). The hyperemia (flare) that spreads beyond a pin-point injury of the skin is thought to arise from axon reflexes that take place entirely between the arborizing collaterals of single sensory nerve fibers. Afferent nerve-mediated vasodilatation may be associated with an increase in vascular permeability and recruitment of leukocytes (Geppetti and Holzer, 1996), and these reactions to sensory nerve stimulation are commonly embodied in the term neurogenic inflammation (Fig. 1).

The sensory nerve fibers that evoke hyperemia and plasma leakage can pharmacologically be manipulated by capsaicin which is a selective excitotoxin for thin afferent neurons (Holzer, 1991). Capsaicin-induced stimulation of sensory nerve fibers increases blood flow and vascular permeability in the skin, whereas pretreatment with a neurotoxic dose of capsaicin prevents these neurogenic changes in vascular function. Capsaicin has in addition been instrumental in the identification of calcitonin gene-related peptide (CGRP) and the tachykinins substance P (SP) and neurokinin A (NKA) as the afferent nerve transmitters which mediate neurogenic vasodilatation and plasma protein extravasation (Brain, 1996; Holzer, 1992). On excitation, these peptides are released from the varicosities of afferent nerve fibers and reach their target receptors by diffusion.

This minireview attempts to briefly outline the basic mechanisms and mediators by which afferent nerve fibers regulate the cutaneous circulation and to discuss the mechanisms that govern activation and termination of neurogenic vasodilatation and plasma leakage with a perspective for inflammatory disease.

NEUROGENIC MEDIATORS OF CUTANEOUS VASODILATATION CGRP

It is widely accepted that CGRP or a closely related peptide is the major mediator of neurogenic vasodilatation in the skin (Fig. 1) CGRP is released from cutaneous afferents in response to nerve stimulation (Holzer, 1992), and is most active in dilating cutaneous arterioles of all species that have been tested (Brain, 1996). The
Neurogenic inflammation

Afferent nerve ending  

Stimulus  

Positive feedback: sensitization  

Negative feedback: inhibition of peptide release  

SP / NKA  

Increased venular permeability (NK1 receptors)  

Arteriolar dilatation (CGRP receptors)  

Proteases  

Histamine  

5-Hydroxytryptamine  

Prostaglandins  

Nitric oxide  

SP  

Mast cells  

Cytokines  

Prostaglandins  

Leukotrienes  

Cytokines  

Nitric oxide  

Opoid peptides  

Leukocytes  

FIGURE 1. Flow chart diagram of neurogenic inflammation in the skin with an outline of the dynamic interactions between afferent nerve fibers, their principal transmitters substance P (SP), neurokinin A (NKA), and calcitonin gene-related peptide (CGRP), arterioles, venules, mast cells, and leukocytes. Dashed lines denote hypothetical interactions.

CGRP

While CGRP per se does not cause protein leakage, it is able to enhance the exudative responses to sensory nerve stimulation, tachykinins, and a variety of inflammatory mediators (Brain, 1996). This facilitation of protein extravasation is thought to result from the peptide's vasodilator activity, although inhibition of SP degradation by CGRP may also play a role (Brain, 1996; Holzer, 1992). Similar mechanisms may be responsible for the effects of VIP and PACAP to augment stimulus-induced protein leakage (Brain, 1996).

NONNEUROGENIC MEDIATORS

OF NEUROGENIC INFLAMMATION

Mast cell-derived mediators

Although there are contact sites between afferent nerve fibers and mast cells in the skin, most mast cells are located in the deeper layers of the dermis, whereas peptidergic afferent nerve fibers abound in the superficial dermis (Kowalski et al., 1990). Accordingly, the initial phase of the exudative response to sensory nerve stimulation takes place primarily in the superficial dermis of the rat and does not
involves mast cell degranulation, whereas the delayed rise in vascular permeability depends in part on mast cell-derived autacoids (Holzer, 1992; Kowalski et al., 1990). Fitting the involvement of mast cells in the postacute phase of neurogenic inflammation (Fig. 1) is the ability of SP to release various autacoids from cutaneous mast cells, whereas NKA and CGRP are rather inactive (Brain, 1996; Holzer, 1992). The stimulant effect of SP on mast cells is not due to activation of tachykinin receptors but related to a direct interaction of the cationic peptide with components of the mast cell membrane (Holzer, 1992). Since micromolar concentrations of SP are needed to activate mast cells, it would appear that mast cell-derived mediators are not essential for the vascular effect of endogenously released SP and come into play only when relatively high doses of exogenous SP are administered (Tausk and Undem, 1995).

Leukocyte-derived mediators

In addition to their action on the vascular system, neuropeptides released from afferent nerve fibers also influence the activity of granulocytes, monocytes, and lymphocytes. SP and CGRP stimulate not only the adhesion of leukocytes to the vessel wall and their emigration into the inflamed tissue of the skin (Brain, 1996; Holzer, 1992; Walsh et al., 1995) but can also affect the release of mediator substances from white blood cells (Fig. 1). SP induces monocytes to release cytokines and prostanoids, stimulates basophils to release histamine, and activates neutrophils to produce cytotoxic oxygen radicals (Brain, 1996; Holzer, 1992). Many of these leukocyte-derived mediators are vasoactive and as such are likely to exert a modulatory action on vessel diameter and vascular permeability. It is important to note in this context, however, that afferent nerve-derived peptides not only stimulate but also suppress certain components of the immune system. This is true for CGRP, which inhibits the production of interleukin-2 in lymphocytes (Wang et al., 1992) and mediates the immunosuppressive effect of ultraviolet irradiation (Gillardon et al., 1995).

ACTIVATION OF VASOACTIVE AFFERENT NEURONS

Experimental stimulation of afferent neurons has been instrumental in the elucidation of neurogenic inflammation, and it is of medical relevance to know which physiological or pathological conditions give rise to neurogenic vasodilator responses, which inflammatory reactions involve a neurogenic component, and which immune/inflammatory mediators are able to activate vasoactive afferent neurons (Fig. 2). The available evidence indicates that afferent nerve-mediated hyperaemia and plasma leakage do play a role in inflammatory disease and that there are important bidirectional interactions between the immune and nervous systems. Vasoactive afferent neurons, which can be stimulated by a variety of inflammatory and immune mediators, represent a positive feedback loop in the inflammatory process (Fig. 1).

Irritation and immune challenge

There is evidence to show that the inflammatory reaction, particularly the flare response, to bites and stings from insects, spiders, and nettles involves afferent vasodilator nerves (Brain, 1996). Inflammation caused by bacterial toxins may depend in part on the release of vasoactive peptides from sensory nerve fibers, which is also true for immediate allergic skin reactions to antigen challenge of sensitized animals and for delayed-type hypersensitivity responses (Brain, 1996; Holzer, 1992). It would hence appear that neurogenic changes

Stimulation of vasoactive afferent nerves in the skin

FIGURE 2. Summary of the stimuli that activate vasoactive afferent nerve fibers in the skin, thus leading to neurogenic vasodilatation and plasma protein extravasation.

in vascular function participate in many forms of acute inflammatory reactions to chemical or immunological challenge of the skin.

Interleukin-1β

The cytokine interleukin-1β can stimulate afferent nerve fibers or augment their excitability through mechanisms that include prostanoids and nitric oxide (Dray, 1995). The ability of interleukin-1β to sensitize nociceptive afferents has a direct bearing on the cytokine’s activity to enhance capsaicin-induced neurogenic vasodilatation in the rat skin (Figs. 2 and 3), which is most probably due to facilitation of neuropeptide release from afferent neurons (Herbert and Holzer, 1994a). Given that interleukin-1β is an important immune mediator it appears conceivable that inflammatory reactions due to immune challenge involve a neurogenic component that is initiated by the cytokine.

Mast cell-derived mediators

The mast cell mediators histamine and 5-hydroxytryptamine are able to sensitize or activate afferent neurons mostly via stimulation of histamine H1 and 5-hydroxytryptamine 5-HT2 receptors, respectively, and to give rise to afferent nerve-mediated vasodilatation and protein exudation (Dray, 1995; Holzer, 1992; Maggi, 1995a). It is important to consider, therefore, that mast cell-derived factors promote inflammation not only by a direct action on the vascular system but also by evoking neurogenic alterations in vascular function (Fig. 2). This possibility is well illustrated by the fact that the flare response to a variety of intradermal chemical stimuli involves histamine as a factor that initiates the axon reflex (Brain, 1996; Holzer, 1992).

Prostaglandins

Prostaglandins are among the most important mediators of inflammatory hyperalgesia, which seems to result from a direct action of these compounds on sensory nerve fibers (Dray, 1995). The stimulant effect of prostaglandin E2 on afferent neurons is associated with neurogenic vasodilatation in the rat skin, a reaction in which CGRP plays a major role (Holzer et al., 1995). Prostaglandins also
Nitric oxide in neurogenic vasodilatation

**FIGURE 3.** Role of nitric oxide in triggering neurogenic vasodilatation in the rat skin in response to chemical irritants, ultraviolet irradiation, nitric oxide donors, and interleukin-1β.

mediate the neurogenic vasodilator response to nitric oxide (Holzer et al., 1995) and the facilitation of neurogenic vasodilatation brought about by interleukin-1β (Herbert and Holzer, 1994a). The role of prostaglandins in facilitating neurogenic inflammatory reactions (Fig. 3) is substantiated by the observation that prostaglandin-evoked plasma protein leakage is attenuated in rats pretreated with a neurotoxic dose of capsaicin (Holzer, 1992).

**Protons and bradykinin**

The production of protons is increased in the inflamed tissue and is likely to be involved in inflammatory hyperalgesia and in the sensation of muscle discomfort after exercise-induced hypoxia (Dray, 1995). Protons are powerful stimulants of capsaicin-sensitive afferents, which results in the release of neuropeptide transmitters and in the manifestation of neurogenic vasodilatation and plasma leakage (Fig. 2). Bradykinin is another inflammatory mediator that is generated in response to tissue injury and acidification (Dray, 1995; Hall and Geppetti, 1995). The proinflammatory action of bradykinin arises from a multiplicity of effects including a direct stimulation of nociceptive afferents that bear bradykinin B2 receptors. There is pharmacological evidence to conclude that the plasma protein leakage induced by low tissue pH is due to the formation of bradykinin, which in turn stimulates the release of SP from afferent nerve fibers (Figini et al., 1995).

**Nitric oxide**

Nitric oxide (NO) synthase inhibitors have no effect on the vasodilator activity of CGRP and SP in the rat and rabbit skin but attenuate edema formation when conjoined with SP, an effect that seems to be a secondary consequence of the reduction of basal blood flow (Brain, 1996; Holzer and Jocić, 1994). It follows that the vascular effects of CGRP and SP in the rat and rabbit skin are independent of NO and that this endothelial autacoid is not a vasorelaxant mediator of neurogenic vasodilatation (Brain, 1996). There is evidence, however, to conclude that the release of neuropeptides from afferent nerve fibers can be stimulated by NO (Holzer and Jocić, 1994; Holzer et al., 1995; Hughes and Brain, 1994). The excitatory effect of NO on cutaneous vasodilator neurons (Fig. 3) has been substantiated by the ability of locally administered NO donors to cause neurogenic vasodilatation via release of CGRP from sensory nerve fibers (Holzer and Jocić, 1994). The stimulant influence of NO on afferent nerve fibers in the skin is mediated by prostaglandins (Fig. 3), and it appears as if the action of interleukin-1β to augment neurogenic vasodilatation is brought about by the same NO-prostaglandin pathway (Herbert and Holzer, 1994a, 1994b; Holzer et al., 1995).

**Heat and ultraviolet injury**

Although the afferent nerve fibers causing neurogenic inflammation are primarily chemonociceptive fibers it has also been shown that the inflammatory reactions caused by heat injury (Siney and Brain, 1996) and ultraviolet irradiation of the skin involve the release of CGRP and SP from afferent nerve fibers (Benrath et al., 1995; Siney and Brain, 1996). Circumstantial evidence suggests that NO may be a factor by which both ultraviolet irradiation (Fig. 3) and heat stimulate cutaneous vasodilator neurons, given that NO can induce neurogenic vasodilatation (Holzer and Jocić, 1994) and the cutaneous erythema due to heating and ultraviolet irradiation is attenuated by inhibition of NO synthesis (Benrath et al., 1995; Goldsmith et al., 1996).

**TERMINATION OF NEUROGENIC INFLAMMATORY REACTIONS**

Although the activation of cutaneous vasodilator neurons by inflammatory and immune mediators has been studied in some detail, comparatively less attention has been devoted to the mechanisms by which neurogenic inflammatory processes cease. The action of neuropeptides on their receptors is terminated by diffusional dilution, enzymatic breakdown (Nadel, 1996) or internalization of the peptide-receptor complexes, which in the case of SP results in desensitization of the cell to the peptide (Bowden et al., 1994). In contrast, the vasodilator action of CGRP in the skin does not undergo desensitization, and intradermal injection of minute amounts of the peptide can cause an erythema that lasts for several hours (Brain, 1996). Which mechanisms eventually stop the action of CGRP in the skin has not yet been investigated in detail but it is worth mentioning...
Neurogenic Vasodilatation and Plasma Leakage in the Skin

Inhibition of local vascular control by cutaneous afferent nerves

Prejunctional inhibition

- CGRP<sub>1</sub> autoreceptors
- Histamine H<sub>3</sub> receptors
- 5-Hydroxytryptamine 5-HT<sub>B</sub> receptors
- Opiate receptors
- α<sub>2</sub>-Adrenoceptors
- Somatostatin receptors

Postjunctional inhibition

- Peptidases
- Receptor internalization
- CGRP<sub>1</sub> receptor antagonists
- NK<sub>1</sub> receptor antagonists
- Galanin
- Corticotropin-releasing factor
- Steroids

FIGURE 4. Summary of the factors that inhibit the activity of cutaneous afferent nerve fibers to elicit vasodilatation and plasma protein extravasation.

here that proteases released by SP from mast cells can cut short the vasodilator activity of CGRP (Brain, 1996).

It is of pathophysiological relevance to know how neurogenic vasodilatation and plasma leakage are terminated because failure of the termination mechanisms may exacerbate inflammatory processes. There is indeed evidence to indicate that the activity of afferent nerve fibers to cause vasodilatation and plasma protein leakage is under the influence of various inhibitory control mechanisms (Fig. 4). Exploitation and enforcement of these negative feedback mechanisms may be an important aspect for the development and optimization of antiinflammatory therapy.

Prejunctional control mechanisms

CGRP-containing vasodilator fibers in the rat mesenteric arteries bear CGRP autoreceptors which regulate transmitter release via a negative feedback mechanism (Nuki et al., 1994) but it is not known whether a similar control mechanism operates in the skin. There is evidence, however, that a number of peptides present in afferent nerve fibers such as somatostatin, leucine-enkephalin and dynorphin, which per se do not influence blood flow and vascular permeability, can modulate the vascular effects of afferent nerve stimulation in an autoinhibitory manner (Fig. 4). Somatostatin reduces the release of SP from afferent neurons and attenuates the vasodilatation and plasma protein leakage evoked by antidromic nerve stimulation (Holzer, 1992). Opiate receptor agonists inhibit the vasodilator and exudative responses to antidromic nerve stimulation through a similar prejunctional mechanism of action (Donnerer and Amann, 1993). It is important to notice that in chronic inflammatory processes that stimulate afferent neurons to cause vasodilatation and plasma protein leakage (Geppetti and Holzer, 1996) and the use of capsaicin to defunctionalize vasoactive afferent nerves (Holzer, 1991).

Postjunctional control mechanisms

Neurogenic inflammation is most efficiently suppressed when the actions of the vasoactive peptide mediators are blocked by specific receptor antagonists (Donnerer and Amann, 1993; Geppetti and Holzer, 1996). Consequently, CGRP and tachykinin receptor antagonists are potential antiinflammatory drugs, a hypothesis that has led to an unrivaled surge of nonpeptide tachykinin receptor antagonists (Maggi, 1995b). Other factors with a postjunctional site of action include corticotropin-releasing factor and galanin, which may be expressed in afferent neurons and which attenuate plasma protein leakage evoked by both antidromic nerve stimulation and exogenous SP (Donnerer and Amann, 1993; Holzer, 1992). Antiinflamm-
matory steroids such as dexamethasone may also inhibit neurogenic inflammatory processes in the skin (Brain, 1996).

SUMMARY AND PATHOPHYSIOLOGICAL IMPLICATIONS

The involvement of peptidergic afferent neurons in vascular effector control needs to be seen in context with their role as nociceptive neurons. The overall function of these neurons is to maintain homeostasis in the face of irritation or trauma to the tissue, a role that is greatly aided by appropriate changes in the microcirculation at the very site of challenge. Hyperemia and increased vascular permeability facilitate the delivery of macromolecules and leukocytes to the tissue and thereby promote defense and repair of injury. Also potentially relevant for homeostasis is the spread of cutaneous flare beyond the site at which the skin is irritated. This propagation of arteriolar dilatation may be considered as a measure to ensure that protective hyperemia takes place not only in the challenged tissue but also in a "safety margin" (Holzer, 1992).

In a pathophysiological perspective it is important to differentiate between the role of vasoactive afferent neurons in acute inflammation and that in the chronically inflamed tissue. Under conditions of acute tissue irritation or injury, the proinflammatory function of these neurons is likely to contribute to the maintenance of tissue integrity, while dysfunction will have a deleterious bearing on skin homeostasis. The impairment of afferent nerve-mediated vasodilatation and axon reflex flare, which is seen in the skin of patients suffering from congenital sensory neuropathy or sensory neuropathies associated with diabetes, herpes zoster, postherpetic neuralgia, or atopic dermatitis (Brain, 1996; Holzer, 1992), is most probably a factor that contributes to the manifestation of the disease. The beneficial role of vasoactive afferent neurons in skin homeostasis is further illustrated by the observations that ablation of capsaicin-sensitive afferent neurons inhibits hair growth, reduces the survival of a musculoskeletal flap, aggravates acid-induced skin lesions, causes the formation of keratitis-like lesions in the cornea, leads to the appearance of persistent skin wounds and delays experimental wound healing (Brain, 1996; Holzer, 1992).

In contrast, chronic inflammation may induce vasoactive afferent neurons to be overactive out of balance and thus to enforce the debilitating and destructive consequences of the disease. Instrumental in this respect is the ability of inflammatory and immune mediators to sensitize and stimulate afferent nerve fibers and thus to provide a continuous drive for these neurons to release their proinflammatory peptide mediators (Dray, 1995; Holzer, 1992). Nerve growth factor whose formation is increased in the inflamed tissue is thought to play a central role in the long-term excitability changes of afferent neurons in chronic inflammation (Brain, 1996; Dray, 1995). An additional aspect is lift up by the ability of SP and other afferent nerve-derived neuropeptides to boost the immune system. It is hence conceivable that sensitized afferent neurons enhance host defense reactions by a direct stimulant action on immunocompetent cells and by way of hyperemia and increased vascular permeability, which facilitate the delivery and accumulation of immune cells and macromolecules in the inflamed tissue. This idea is supported by the ability of afferent nerve-derived peptides to enhance allergic contact dermatitis in mice (Brain, 1996; Holzer, 1992). Furthermore, vasoactive afferent neurons participate in the vascular reactions of the skin to allergen challenge and in the vascular manifestations of acquired cold and heat urticaria. Hyperreactive disorders of the skin such as psoriasis, bullous pemphigoid, eczema, and photodermatoses may also involve peptidergic afferent neurons (Geppetti and Holzer, 1996), and it is here that pharmacological interventions to depress the stimulation or activity of vasoactive afferent neurons may be of considerable therapeutic benefit.

References


I am very grateful to Dr. Ulrike Holzer-Petsche for preparing the computer-generated graphs. Research done in the author’s laboratory was supported by the Austrian Scientific Research Foundation (grants 9473 and 11654) and the Jubilee Foundation of the Austrian National Bank (grant 49025).


