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REVIEW ARTICLE

The Role of Substance P in Inflammatory Disease

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The diffuse neuroendocrine system consists of specialised endocrine cells and peptidergic nerves and is present in all organs of the body. Substance P (SP) is secreted by nerves and inflammatory cells such as macrophages, eosinophils, lymphocytes, and dendritic cells and acts by binding to the neurokinin-1 receptor (NK-1R). SP has proinflammatory effects in immune and epithelial cells and participates in inflammatory diseases of the respiratory, gastrointestinal, and musculoskeletal systems. Many substances induce neuropeptide release from sensory nerves in the lung, including allergen, histamine, prostaglandins, and leukotrienes. Patients with asthma are hyperresponsive to SP and NK-1R expression is increased in their bronchi. Neurogenic inflammation also participates in virus-associated respiratory infection, non-productive cough, allergic rhinitis, and sarcoidosis. SP regulates smooth muscle contractility, epithelial ion transport, vascular permeability, and immune function in the gastrointestinal tract. Elevated levels of SP and upregulated NK-1R expression have been reported in the rectum and colon of patients with inflammatory bowel disease (IBD), and correlate with disease activity. Increased levels of SP are found in the synovial fluid and serum of patients with rheumatoid arthritis (RA) and NK-1R mRNA is upregulated in RA synoviocytes. Glucocorticoids may attenuate neurogenic inflammation by decreasing NK-1R expression in epithelial and inflammatory cells and increasing production of neutral endopeptidase (NEP), an enzyme that degrades SP. Preventing the proinflammatory effects of SP using tachykinin receptor antagonists may have therapeutic potential in inflammatory diseases such as asthma, sarcoidosis, chronic bronchitis, IBD, and RA. In this paper, we review the role that SP plays in inflammatory disease. J. Cell. Physiol. 201: 167–180, 2004. © 2004 Wiley-Liss, Inc.

The neuro-immune axis is a bidirectional pathway of intersystem communication. Immune responses alter neural function, and in turn, neural activity modifies immunologic function. Inter-system cross-talk is mediated via a common biochemical language of shared ligands such as cytokines and neuropeptides, and their receptors. Mediators classically thought to be synthesized exclusively by the nervous system, are now known to be produced by immunocytes, and vice versa. The diffuse neuroendocrine system consists of specialised endocrine cells and peptidergic nerves and is present in all organs of the body, including the respiratory tract.

Tachykinins are a family of neuropeptides that share the carboxy-terminal sequence Phe-X-Gly-Leu-Met-NH_2, where X is an aromatic (Tyr or Phe) or hydrophobic (Val or Ile) amino acid (Uddman et al., 1997). This common sequence is essential for the tachykinin's receptor interaction and activation, whereas the distinct amino-terminal sequences of the tachykinins provide their receptor subtype specificity. Five tachykinin peptides have been identified in mammals: substance P (SP), neurokinin A, neuropeptide K, neuropeptide-γ, and neurokinin B.

In mammals, two separate genes encode the tachykinins designated preprotachykinin I (PPT-I) and preprotachykinin II (PPT-II) (Table 1) (Nawa et al., 1984). The *PPT-I* gene can express four distinct forms of mRNA through alternative splicing, two of which (the β and γ forms) encode synthesis of both SP and NKA, whilst the other two, the α and δ forms, encode SP only (Nawa et al., 1984). The β and γ forms of PPT-I mRNA also encode the synthesis of neuropeptide K and neuropeptide-γ, which are amino-terminally extended forms of NKA, although their function has not been fully clarified (Tatemoto et al., 1985; Kage et al., 1988). The *PPT-II* gene gives rise to neurokinin B (Hokfelt et al., 2001).

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TABLE 1. Genes encoding synthesis of mammalian tachykinins

Preprotachykinin I gene	Preprotachykinin II gene
α-*PPT-I* mRNA	*PPT-II* mRNA
Substance P (SP)	Neurokinin B
β-*PPT-I* mRNA	
SP, neurokinin A, neuropeptide K	
γ-*PPT-I* mRNA	
SP, neurokinin A, neuropeptide-γ	
δ-*PPT-I* mRNA	
SP	

SUBSTANCE P

SP was discovered by von Euler and Gaddum (1931). They reported that extracts of equine brain and intestine contained a hypotensive and spasmogenic factor. The preparation, termed preparation P, was later found to be proteinaceous. The isolation from bovine hypothalamus and characterization of SP was carried out by Leeman's group in 1970–1971 (Chang et al., 1971). The structure of SP is as follows.

SP : Arg-Pro-Lys-Pro-Gln-Gln-Phe-Phe
-Gly-Leu-Met-NH_2

SP is synthesized in the ribosome as a larger protein and then enzymatically converted into the active undecapeptide. The peptide is widely distributed in the central and peripheral nervous systems of vertebrates. In the central nervous system, SP is thought to participate in various behavioral responses and in regulating neuronal survival and degeneration. SP also regulates cardiovascular and respiratory function and is involved in activating the emetic reflex. In the spinal cord, SP participates in neurotransmission of pain and noxious stimuli and modulates autonomic reflexes, including the micturition reflex. In the peripheral system, SP is localized in the primary sensory neurons and neurons intrinsic to the gastrointestinal, respiratory tracts, and genitourinary tracts (Maggi, 2000).

Tachykinin receptors

Tachykinin effects on target cells are mediated by at least three specific receptors, the neurokinin-1 receptor (NK-1R), NK-2R, and NK-3R. These receptors are members of the superfamily of guanine nucleotide binding-coupled receptors, which interact with G-proteins to promote high-affinity binding and signal transduction (Hershey and Krause, 1990). These receptors are glycoproteins with seven putative α-helical transmembrane segments, an extracellular amino-terminus and an intracellular carboxyl tail (Fig. 1). G-protein-linked receptors are generally associated with low abundance mRNA. For example, only 15 transcripts per cell have been reported for the β2-adrenergic receptor (Hadcock et al., 1989).

Each tachykinin appears to preferentially activate a distinct tachykinin receptor, although at high ligand concentration, each tachykinin can activate each of the tachykinin receptors. The NK-1R is activated preferentially by SP, the NK-2R by NKA, and the NK-3R by NKB (Nakanishi, 1991). The conserved carboxyl terminal domain of tachykinins interacts with the neurokinin receptors, while the unique amino terminal sequences of tachykinins dictate receptor specificity. The relative affinity of NK-1R for neurokinin A and neurokinin B is 100- and 500-fold lower than for SP, respectively (Gerard et al., 1991). Recently, a fourth tachykinin receptor has been cloned (Donaldson et al., 2001).

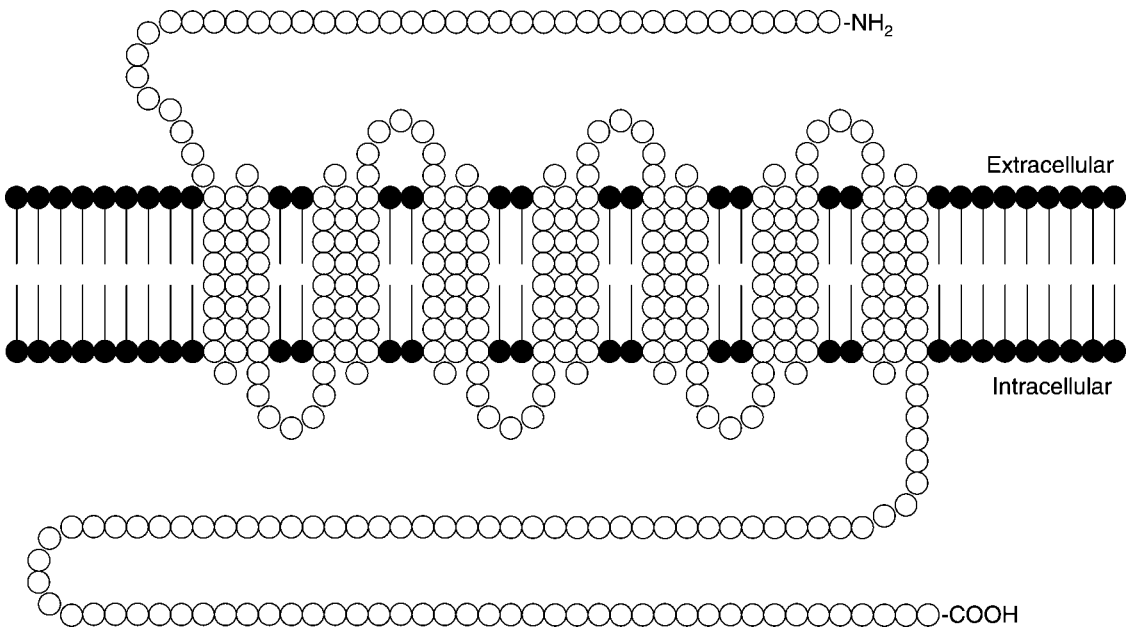

Fig. 1. The neurokinin-1 receptor (NK-1R) is a glycoprotein with seven putative α-helical transmembrane segments, an extracellular amino-terminus and an intracellular carboxyl tail. The human NK-1R consists of 407 amino acid residues and has a relative molecular mass of 46 kDa. The second and third membrane-spanning domains are involved in agonist/antagonist binding, the third cytoplasmic loop is responsible for G-protein interaction, while the cytoplasmic carboxyl terminal contains many serine and threonine residues which when phosphorylated, cause desensitization of the receptor in response to repeated application of agonist.

The human NK-1R consists of 407 amino acid residues and has a relative molecular mass of 46 kDa (Hopkins et al., 1991). The second and third membrane-spanning domains are involved in agonist/antagonist binding, the third cytoplasmic loop is responsible for G-protein interaction, while the cytoplasmic carboxyl terminal contains many serine and threonine residues which when phosphorylated, cause desensitization of the receptor in response to repeated application of agonist. Binding of SP to NK-1R mediates rapid endocytosis and internalization of the receptor; this also contributes to desensitization of cells to SP signaling. Agonist stimulation of the NK-1R in many tissue and cell types causes activation of phospholipase C, which catalyzes the hydrolysis of phosphoinositides into inositol 1,4,5-trisphosphate and diacylglycerol. These second messengers are then available for the mobilization of calcium from internal reticular stores, and for the activation of protein kinase C. In Chinese hamster ovary cells, rat NK-1R has been shown to activate both phospholipase C and adenylate cyclase, and thus to stimulate both phosphoinositide metabolism and cAMP formation (Li et al., 1997).

The gene for the human NK-1R is located on chromosome 2, spans 45–60 kb and is contained in five exons, with introns interrupting at sites homologous to those in the *NK-2R* gene (Gerard et al., 1991). The 5′ flanking region of the *NK-1R* gene has several putative transcriptional regulatory DNA elements, such as a cAMP responsive element, an AP-1, AP-2, AP-4, NF-κB, OCT-2, and a Sp-1 site. Comparison of rat and human NK-1R sequences reveals 94.5% homology (Takeda et al., 1991).

THE ROLE OF SP IN INFLAMMATION

Evidence to support the involvement of SP in the pathophysiology of inflammatory disease stems from observations of aberrant levels of SP and of SP-nerve fibres in diseased tissue, aberrant expression of NK-1R in diseased tissue, and beneficial effect of NK-1R antagonists and NK-1R knockout in animal models of inflammatory disease. Tachykinins are biologically active at extremely low concentrations (Kraneveld and Nijkamp, 2001).

Although SP has been described as a peptide of neuronal origin, studies in rodents have demonstrated its production by inflammatory cells such as macrophages, eosinophils, lymphocytes, and dendritic cells (Weinstock et al., 1988; Bost et al., 1992; Killingsworth et al., 1997; Joos et al., 2000). SP enhances lymphocyte proliferation and immunoglobulin production, and enhances cytokine secretion from lymphocytes, monocytes, macrophages, and mast cells (Stanisz et al., 1986; Lotz et al., 1988; Scicchitano et al., 1988; Pascual et al., 1991; Bost and Pascual, 1992; Calvo et al., 1992; Ansel et al., 1993; Covas et al., 1994; Ho et al., 1996; Maggi, 1997). SP-induced release of inflammatory mediators such as cytokines, oxygen radicals, arachidonic acid derivatives, and histamine potentiates tissue injury, and stimulates further leukocyte recruitment, thereby amplifying the inflammatory response (Holzer and Holzer-Petsche, 1997).

SP elicits local vasodilatation and alters vascular permeability, thus enhancing the delivery and accumulation of leukocytes to tissues for the expression of local immune responses (Pernow, 1983). SP can specifically stimulate the chemotaxis of lymphocytes, monocytes, neutrophils, and fibroblasts (Haines et al., 1993; Kahler et al., 1993; Schratzberger et al., 1997). SP has been reported to induce the expression of endothelial-leukocyte adhesion molecule-1 on human microvascular endothelium, to increase the expression of the leukocyte integrin CD11b on human neutrophils, and to enhance the expression of intercellular adhesion molecular-1 and leucocyte function-associated antigen-1 on murine endothelial cells, and lymphocytes (Matis et al., 1990; DeRose et al., 1994; Vishwanath and Mukherjee, 1996). SP induces a rapid influx of neutrophils and eosinophils in human dermis, occurring in parallel with translocation of P-selectin and upregulation of E-selectin (Smith et al., 1993). By promoting vasodilatation, leukocyte chemotaxis, and leukocyte/endothelial cell adhesion, SP ensures the extravasation, migration, and subsequent accumulation of leukocytes at sites of injury.

SP has also been implicated in the resolution of inflammation. Evidence of a role for SP in tissue repair has been primarily derived from studies detailing its proliferative effect on a variety of cells. SP acts as a mitogen for smooth muscle cells, fibroblasts, endothelial cells, and synoviocytes (Nilsson et al., 1985; Lotz et al., 1987; Ziche et al., 1990; Rameshwar et al., 1997). Indeed, a role for SP in angiogenesis has been proposed (Fan et al., 1993). SP has been implicated in inflammatory reactions of such diverse tissues as the lung (Adcock et al., 1993a; Bozic et al., 1996; Colten and Krause, 1997), the gut (Mantyh et al., 1988, 1994), the joints (Levine et al., 1984; Krause et al., 1995), the skin (Foreman, 1987), and the eyes (Bill et al., 1979). The proinflammatory effects of SP on various immune cells are summarized in Table 2.

SP and immunoregulation: lymphocytes

A diverse spectrum of immunoregulatory effects on lymphocyte function has been described for SP (Payan and Goetzl, 1985). SP is a chemoattractant for human lymphocytes and increases lymphocyte traffic and lymph flow through the peripheral lymph nodes of sheep (Moore et al., 1989; Schratzberger et al., 1997). SP acts as a B lymphocyte differentiation cofactor and increases immunoglobulin secretion (particularly IgA) by murine Peyer's patches, splenic lymphocytes, and mesenteric lymph nodes in an isotype-specific manner (Stanisz et al., 1986; Scicchitano et al., 1988; Bost and Pascual, 1992). In experiments using established B cell lymphoma clones, SP directly stimulated IgA, but not IgM secretion. However, in the presence of LPS, SP stimulated a threefold increase in IgM secretion (Pascual et al., 1991). Depletion of SP in rodents by capsaicin administration or treatment with the SP antagonist spantide, reduced the number of antibody secreting cells (Eglezos et al., 1990). SP can also enhance immunoglobulin secretion by human B-cells and IL-2 production by human T cells and murine T cell lines (Bost and Pascual, 1992; Calvo et al., 1992). SP enhances macrophage inflammatory protein-1β expression and natural killer activity in T lymphocytes (Croitoru et al., 1990; Guo et al., 2002). SP can stimulate the proliferation of human T lymphocytes at concentrations as low as

TABLE 2. Proinflammatory effects of SP in immune cells

Cell type	Effects
Lymphocytes	Potent chemoattractant
	B lymphocyte differentiation cofactor
	Increases immunoglobulin secretion
	Stimulates T lymphocyte proinflammatory cytokine release
	Stimulates T lymphocyte natural killer activity
	Stimulates T lymphocyte proliferation
	Lymphocytes can produce SP
Monocytes/macrophages	Potent chemoattractant
	Stimulates proinflammatory cytokine release
	Induces oxidative burst
	Stimulates synthesis and release of arachidonic acid metabolites
	Macrophages express NK-1R and secrete SP
Neutrophils	Chemoattractant
	Stimulates degranulation and respiratory burst
	Increases adherence to epithelial cells (upregulates adhesion molecule expression)
	Stimulates proinflammatory cytokine release
Mast cells	Close contact between mast cells and nerves
	Induces degranulation
	Induces histamine and serotonin release
	Stimulates proinflammatory cytokine release
Eosinophils	Chemoattractant
	Stimulate activation, degranulation, release of O_2^-, and thromboxane
	Eosinophils can secrete SP

10^{-10} M (Covas et al., 1994). Inhibitory effects of SP on lymphocyte proliferation have also been documented (Krco et al., 1986).

SP and immunoregulation: monocytes/macrophages

SP is a chemoattractant for human monocytes (Schratzberger et al., 1997). The chemotactic activity of SP resides in its C-terminal amino acid sequence. SP can also stimulate the secretion of cytokines such as IL-1, TNF-α, and IL-6 from monocytes and macrophages (Bill et al., 1979; Lotz et al., 1988; Ho et al., 1996). SP induces oxidative burst, and stimulates the synthesis and release of arachidonic acid metabolites, prostaglandin E_2, thromboxane B_2, and toxic oxygen radicals in guinea-pig peritoneal macrophages (Murris-Espin et al., 1995). SP enhances the phagocytosis of murine macrophages via its N-terminus (Bar-Shavit et al., 1980). Recently, the expression of SP and the NK-1R by human peripheral monocytes and sputum macrophages has been shown, suggesting that macrophages may be a major source of SP in inflammatory airway diseases (Ho et al., 1997; Germonpre et al., 1999). The expression of SP and NK-1R by monocytes/macrophages is upregulated by endotoxin (Bost et al., 1992; Germonpre et al., 1999).

SP and immunoregulation: neutrophils

SP induces the chemotaxis and degranulation of human neutrophils and stimulates respiratory burst, H_2O_2 production, and secretion of the constituents of granules (Serra et al., 1988; Haines et al., 1993). SP potently affects the migratory and cytotoxic functions of human leukocytes, suggesting that neurogenic stimuli may prepare neutrophils for an exaggerated inflammatory response to other mediators (Perianin et al., 1989). SP increases neutrophil adherence to lung epithelial cells and induces IL-1β and TNF-α release (DeRose et al., 1994; Kuo et al., 2000). The adherence-promoting activity of SP resides in its C-terminus, an effect that is NK-1R-mediated (Kroegel et al., 1990).

SP and immunoregulation: mast cells

Direct evidence for a close contact between mast cells and nerves has been obtained. A large proportion of rat intestinal mucosal cells are in direct contact with nerves, some of which contain SP or CGRP (Stead et al., 1987). Such an association has also been found in rat lung, human intestine, and skin (Stead et al., 1989; Nilsson et al., 1990; Naukkarinen et al., 1993). SP induces degranulation and histamine and serotonin release from human and rat mast cells by a receptor-independent mechanism (Shanahan et al., 1985; Repke and Bienert, 1987). SP-triggering of mast cells involves insertion of the amphiphilic SP molecule into the cell membrane, thus enabling direct activation of G proteins (Mousli et al., 1990). Stimulation of murine mast cells with SP activates TNF-α gene expression and induces TNF-α secretion (Ansel et al., 1993).

SP and immunoregulation: eosinophils

Eosinophil activation by SP is reported to cause their degranulation, release of O_2^-, and thromboxane B_2. The stimulatory effect of SP on the degranulation of guinea pig eosinophils is mediated via its N-terminus, and is thought to be receptor-independent (Kroegel et al., 1990). Eosinophils from allergic and normal subjects differ in their chemotactic response to SP. SP alone was not chemotactic for eosinophils, whereas the chemotactic response to platelet-activating factor and leukotriene B4 of eosinophils derived from asthmatic but not normal subjects was enhanced by pre-treatment with low concentrations of SP (Numao and Agrawal, 1992).

SP and immunoregulation: lung epithelium

There is an extensive apical and basal plexus of nerves in the bronchial epithelium, most of which contain SP. Recent studies have demonstrated the existence of a

continuous pathway of sensory nerves containing SP from the epithelium to arterioles in bronchial mucosa, providing structural support for a local axon reflex (Lamb and Sparrow, 2002). Neuropeptides and capsaicin stimulate the release of inflammatory cytokines in a human bronchial epithelial cell line. Exposure to SP resulted in immediate increases in intracellular calcium, followed by the synthesis of the transcripts for and the release of the proteins of the inflammatory cytokines IL-6, IL-8, and TNF-α (Veronesi et al., 1999). Neuropeptides may also stimulate proliferation of human airway epithelial cells, suggesting a contrasting role of repair after epithelial injury (Kim et al., 1995).

SP and immunoregulation: hematopoiesis

SP-immunoreactive nerve fibers have been detected in bone marrow (Weihe et al., 1991). Electron microscopic studies have demonstrated direct synapse-like contacts between nerve endings and the cytoplasmic processes of reticular and fibroblastoid cells of bone marrow. SP stimulates bone marrow progenitors of both erythroid and myeloid lineages, and has, therefore, been implicated in hematopoiesis (Rameshwar et al., 1993). The hematopoietic effect of SP is primarily due to its stimulatory influence on bone marrow stroma (macrophages, reticular adventitial cells, adipocytes, endothelial cells, and fibroblasts). SP induces production of essential hematopoietic growth factors such as IL-1 and stem cell factor by murine bone marrow stroma, and these cytokines in turn regulate stromal expression of NK-1R (Rameshwar and Gascon, 1995). SP also induces IL-3 and granulocyte-macrophage colony stimulating factor by human bone marrow mononuclear cells, and the expression of NK-1R mRNA in human bone marrow fibroblasts, which in turn, proliferate in response to SP stimulation (Rameshwar et al., 1997).

SP and apoptosis

SP has been shown to have antiapoptotic effects in many cells, including macrophages, neutrophils, and thymocytes (DeFea et al., 2000; Dimri et al., 2000; Bockmann et al., 2001; Kang et al., 2001). Glucocorticoids and SP appear to have opposing effects in thymocytes, with glucocorticoids acting as strong inducers of apoptosis and SP counteracting this effect (Dimri et al., 2000). Lung epithelial apoptosis contributes to the pathophysiology of asthma, idiopathic pulmonary fibrosis (IPF), and acute lung injury (Kuwano et al., 1999; Matute-Bello et al., 1999; Trautmann et al., 2002). The antiapoptotic effect of SP may protect against epithelial cell injury in these diseases. Tachykinins may stimulate growth in small cell lung cancer, and blocking the effects of SP increases apoptosis in cancer cells (Bepler et al., 1988).

In contrast, SP may promote apoptosis in other biological systems, either directly or indirectly through its ability to induce secretion of proinflammatory cytokines which may promote apoptosis. SP potentiates and NK-1R antagonists protect mice from CD95 and TNF-α-mediated apoptotic liver damage (Bang et al., 2003). SP can also induce a non-apoptotic form of programmed cell death that is independent of caspase activation (Castro-Obregon et al., 2002). Therefore, the effects of SP on cell death and survival are tissue specific and may also depend on the inflammatory microenvironment of the tissue.

SP IN THE LUNG

In the human respiratory tract, SP-immunoreactive nerves are located beneath and within the epithelium, around submucosal bronchial glands, bronchial blood vessels, and to a lesser extent within airway smooth muscle (Helke et al., 1990; Barnes et al., 1991; Solway and Leff, 1991). Significant amounts of SP are found in central and peripheral airway tissues, as well as bronchoalveolar lavage (BAL) fluid and sputum (Nieber et al., 1992; Lilly et al., 1995a; Tomaki et al., 1995). The content of SP in human lung may decrease with age and after denervation (Hislop et al., 1990).

Neural regulation of the airways consists of cholinergic excitatory, adrenergic inhibitory nerves, and nonadrenergic, non-cholinergic (NANC) nerves. Cholinergic nerves form the predominant bronchoconstrictor neural pathway in human airways. Acetylcholine controls neuronal and non-neuronal target cells via a short-lived action at nicotinic and muscarinic receptors. NANC nerves can be either inhibitory or excitatory. Noncholinergic excitatory nerves generate antidromic pulses and a local axon reflex, which leads to noncholinergic bronchoconstriction, plasma extravasation, and vasodilatation (Joos et al., 1987b). SP and neurokinin A are thought to mediate the excitatory part of the NANC nervous system. NK-2Rs are present on smooth muscle of both large and small airways and mediate part of the bronchoconstrictor effect of tachykinins. Most of the proinflammatory effects of SP are mediated by the NK-1R. An extensive cross-talk exists between nerves and the immune system. The complexity of the picture has increased further as it has become clear that classical neurotransmitters, such as acetylcholine and neuropeptides, are produced by non-neuronal cells (Joos, 2001).

Tachykinins released from sensory C-fibres induce neurogenic inflammation, characterized by vasodilatation, increased postcapillary venule permeability, and neutrophil adherence to blood vessels (Umeno et al., 1990; Piedimonte et al., 1993). Neuropeptides can be released from sensory nerves by a range of substances, including allergen, ozone, or bronchoactive agonists such as histamine, prostaglandins, and leukotrienes (Kaufman et al., 1980; Martins et al., 1991b; Takebayashi et al., 1998). Tachykinin-mediated inflammatory responses may be enhanced in endotoxaemia and implicated in endotoxin-induced lung injury. LPS enhances SP-mediated neutrophil accumulation in the lungs and vascular permeability in guinea pig airways (Kuo et al., 1998). Tachykinins, especially SP through the NK-1 receptor, induce a series of leukocyte responses to trigger and amplify the inflammatory processes, including upregulation of ICAM-1 expression on vascular endothelial cells and enhancement of neutrophil transendothelial migration, mediating leukocyte adhesion to the endothelial or epithelial cells in the airways (DeRose et al., 1994; Baluk et al., 1995; Nakagawa et al., 1995; Bhatia et al., 1998). SP also stimulates human monocytes to release IL-1, IL-6, and TNF (Lotz et al., 1988).

Animal studies using NK-1R antagonists and *NK-1R* gene knockout mice further support the theory that the NK-1R may be involved in the pathogenesis of airway inflammation (Bozic et al., 1996; Kaltreider et al., 1997). Increased expression of NK-1R mRNA by alveolar macrophages was observed in a murine model of antigen-induced airway inflammation (Kaltreider et al., 1997).

Neurokinin receptors in the lung

NK-1Rs and NK-2Rs are present in several structures of human central airways, including smooth muscle, glands, vessels, and pulmonary arteries. NK-1R and NK-2R mRNA are found in equal abundance in bronchi and subpleural lung, suggesting multiple sites of action of neuropeptides (Solway and Leff, 1991). Different tachykinin receptors are involved in the direct and indirect bronchoconstrictor effect of tachykinins in the rat (Joos et al., 1994). In other words, tachykinins may cause bronchoconstriction directly at high concentrations, mediated by direct inteaction with NK-1Rs, and indirectly through mast cells and cholinergic nerves.

Tachykinin-induced pulmonary stretch receptor activation and airway effects of capsaicin are mediated by the activation of NK-2Rs (Matsumoto et al., 1997; Vieira et al., 1997). Stimulation of NK-1 receptors causes relaxation of human pulmonary arteries which is mediated largely by nitric oxide and prostacyclin released from the endothelium (Corboz et al., 1998).

Neutral endopeptidase

Tachykinins are degraded by the enzymes neutral endopeptidase (NEP) and angiotensin converting enzyme (ACE) (Joos et al., 2000). NEP is more important than ACE in airway neuropeptide metabolism (Martins et al., 1991a). NEP maintains low levels of SP in the extra-cellular fluid under basal conditions and terminates its proinflammatory effects. NEP has been localized in the lungs of different animal species (Nadel and Borson, 1991). NEP is present in the basal cells of the epithelium, type II alveolar cells, neutrophils, submucosal glands, airway smooth muscle, and postcapillary venules and nerves. Inhibition of NEP enhances various airway effects of exogenously administered SP and tachykinins, including airway smooth muscle contraction, plasma extravasation, and airway mast cell activation (Nadel and Borson, 1991; Roques et al., 1993). Moreover, the airway effects of endogenous sensory neuropeptides are enhanced in the presence of a NEP inhibitor (Cheung et al., 1993). NEP and ACE participate in the metabolism of SP when administered intravascularly, whilst NEP degrades tachykinins administered by aerosol (Shore et al., 1988).

A variety of environmental irritants and sensitizers, such as toluene diisocyanate, cigarette smoke, and hypochlorous acid, and pathogens, such as the human influenza virus A/Taiwan, the Sendai virus and Mycoplasma, decrease airway NEP and increase the response of airways to SP and NKA (Borson, 1991; Nadel, 1991). Glucocorticoids can reduce the magnitude of plasma extravasation produced in the rat trachea, mediated, in part, by an upregulation in NEP synthesis (Piedimonte et al., 1990). Intestinal inflammation results in down-regulation of NEP, which may contribute to uncontrolled inflammation (Sturiale et al., 1999).

Asthma

SP-immunoreactive nerves are increased in the submucosa of patients with severe or fatal asthma and NEP in tissue may be reduced (Nadel and Borson, 1991; Ollerenshaw et al., 1991). Airway epithelial damage may stimulate sub-epithelial sensory nerves, with subsequent release of neuropeptides (Laitinen et al., 1985). Increased amounts of SP are found in sputum and BAL fluid of asthmatics and levels in BAL fluid increase after intra-segmental allergen challenge in atopics (Nieber et al., 1992; Tomaki et al., 1995). SP causes many of the typical changes observed in asthmatic airways (Fig. 2), including bronchoconstriction, increased mucus secretion, facilitation of cholinergic neurotransmission, vasodilatation, and plasma leakage (Lundberg et al., 1983; Laitinen et al., 1985; Martling et al., 1987; Helke et al., 1990; Kuo et al., 1990; Barnes et al., 1991; Mantyh, 1991; Solway and Leff, 1991). Tachykinins cause macrophage chemotaxis, mast-cell degranulation, T-cell recruitment, and B-cell immunoglobulin production in respiratory tissues (Goetzl and Sreedharan, 1992; Tiberio et al., 1997). Furthermore, SP and NKA are potent mitogens of smooth muscle cells, endothelial cells, epithelial cells, and fibroblasts and hence are potential mediators of the thickened airways found in asthma (Nilsson et al., 1985; Kuwano et al., 1993; Harrison et al., 1995; Kim et al., 1995).

NK-1Rs are predominantly localized to bronchial vessels, epithelial cells, submucosal glands, and vascular endothelium, whereas NK-2Rs are predominantly localized to airway smooth muscle (Bai et al., 1995; Strigas and Burcher, 1996). SP is more potent than NKA in stimulating airway mucus secretion, microvascular leakage, and vasodilation whereas NKA is a more potent constrictor of human bronchi than SP (Sheldrick et al., 1995; Van Rensen et al., 2002). Conflicting data exist with regard to NK-1R expression in asthma. Some studies have shown upregulated NK-1R expression in asthmatic lung compared to normal controls, whereas others have shown no differences (Solway and Leff, 1991; Adcock et al., 1993a; Chu et al., 2000).

Asthmatics are hyperresponsive to SP and NKA (Joos et al., 1987a; Cheung et al., 1994). Incubating human bronchi with serum from asthmatic patients atopic to *Dermatophagoides pteronyssimus* caused an enhanced sensitivity and contractile response to SP and NKA (Ben-Jebria et al., 1993). Normal airway smooth muscle becomes hyperresponsive to acetylcholine and tachykinins following exposure to IL-1β and TNF-α. IL-1β-enhanced cholinergic airway smooth muscle contractile responses are mediated by the actions of SP released from intrinsic airway neurons (Wu et al., 2002). Animal and human studies have suggested that tachykinins may cause bronchoconstriction both directly and indirectly, by activation of postganglionic cholinergic nerves and mast cells (Lundberg et al., 1983; Joos et al., 1988; Mantyh, 1991). SP can liberate histamine from airway and BAL fluid mast cells, but the lack of effect of specific H1-antagonists on tachykinin-induced bronchoconstriction in asthmatics suggests that histamine does not play a major role (Crimi et al., 1990; Lilly et al., 1995b).

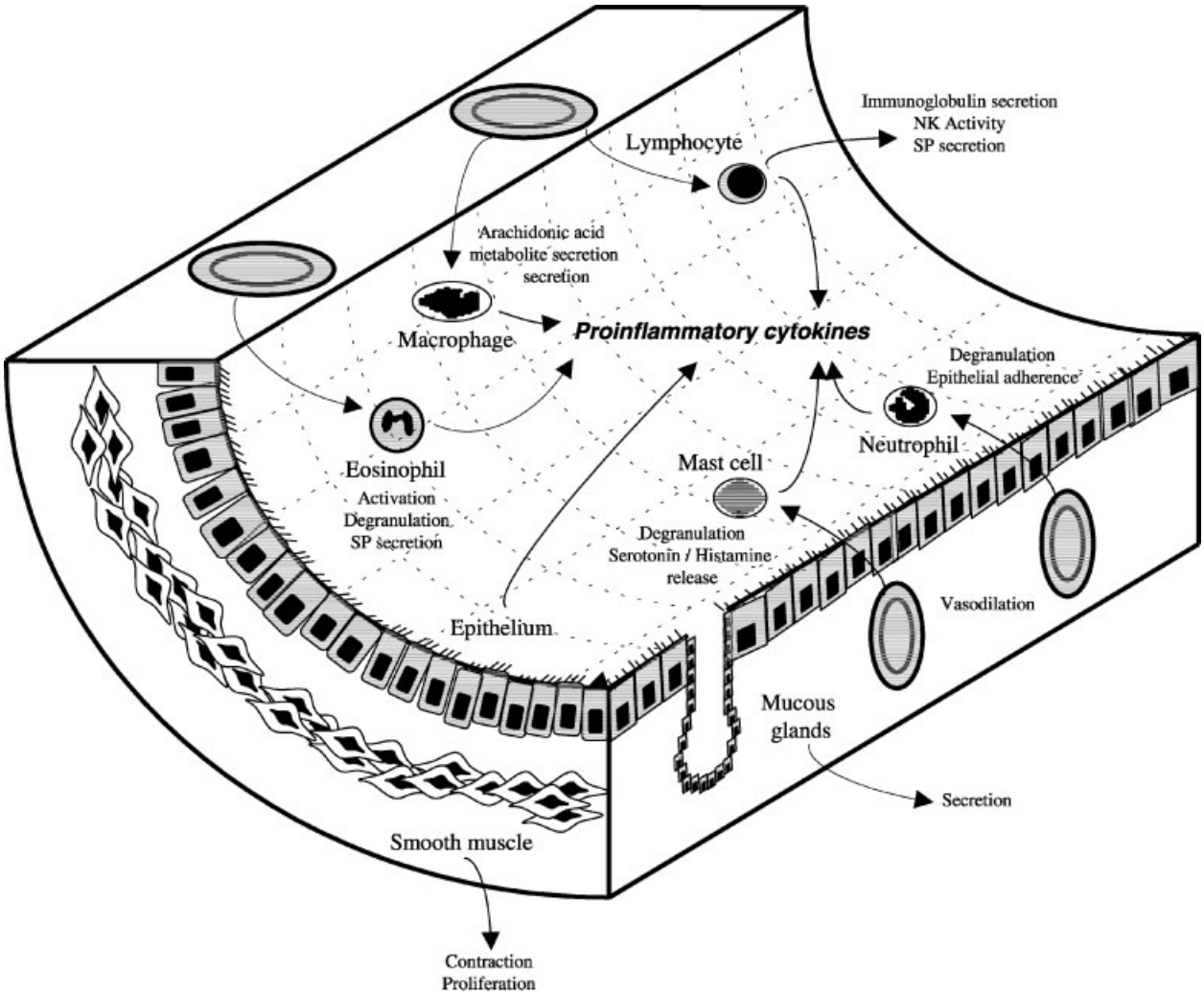

Fig. 2. Substance P (SP) leads to many of the typical features of asthmatic inflammation in the airway, including eosinophil and mast cell activation and degranulation, mucus cell hypersecretion and smooth muscle contraction. SP induces proinflammatory cytokine secretion from inflammatory and epithelial cells in the airway, which amplifies local inflammation.

Genetic factors appear to determine the magnitude of the airway response to tachykinins (Pauwels et al., 1993).

Corticosteroids may attenuate neurogenic inflammation in asthmatic lung. The *NK-1R* gene has glucocorticoid-responsive elements, indicating that steroids may modulate transcription (Ihara and Nakanishi, 1990). A comparison of the *NK-1R* gene with known promoter elements has indicated a region at −52 to −45 from the transcription start site that resembles a consensus activation protein-1-binding site (Sheng et al., 1988). Binding at these sites is stimulated by long-term mediators of inflammation, such as cytokines, and reduced by glucocorticoids (Adcock et al., 1993b). This suggests increased NK-1R gene expression due to the chronic inflammatory process that can be down-regulated by steroid therapy. Experiments in rat pancreatic acinar cells and human IM-9 lymphoblasts indicate a decrease in NK-1R mRNA after glucocorticoid treatment and NK-1R mRNA is reduced in asthmatic lung specimens after incubation with dexamethasone (Ihara and Nakanishi, 1990; Gerard et al., 1991; Adcock et al., 1993a). Recent studies have shown that inhaled steroid reduces bronchial responsiveness to tachykinins in patients with asthma (Van Schoor et al., 2002).

NK-1R antagonists may have therapeutic potential in patients with asthma. Pretreatment of antigen-sensitized guinea pigs with NK-1R antagonists prevented the development of bronchial hyperreactivity. Tachykinin antagonists prevent the bronchoconstriction and increased permeability in an animal model of exercise-induced asthma, and a combined NK1/NK2 antagonist prevents bradykinin-induced bronchoconstriction in asthmatics (Ichinose et al., 1992; Solway et al., 1993).

The role of SP in other diseases of the respiratory tract

Neurogenic inflammation participates in virus-associated respiratory infection. Respiratory syncitial virus (RSV) makes the airway susceptible to the

proinflammatory effects of SP by upregulating SP NK-1R expression on airway cells (King et al., 2001). This effect may contribute to the inflammatory reaction to the virus and could be a target for the therapy of RSV disease (Piedimonte et al., 1999). Upregulation of lymphocyte NK-1R expression may participate in the development of primary and secondary immune responses to respiratory virus infections (Tripp et al., 2002). Parainfluenza virus-induced M2 receptor dysfunction and hyperresponsiveness are prevented by a NK-1R antagonist, but not by a NK-2R antagonist, whereas both antagonists had similar anti-inflammatory effects (Jacoby et al., 2000). The acute airway inflammation observed in patients after exposure to adenoviral vectors may also exhibit a neurogenic component (Piedimonte et al., 1997).

Cigarette smoking releases SP from sensory nerves, induces adhesion of leukocytes to tracheal mucosa, decreases NEP activity, and exaggerates neurogenic inflammatory responses (Lundberg and Saria, 1983; Dusser et al., 1989; Baluk et al., 1996). Chronic smoke-induced airway hyperresponsiveness is related to an increase in SP synthesis and release in neurons innervating the lungs and airways (Kwong et al., 2001). Although some studies have shown no differences, others have shown that NK-1R and NK-2R expression are increased in the lungs of smokers (Solway and Leff, 1991; Mapp et al., 2000). Smoke-induced bronchoconstriction in guinea pigs consists of an early phase induced by both a cholinergic reflex and tachykinin release, probably evoked by the activation of bronchopulmonary C fibres, and a late phase caused by the action of arachidonic acid metabolites (Hong et al., 1995). These findings may help explain the increased incidence of airway hyperresponsiveness and cough in people exposed to tobacco smoke.

Tachykinin-containing capsaicin-sensitive nerves may play a role in the generation of non-productive cough (Karlsson, 1993). Capsaicin stimulates airway C-fibres, and is one of the most potent tussigenic stimuli known. Airway rapidly adapting afferent nerves also participate in the cough reflex. These nerves do not normally contain tachykinins, but begin to produce them in inflamed airways of allergic inflammation and viral infection (Hunter et al., 2000; Carr et al., 2002). Patients with non-asthmatic chronic cough have high levels of SP and interleukin-8 in sputum, as well as mild neutrophilia (Pizzichini et al., 1999). SP is released during allergic reactions in the nose and causes an increase in nasal microvascular leakage in patients with allergic rhinitis (Braunstein et al., 1991).

Increased levels of SP have been found in BAL fluid recovered from patients with IPF and sarcoidosis, and SP activates monocytes recovered from BAL fluid more in patients with IPF and sarcoidosis than healthy volunteers (Takeyama et al., 1996; Brunelleschi et al., 2000). We recently demonstrated upregulated NK-1R expression in BAL cells, bronchial epithelium, and granulomas of patients with sarcoidosis compared with normal controls (O'Connor et al., 2003). SP, by activating the NK-1R, stimulates the secretion of TNF-α, a critical cytokine in the pathogenesis of granulomatous inflammation in sarcoidosis, from alveolar macrophages and epithelial cells (Ho et al., 1996; Kuo et al., 2000). SP, acting through upregulated NK-1R expression, may increase proinflammatory cytokine production in the lungs of patients with sarcoidosis and thus amplify localized pulmonary inflammatory responses. In contrast to untreated patients with sarcoidosis, we were unable to detect NK-1R expression in BAL cells and endobronchial biopsies of a patient with sarcoidosis who was taking high dose corticosteroids (O'Connor et al., 2003). Furthermore, culture of alveolar and bronchial epithelial cells in dexamethasone downregulated NK-1R expression on these cells (O'Connor et al., 2003). We suggest that the downregulatory effect of corticosteroids on NK-1R expression may explain, in part, the disease modifying effect of corticosteroids in sarcoidosis.

SP IN THE GASTROINTESTINAL TRACT

The enteric nervous system of the gut is comprised of approximately 10^8 neurons, which contain a plethora of peptidergic neurotransmitters. The gastrointestinal mucosa, rich in peptidergic innervation and immunocyte content, therefore, provides the ideal milieu for neuro-immune interactions to occur. SP regulates smooth muscle contractility, epithelial ion transport, vascular permeability, and immune function in the gastrointestinal tract (Pernow, 1983; Lordal et al., 1996). In the human colon, SP nerve fibres ramify throughout the lamina propria and to form dense networks beneath the epithelium (Keast et al., 1985). SP-containing subepithelial nerves are in intimate contact with intestinal mucosal mast cells of the rat (Stead et al., 1989). However, neurons are not the exclusive source of SP. Enteroendocrine cells, human colonic eosinophils, rat ileal macrophages, and mouse colonic glia express SP (Keast et al., 1985; Bernstein and Vidrich, 1994; Metwali et al., 1994; Castagliuolo et al., 1997).

Repeated oesophageal acidification caused by relaxation of the lower oesophageal sphincter occurs in gastro-oesophageal reflux disease. Intra-oesophageal acid causes SP release from extrinsic afferent nerve endings which activates local inhibitory pathways to the lower oesophageal sphincter via NK-1Rs (Blackshaw and Dent, 1997). SP acts on NK-1R cholinergic vagal neurons in the dorsal motor nucleus of the vagus nerve, which control enteric NANC motor inhibition of the gastric fundus (Chang et al., 1999). Therefore, an antiemetic site of action of NK-1R antagonists may be in the dorsal motor nucleus to prevent excitation of neurons controlling fundic relaxation (Ladabaum and Hasler, 1999). Tachykinins stimulate duodenal contraction and bicarbonate secretion and are potent pancreatic circulatory stimulants and secretagogues (Pawlik et al., 1992). SP regulates the severity of acute pancreatitis and pancreatitis-associated lung injury (Bhatia et al., 1998; Grady et al., 2000). Localization of IL-8 positive immune cells around pancreatic nerves in chronic pancreatitis supports the existence of a neuroimmune interaction (Di Sebastiano et al., 2000).

SP and NKA are synthesized by enteric cholinergic motor neurons that project to the longitudinal and circular muscle of the intestine (Maggi, 1990). NK-1R is the primary tachykinin receptor involved in NANC transmission (Saban et al., 1999). NK-1R stimulation evokes a myogenic excitatory and a neurogenic inhibitory motor

response (Lecci et al., 1999). Interstitial cells of Cajal function as pacemakers of rhythmic activity and intermediaries in neural input from the enteric nervous system to the muscle, and express NK-1R (Epperson et al., 2000). Increased NK-1R expression is seen in human colonic mucosal mononuclear cells when compared to peripheral blood mononuclear cells, suggesting a direct role for SP in mucosal immunomodulation (Goode et al., 1998, 2000b).

Inflammatory bowel disease

The pathogenesis of IBD represents an interaction between genetic predisposing factors, exogenous, and endogenous triggers, and modifying factors, resulting in a spontaneously relapsing and remitting inflammatory process (Shanahan, 1993). Elevated levels of SP have been reported in the rectum and colon of UC patients, and correlate with disease activity (Koch et al., 1987; Mazumdar and Das, 1992; Bernstein et al., 1993). Other studies show an increase in SP nerves in hypervascular lesions, but a decrease in severe inflammatory lesions of UC colon (Kimura et al., 1994). Conflicting results also exist regarding SP in Crohn's disease (CD). Mucosal levels of SP were found to be significantly decreased in the rectum of patients with CD (Bernstein et al., 1993). Others have reported no significant difference between mucosal levels of SP from CD patients and those of controls (Koch et al., 1987). An increased density of SP-immunoreactive fibres has been demonstrated in hypervascular lesions of CD and in CD colon (Mazumdar and Das, 1992; Kimura et al., 1994). Autoradiographic studies demonstrated a 1,000-fold upregulation of SP binding sites in the lymphoid follicles and submucosal vasculature of patients with IBD (Mantyh et al., 1988, 1994). We have recently shown that proinflammatory cytokines induce NK-1R expression in colonic epithelial cells, suggesting that colonic inflammation may potentiate further SP-induced inflammatory and proliferative effects (Goode et al., 2003). Furthermore, we have shown marked upregulation of NK-1R mRNA levels in IBD colonic mucosal biopsies compared with non-inflamed mucosal expression levels (Goode et al., 2000a).

Considerable evidence has implicated SP in the pathophysiology of experimental models of IBD. Elevated levels of SP have been associated with *Trichinella spiralis*-induced enteritis (Swain et al., 1992; Agro and Stanisz, 1993). Increased SP levels were dependent on the presence of lymphocytes, as the effect was abolished in athymic rats (Swain et al., 1992). Furthermore, blockade of SP with either SP antibodies, or with the NK-1R antagonist CP 96,345, reduced jejunal inflammation (Agro and Stanisz, 1993; Kataeva et al., 1994).

SP plays an important role in *Clostridium difficile* toxin A-induced enterocolitis. Intraluminal administration of toxin A induced release of SP from primary afferent neurons, while capsaicin pre-treatment inhibited toxin A-mediated fluid secretion, neutrophil infiltration, myeloperoxidase activity, and rat mast cell protease II release in the rat ileum (Mantyh et al., 1996b; Wershil et al., 1998). Administration of specific SP antagonists inhibited toxin A-mediated TNF-α release from isolated intestinal macrophages (Pothoulakis et al., 1994; Castagliuolo et al., 1997). Mice genetically deficient in the NK-1R were protected from the secretory and inflammatory changes (Castagliuolo et al., 1998). Increased SP binding sites have been demonstrated in the lymphoid aggregates and vasculature of a patient with *C. difficile* toxin A-induced pseudomembranous colitis (Mantyh et al., 1996a). In contrast, NK-1R mRNA expression is significantly reduced in patients with HIV infection. This may contribute to the mucosal abnormality, altered intestinal motility and GI symptoms associated with HIV infection (McGowan et al., 1997).

SP AND ARTHRITIS

Substantial evidence indicates that SP contributes to the pathophysiology of joint inflammation. Studies demonstrate the involvement of neurogenic inflammation in adjuvant-induced experimental arthritis in rats (Levine et al., 1984, 1985). They found that joints which developed severe arthritis had a dense innervation of SP-containing sensory neurons, and a higher SP content, than joints that developed mild arthritis. They also reported that infusion of SP into the knee exacerbated the severity of experimental arthritis, whereas infusion of a SP antagonist had no effect. Neural depletion of SP by capsaicin administration, eliminates paw swelling and tenderness within the inflamed joint (Colpaert et al., 1983). Furthermore, elevated SP levels and accelerated cartilage degradation has been reported in rabbit knees injected with IL-1β or TNF-α (O'Byrne et al., 1990).

Increased levels of SP in the synovial fluid and serum of patients with rheumatoid arthritis (RA) have been documented (Marshall et al., 1990; Menkes et al., 1993). Others demonstrated tachykinin-immunoreactivity in the stromal nerves of normal tissues, but not of RA tissues (Gronblad et al., 1988). SP stimulates prostaglandin E_2, and collagenase release from RA synoviocytes and increases proliferation of RA synoviocytes (Lotz et al., 1987). These findings support a role for SP in the cartilage destruction, bone lesion development, and pannus formation of arthritis. NK-1R mRNA is expressed by RA synoviocytes, but not by normal synoviocytes (Krause et al., 1995).

CONCLUSIONS

The wide range of inflammatory diseases in which SP participates suggests a major future role for neurokinin receptor antagonists in the management of these diseases. Studies with neurokinin receptor antagonists suggest that blocking the binding of SP to the NK-1R interrupts the inflammatory cascade that triggers and maintains intestinal lesions of IBD (Sonea et al., 2002). Tachykinins may participate in the gastrointestinal dysmotility associated with infection, inflammation, stress, and pain. Tachykinin agonists and antagonists may become adjuncts to the treatment of motor disorders that involve pathological disturbances of the gastrointestinal tachykinin system and may ultimately have a role as spasmolytic, antidiarrheal, antiinflammatory, antiemetic, and antinociceptive drugs (Holzer and Holzer-Petsche, 1997). SP derivatives cause apoptosis in small cell lung cancer cells, and block calcium mobilization induced by neuropeptides, and thus are potential therapeutic compounds for the treatment of small cell lung cancer (Rosati et al., 1998). At present, trials with neurokinin receptor antagonists in patients

with asthma, IBD, and depression are under way. Ultimately, tachykinin receptor antagonists may have therapeutic potential in other inflammatory diseases such as sarcoidosis, chronic bronchitis and chronic cough and RA (Chung and Chang, 2002).

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LITERATURE CITED

Adcock IM, Peters M, Gelder C, Shirasaki H, Brown CR, Barnes PJ. 1993a. Increased tachykinin receptor gene expression in asthmatic lung and its modulation by steroids. J Mol Endocrinol 11(1):1–7.

Adcock IM, Peters MJ, Brown CR, Gelder CM, Barnes PJ. 1993b. Transcription factor interactions in human lung. Biochem Soc Trans 21(Pt 3):277S.

Agro A, Stanisz AM. 1993. Inhibition of murine intestinal inflammation by anti-substance P antibody. Reg Immunol 5(2):120–126.

Ansel JC, Brown JR, Payan DG, Brown MA. 1993. Substance P selectively activates TNF-alpha gene expression in murine mast cells. J Immunol 150(10):4478–4485.

Bai TR, Zhou D, Weir T, Walker B, Hegele R, Hayashi S, McKay K, Bondy GP, Fong T. 1995. Substance P (NK1)- and neurokinin A (NK2)-receptor gene expression in inflammatory airway diseases. Am J Physiol 269(3 Pt 1):L309–L317.

Baluk P, Bertrand C, Geppetti P, McDonald DM, Nadel JA. 1995. NK1 receptors mediate leukocyte adhesion in neurogenic inflammation in the rat trachea. Am J Physiol 268(2 Pt 1):L263–L269.

Baluk P, Bertrand C, Geppetti P, McDonald DM, Nadel JA. 1996. NK1 receptor antagonist CP-99,994 inhibits cigarette smoke-induced neutrophil and eosinophil adhesion in rat tracheal venules. Exp Lung Res 22(4):409–418.

Bang R, Biburger M, Neuhuber WL, Tiegs G. 2004. Neurokinin-receptor antagonists protect mice from CD95- and TNF{alpha}-mediated apoptotic liver damage. J Pharmacol Exp Ther 308:926–934.

Bar-Shavit Z, Goldman R, Stabinsky Y, Gottlieb P, Fridkin M, Teichberg VI, Blumberg S. 1980. Enhancement of phagocytosis—a newly found activity of substance P residing in its N-terminal tetrapeptide sequence. Biochem Biophys Res Commun 94(4):1445–1451.

Barnes PJ, Baraniuk JN, Belvisi MG. 1991. Neuropeptides in the respiratory tract. Part I. Am Rev Respir Dis 144(5):1187–1198.

Ben-Jebria A, Marthan R, Rossetti M, Savineau JP. 1993. Effect of passive sensitization on the mechanical activity of human isolated bronchial smooth muscle induced by substance P, neurokinin A, and VIP. Br J Pharmacol 109(1):131–136.

Bepler G, Rotsch M, Jaques G, Haeder M, Heymanns J, Hartogh G, Kiefer P, Havemann K. 1988. Peptides and growth factors in small cell lung cancer: Production, binding sites, and growth effects. J Cancer Res Clin Oncol 114(3):235–244.

Bernstein CN, Vidrich A. 1994. Isolation, identification, and culture of normal mouse colonic glia. Glia 12(2):108–116.

Bernstein CN, Robert ME, Eysselein VE. 1993. Rectal substance P concentrations are increased in ulcerative colitis but not in Crohn's disease. Am J Gastroenterol 88(6):908–913.

Bhatia M, Saluja AK, Hofbauer B, Frossard JL, Lee HS, Castagliuolo I, Wang CC, Gerard N, Pothoulakis C, Steer ML. 1998. Role of substance P and the neurokinin 1 receptor in acute pancreatitis and pancreatitis-associated lung injury. Proc Natl Acad Sci USA 95(8):4760–4765.

Bill A, Stjernschantz J, Mandahl A, Brodin E, Nilsson G. 1979. Substance P: Release on trigeminal nerve stimulation, effects in the eye. Acta Physiol Scand 106(3):371–373.

Blackshaw LA, Dent J. 1997. Lower oesophageal sphincter responses to noxious oesophageal chemical stimuli in the ferret: Involvement of tachykinin receptors. J Auton Nerv Syst 66(3):189–200.

Bockmann S, Seep J, Jonas L. 2001. Delay of neutrophil apoptosis by the neuropeptide substance P: Involvement of caspase cascade. Peptides 22(4):661–670.

Borson DB. 1991. Roles of neutral endopeptidase in airways. Am J Physiol 260(4 Pt 1):L212–L225.

Bost KL, Pascual DW. 1992. Substance P: A late-acting B lymphocyte differentiation cofactor. Am J Physiol 262(3 Pt 1):C537–C545.

Bost KL, Breeding SA, Pascual DW. 1992. Modulation of the mRNAs encoding substance P and its receptor in rat macrophages by LPS. Reg Immunol 4(2):105–112.

Bozic CR, Lu B, Hopken UE, Gerard C, Gerard NP. 1996. Neurogenic amplification of immune complex inflammation. Science 273(5282):1722–1725.

Braunstein G, Fajac I, Lacronique J, Frossard N. 1991. Clinical and inflammatory responses to exogenous tachykinins in allergic rhinitis. Am Rev Respir Dis 144(3 Pt 1):630–635.

Brunelleschi S, Nicali R, Lavagno L, Viano I, Pozzi E, Gagliardi L, Ghio P, Albera C. 2000. Tachykinin activation of human monocytes from patients with interstitial lung disease, healthy smokers, or healthy volunteers. Neuropeptides 34(1):45–50.

Calvo CF, Chavanel G, Senik A. 1992. Substance P enhances IL-2 expression in activated human T cells. J Immunol 148(11):3498–3504.

Carr MJ, Hunter DD, Jacoby DB, Undem BJ. 2002. Expression of tachykinins in nonnociceptive vagal afferent neurons during respiratory viral infection in guinea pigs. Am J Respir Crit Care Med 165(8):1071–1075.

Castagliuolo I, Keates AC, Qiu B, Kelly CP, Nikulasson S, Leeman SE, Pothoulakis C. 1997. Increased substance P responses in dorsal root ganglia and intestinal macrophages during *Clostridium difficile* toxin A enteritis in rats. Proc Natl Acad Sci USA 94(9):4788–4793.

Castagliuolo I, Riegler M, Pasha A, Nikulasson S, Lu B, Gerard C, Gerard NP, Pothoulakis C. 1998. Neurokinin-1 (NK-1) receptor is required in *Clostridium difficile*-induced enteritis. J Clin Invest 101(8):1547–1550.

Castro-Obregon S, Del Rio G, Chen SF, Swanson RA, Frankowski H, Rao RV, Stoka V, Vesce S, Nicholls DG, Bredesen DE. 2002. A ligand-receptor pair that triggers a non-apoptotic form of programmed cell death. Cell Death Differ 9(8):807–817.

Chang MM, Leeman SE, Niall HD. 1971. Amino-acid sequence of substance P. Nat New Biol 232(29):86–87.

Chang FY, Lee SD, Yeh GH, Wang PS. 1999. Rat gastrointestinal motor responses mediated via activation of neurokinin receptors. J Gastroenterol Hepatol 14(1):39–45.

Cheung D, Timmers MC, Zwinderman AH, den Hartigh J, Dijkman JH, Sterk PJ. 1993. Neutral endopeptidase activity and airway hyperresponsiveness to neurokinin A in asthmatic subjects in vivo. Am Rev Respir Dis 148(6 Pt 1):1467–1473.

Cheung D, van der Veen H, den Hartigh J, Dijkman JH, Sterk PJ. 1994. Effects of inhaled substance P on airway responsiveness to methacholine in asthmatic subjects in vivo. J Appl Physiol 77(3):1325–1332.

Chu HW, Kraft M, Krause JE, Rex MD, Martin RJ. 2000. Substance P and its receptor neurokinin 1 expression in asthmatic airways. J Allergy Clin Immunol 106(4):713–722.

Chung KF, Chang AB. 2002. Therapy for cough: Active agents. Pulm Pharmacol Ther 15(3):335–338.

Colpaert FC, Donnerer J, Lembeck F. 1983. Effects of capsaicin on inflammation and on the substance P content of nervous tissues in rats with adjuvant arthritis. Life Sci 32(16):1827–1834.

Colten HR, Krause JE. 1997. Pulmonary inflammation—a balancing act. N Engl J Med 336(15):1094–1096.

Corboz MR, Rivelli MA, Ramos SI, Rizzo CA, Hey JA. 1998. Tachykinin NK1 receptor-mediated vasorelaxation in human pulmonary arteries. Eur J Pharmacol 350(1):R1–R3.

Covas MJ, Pinto LA, Victorino RM. 1994. Disturbed immunoregulatory properties of the neuropeptide substance P on lymphocyte proliferation in HIV infection. Clin Exp Immunol 96(3):384–388.

Crimi N, Palermo F, Oliveri R, Palermo B, Vancheri C, Polosa R, Mistretta A. 1990. Influence of antihistamine (astemizole) and anticholinergic drugs (ipratropium bromide) on bronchoconstriction induced by substance P. Ann Allergy 65(2):115–120.

Croitoru K, Ernst PB, Bienenstock J, Padol I, Stanisz AM. 1990. Selective modulation of the natural killer activity of murine intestinal intraepithelial leucocytes by the neuropeptide substance P. Immunology 71(2):196–201.

DeFea KA, Vaughn ZD, O'Bryan EM, Nishijima D, Dery O, Bunnett NW. 2000. The proliferative and antiapoptotic effects of substance P are facilitated by formation of a beta-arrestin-dependent scaffolding complex. Proc Natl Acad Sci USA 97(20):11086–11091.

DeRose V, Robbins RA, Snider RM, Spurzem JR, Thiele GM, Rennard SI, Rubinstein I. 1994. Substance P increases neutrophil adhesion to bronchial epithelial cells. J Immunol 152(3):1339–1346.

Di Sebastiano P, di Mola FF, Di Febbo C, Baccante G, Porreca E, Innocenti P, Friess H, Buchler MW. 2000. Expression of interleukin 8 (IL-8) and substance P in human chronic pancreatitis. Gut 47(3): 423–428.

Dimri R, Sharabi Y, Shoham J. 2000. Specific inhibition of glucocorticoid-induced thymocyte apoptosis by substance P. J Immunol 164(5):2479–2486.

Donaldson LF, Haskell CA, Hanley MR. 2001. Messenger RNA localization and further characterisation of the putative tachykinin receptor NK4 (NK3B). Receptors Channels 7(4):259–272.

Dusser DJ, Djokic TD, Borson DB, Nadel JA. 1989. Cigarette smoke induces bronchoconstrictor hyperresponsiveness to substance P and inactivates airway neutral endopeptidase in the guinea pig. Possible role of free radicals. J Clin Invest 84(3):900–906.

Eglezos A, Andrews PV, Boyd RL, Helme RD. 1990. Effects of capsaicin treatment on immunoglobulin secretion in the rat: Further evidence for involvement of tachykinin-containing afferent nerves. J Neuroimmunol 26(2):131–138.

Epperson A, Hatton WJ, Callaghan B, Doherty P, Walker RL, Sanders KM, Ward SM, Horowitz B. 2000. Molecular markers expressed in cultured and freshly isolated interstitial cells of Cajal. Am J Physiol Cell Physiol 279(2):C529–C539.

Fan TP, Hu DE, Guard S, Gresham GA, Watling KJ. 1993. Stimulation of angiogenesis by substance P and interleukin-1 in the rat and its inhibition by NK1 or interleukin-1 receptor antagonists. Br J Pharmacol 110(1):43–49.

Foreman JC. 1987. Substance P and calcitonin gene-related peptide: Effects on mast cells and in human skin. Int Arch Allergy Appl Immunol 82(3–4):366–371.

Gerard NP, Garraway LA, Eddy RL, Jr., Shows TB, Iijima H, Paquet JL, Gerard C. 1991. Human substance P receptor (NK-1): Organization of the gene, chromosome localization, and functional expression of cDNA clones. Biochemistry 30(44):10640–10646.

Germonpre PR, Bullock GR, Lambrecht BN, Van De Velde V, Luyten WH, Joos GF, Pauwels RA. 1999. Presence of substance P and neurokinin 1 receptors in human sputum macrophages and U-937 cells. Eur Respir J 14(4):776–782.

Goetzl EJ, Sreedharan SP. 1992. Mediators of communication and adaptation in the neuroendocrine and immune systems. FASEB J 6(9):2646–2652.

Goode T, O'Connell J, Sternini C, Anton P, Wong H, O'Sullivan GC, Collins JK, Shanahan F. 1998. Substance P (neurokinin-1) receptor is a marker of human mucosal but not peripheral mononuclear cells: Molecular quantitation and localization. J Immunol 161(5):2232–2240.

Goode T, O'Connell J, Anton P, Wong H, Reeve J, O'Sullivan GC, Collins JK, Shanahan F. 2000a. Neurokinin-1 receptor expression in inflammatory bowel disease: Molecular quantitation and localisation. Gut 47(3):387–396.

Goode T, O'Connell J, Ho WZ, O'Sullivan GC, Collins JK, Douglas SD, Shanahan F. 2000b. Differential expression of neurokinin-1 receptor by human mucosal and peripheral lymphoid cells. Clin Diagn Lab Immunol 7(3):371–376.

Goode T, O'Connor T, Hopkins A, Moriarty D, O'Sullivan GC, Collins JK, O'Donoghue D, Baird AW, O'Connell J, Shanahan F. 2003. Neurokinin-1 receptor (NK-1R) expression is induced in human colonic epithelial cells by proinflammatory cytokines and mediates proliferation in response to substance P. J Cell Physiol 197(1): 30–41.

Grady EF, Yoshimi SK, Maa J, Valeroso D, Vartanian RK, Rahim S, Kim EH, Gerard C, Gerard N, Bunnett NW, Kirkwood KS. 2000. Substance P mediates inflammatory oedema in acute pancreatitis via activation of the neurokinin-1 receptor in rats and mice. Br J Pharmacol 130(3):505–512.

Gronblad M, Konttinen YT, Korkala O, Liesi P, Hukkanen M, Polak JM. 1988. Neuropeptides in synovium of patients with rheumatoid arthritis and osteoarthritis. J Rheumatol 15(12):1807–1810.

Guo CJ, Lai JP, Luo HM, Douglas SD, Ho WZ. 2002. Substance P upregulates macrophage inflammatory protein-1beta expression in human T lymphocytes. J Neuroimmunol 131(1–2):160–167.

Hadcock JR, Williams DL, Malbon CC. 1989. Physiological regulation at the level of mRNA: Analysis of steady-state levels of specific mRNAs by DNA-excess solution hybridization. Am J Physiol 256(3 Pt 1):C457–C465.

Haines KA, Kolasinski SL, Cronstein BN, Reibman J, Gold LI, Weissmann G. 1993. Chemoattraction of neutrophils by substance P and transforming growth factor-beta 1 is inadequately explained by current models of lipid remodeling. J Immunol 151(3):1491–1499.

Harrison NK, Dawes KE, Kwon OJ, Barnes PJ, Laurent GJ, Chung KF. 1995. Effects of neuropeptides on human lung fibroblast proliferation and chemotaxis. Am J Physiol 268(2 Pt 1):L278–L283.

Helke CJ, Krause JE, Mantyh PW, Couture R, Bannon MJ. 1990. Diversity in mammalian tachykinin peptidergic neurons: Multiple peptides, receptors, and regulatory mechanisms. FASEB J 4(6): 1606–1615.

Hershey AD, Krause JE. 1990. Molecular characterization of a functional cDNA encoding the rat substance P receptor. Science 247(4945):958–962.

Hislop AA, Wharton J, Allen KM, Polak JM, Haworth SG. 1990. Immunohistochemical localization of peptide-containing nerves in human airways: Age-related changes. Am J Respir Cell Mol Biol 3(3):191–198.

Ho WZ, Kaufman D, Uvaydova M, Douglas SD. 1996. Substance P augments interleukin-10 and tumor necrosis factor-alpha release by human cord blood monocytes and macrophages. J Neuroimmunol 71(1–2):73–80.

Ho WZ, Lai JP, Zhu XH, Uvaydova M, Douglas SD. 1997. Human monocytes and macrophages express substance P and neurokinin-1 receptor. J Immunol 159(11):5654–5660.

Hokfelt T, Pernow B, Wahren J. 2001. Substance P: A pioneer amongst neuropeptides. J Int Med 249(1):27–40.

Holzer P, Holzer-Petsche U. 1997. Tachykinins in the gut. Part II. Roles in neural excitation, secretion, and inflammation. Pharmacol Ther 73(3):219–263.

Hong JL, Rodger IW, Lee LY. 1995. Cigarette smoke-induced bronchoconstriction: Cholinergic mechanisms, tachykinins, and cyclooxygenase products. J Appl Physiol 78(6):2260–2266.

Hopkins B, Powell SJ, Danks P, Briggs I, Graham A. 1991. Isolation and characterisation of the human lung NK-1 receptor cDNA. Biochem Biophys Res Commun 180(2):1110–1117.

Hunter DD, Myers AC, Undem BJ. 2000. Nerve growth factor-induced phenotypic switch in guinea pig airway sensory neurons. Am J Respir Crit Care Med 161(6):1985–1990.

Ichinose M, Nakajima N, Takahashi T, Yamauchi H, Inoue H, Takishima T. 1992. Protection against bradykinin-induced bronchoconstriction in asthmatic patients by neurokinin receptor antagonist. Lancet 340(8830):1248–1251.

Ihara H, Nakanishi S. 1990. Selective inhibition of expression of the substance P receptor mRNA in pancreatic acinar AR42J cells by glucocorticoids. J Biol Chem 265(36):22441–22445.

Jacoby DB, Yost BL, Elwood T, Fryer AD. 2000. Effects of neurokinin receptor antagonists in virus-infected airways. Am J Physiol Lung Cell Mol Physiol 279(1):L59–L65.

Joos GF. 2001. The role of neuroeffector mechanisms in the pathogenesis of asthma. Curr Allergy Asthma Rep 1(2):134–143.

Joos G, Pauwels R, van der Straeten M. 1987a. Effect of inhaled substance P and neurokinin A on the airways of normal and asthmatic subjects. Thorax 42(10):779–783.

Joos GF, Pauwels RA, Van der Straeten ME. 1987b. The role of neuropeptides as neurotransmitters of non-adrenergic, non-cholinergic nerves in bronchial asthma. Bull Eur Physiopathol Respir 23(6): 619–637.

Joos GF, Pauwels RA, van der Straeten ME. 1988. The mechanism of tachykinin-induced bronchoconstriction in the rat. Am Rev Respir Dis 137(5):1038–1044.

Joos GF, Kips JC, Pauwels RA. 1994. In vivo characterization of the tachykinin receptors involved in the direct and indirect bronchoconstrictor effect of tachykinins in two inbred rat strains. Am J Respir Crit Care Med 149(5):1160–1166.

Joos GF, Germonpre PR, Pauwels RA. 2000. Role of tachykinins in asthma. Allergy 55(4):321–337.

Kage R, McGregor GP, Thim L, Conlon JM. 1988. Neuropeptide-gamma: A peptide isolated from rabbit intestine that is derived from gamma-preprotachykinin. J Neurochem 50(5):1412–1417.

Kahler CM, Sitte BA, Reinisch N, Wiedermann CJ. 1993. Stimulation of the chemotactic migration of human fibroblasts by substance P. Eur J Pharmacol 249(3):281–286.

Kaltreider HB, Ichikawa S, Byrd PK, Ingram DA, Kishiyama JL, Sreedharan SP, Warnock ML, Beck JM, Goetzl EJ. 1997. Upregulation of neuropeptides and neuropeptide receptors in a murine model of immune inflammation in lung parenchyma. Am J Respir Cell Mol Biol 16(2):133–144.

Kang BN, Jeong KS, Park SJ, Kim SJ, Kim TH, Kim HJ, Ryu SY. 2001. Regulation of apoptosis by somatostatin and substance P in peritoneal macrophages. Regul Pept 101(1–3):43–49.

Karlsson JA. 1993. A role for capsaicin sensitive, tachykinin containing nerves in chronic coughing and sneezing but not in asthma: A hypothesis. Thorax 48(4):396–400.
Kataeva G, Agro A, Stanisz AM. 1994. Substance-P-mediated intestinal inflammation: Inhibitory effects of CP 96,345 and SMS 201-995. Neuroimmunomodulation 1(6):350–356.
Kaufman MP, Coleridge HM, Coleridge JC, Baker DG. 1980. Bradykinin stimulates afferent vagal C-fibers in intrapulmonary airways of dogs. J Appl Physiol 48(3):511–517.
Keast JR, Furness JB, Costa M. 1985. Distribution of certain peptide-containing nerve fibres and endocrine cells in the gastrointestinal mucosa in five mammalian species. J Comp Neurol 236(3):403–422.
Killingsworth CR, Shore SA, Alessandrini F, Dey RD, Paulauskis JD. 1997. Rat alveolar macrophages express preprotachykinin gene-I mRNA-encoding tachykinins. Am J Physiol 273(5 Pt 1): L1073–L1081.
Kim JS, Rabe KF, Magnussen H, Green JM, White SR. 1995. Migration and proliferation of guinea pig and human airway epithelial cells in response to tachykinins. Am J Physiol 269(1 Pt 1): L119–L126.
Kimura M, Masuda T, Hiwatashi N, Toyota T, Nagura H. 1994. Changes in neuropeptide-containing nerves in human colonic mucosa with inflammatory bowel disease. Pathol Int 44(8):624–634.
King KA, Hu C, Rodriguez MM, Romaguera R, Jiang X, Piedimonte G. 2001. Exaggerated neurogenic inflammation and substance P receptor upregulation in RSV-infected weanling rats. Am J Respir Cell Mol Biol 24(2):101–107.
Koch TR, Carney JA, Go VL. 1987. Distribution and quantitation of gut neuropeptides in normal intestine and inflammatory bowel diseases. Dig Dis Sci 32(4):369–376.
Kraneveld AD, Nijkamp FP. 2001. Tachykinins and neuro-immune interactions in asthma. Int Immunopharmacol 1(9–10):1629–1650.
Krause JE, DiMaggio DA, McCarson KE. 1995. Alterations in neurokinin 1 receptor gene expression in models of pain and inflammation. Can J Physiol Pharmacol 73(7):854–859.
Krco CJ, Gores A, Go VL. 1986. Gastrointestinal regulatory peptides modulate in vitro immune reactions of mouse lymphoid cells. Clin Immunol Immunopathol 39(2):308–318.
Kroegel C, Giembycz MA, Barnes PJ. 1990. Characterization of eosinophil cell activation by peptides. Differential effects of substance P, melittin, and FMET-Leu-Phe. J Immunol 145(8):2581–2587.
Kuo HP, Rohde JA, Tokuyama K, Barnes PJ, Rogers DF. 1990. Capsaicin and sensory neuropeptide stimulation of goblet cell secretion in guinea-pig trachea. J Physiol 431:629–641.
Kuo HP, Hwang KH, Lin HC, Wang CH, Liu CY, Lu LC. 1998. Lipopolysaccharide enhances neurogenic plasma exudation in guinea-pig airways. Br J Pharmacol 125(4):711–716.
Kuo HP, Lin HC, Hwang KH, Wang CH, Lu LC. 2000. Lipopolysaccharide enhances substance P-mediated neutrophil adherence to epithelial cells and cytokine release. Am J Respir Crit Care Med 162(5):1891–1897.
Kuwano K, Bosken CH, Pare PD, Bai TR, Wiggs BR, Hogg JC. 1993. Small airways dimensions in asthma and in chronic obstructive pulmonary disease. Am Rev Respir Dis 148(5):1220–1225.
Kuwano K, Miyazaki H, Hagimoto N, Kawasaki M, Fujita M, Kunitake R, Kaneko Y, Hara N. 1999. The involvement of Fas–Fas ligand pathway in fibrosing lung diseases. Am J Respir Cell Mol Biol 20(1):53–60.
Kwong K, Wu ZX, Kashon ML, Krajnak KM, Wise PM, Lee LY. 2001. Chronic smoking enhances tachykinin synthesis and airway responsiveness in guinea pigs. Am J Respir Cell Mol Biol 25(3): 299–305.
Ladabaum U, Hasler WL. 1999. Novel approaches to the treatment of nausea and vomiting. Dig Dis 17(3):125–132.
Laitinen LA, Heino M, Laitinen A, Kava T, Haahtela T. 1985. Damage of the airway epithelium and bronchial reactivity in patients with asthma. Am Rev Respir Dis 131(4):599–606.
Lamb JP, Sparrow MP. 2002. Three-dimensional mapping of sensory innervation with substance P in porcine bronchial mucosa: Comparison with human airways. Am J Respir Crit Care Med 166(9): 1269–1281.
Lecci A, De Giorgio R, Bartho L, Sternini C, Tramontana M, Corinaldesi R, Giuliani S, Maggi CA. 1999. Tachykinin NK(1)receptor-mediated inhibitory responses in the guinea-pig small intestine. Neuropeptides 33(1):91–97.
Levine JD, Clark R, Devor M, Helms C, Moskowitz MA, Basbaum AI. 1984. Intraneuronal substance P contributes to the severity of experimental arthritis. Science 226(4674):547–549.
Levine JD, Moskowitz MA, Basbaum AI. 1985. The contribution of neurogenic inflammation in experimental arthritis. J Immunol 135(2 Suppl):843s–847s.
Li H, Leeman SE, Slack BE, Hauser G, Saltsman WS, Krause JE, Blusztajn JK, Boyd ND. 1997. A substance P (neurokinin-1) receptor mutant carboxyl-terminally truncated to resemble a naturally occurring receptor isoform displays enhanced responsiveness and resistance to desensitization. Proc Natl Acad Sci USA 94(17): 9475–9480.
Lilly CM, Bai TR, Shore SA, Hall AE, Drazen JM. 1995a. Neuropeptide content of lungs from asthmatic and nonasthmatic patients. Am J Respir Crit Care Med 151(2 Pt 1):548–553.
Lilly CM, Hall AE, Rodger IW, Kobzik L, Haley KJ, Drazen JM. 1995b. Substance P-induced histamine release in tracheally perfused guinea pig lungs. J Appl Physiol 78(4):1234–1241.
Lordal M, Hallgren A, Nylander O, Hellstrom PM. 1996. Tachykinins increase vascular permeability in the gastrointestinal tract of the rat. Acta Physiol Scand 156(4):489–494.
Lotz M, Carson DA, Vaughan JH. 1987. Substance P activation of rheumatoid synoviocytes: Neural pathway in pathogenesis of arthritis. Science 235(4791):893–895.
Lotz M, Vaughan JH, Carson DA. 1988. Effect of neuropeptides on production of inflammatory cytokines by human monocytes. Science 241(4870):1218–1221.
Lundberg JM, Saria A. 1983. Capsaicin-induced desensitization of airway mucosa to cigarette smoke, mechanical, and chemical irritants. Nature 302(5905):251–253.
Lundberg JM, Martling CR, Saria A. 1983. Substance P and capsaicin-induced contraction of human bronchi. Acta Physiol Scand 119(1): 49–53.
Maggi CA. 1990. Capsaicin-sensitive nerves in the gastrointestinal tract. Arch Int Pharmacodyn Ther 303:157–166.
Maggi CA. 1997. The effects of tachykinins on inflammatory and immune cells. Regul Pept 70(2–3):75–90.
Maggi CA. 2000. Principles of tachykininergic co-transmission in the peripheral and enteric nervous system. Regul Pept 93(1–3):53–64.
Mantyh PW. 1991. Substance P and the inflammatory and immune response. Ann NY Acad Sci 632:263–271.
Mantyh CR, Gates TS, Zimmerman RP, Welton ML, Passaro EP, Jr., Vigna SR, Maggio JE, Kruger L, Mantyh PW. 1988. Receptor binding sites for substance P, but not substance K or neuromedin K, are expressed in high concentrations by arterioles, venules, and lymph nodules in surgical specimens obtained from patients with ulcerative colitis and Crohn disease. Proc Natl Acad Sci USA 85(9):3235–3239.
Mantyh CR, Vigna SR, Maggio JE, Mantyh PW, Bollinger RR, Pappas TN. 1994. Substance P binding sites on intestinal lymphoid aggregates and blood vessels in inflammatory bowel disease correspond to authentic NK-1 receptors. Neurosci Lett 178(2):255–259.
Mantyh CR, Maggio JE, Mantyh PW, Vigna SR, Pappas TN. 1996a. Increased substance P receptor expression by blood vessels and lymphoid aggregates in *Clostridium difficile*-induced pseudomembranous colitis. Dig Dis Sci 41(3):614–620.
Mantyh CR, Pappas TN, Lapp JA, Washington MK, Neville LM, Ghilardi JR, Rogers SD, Mantyh PW, Vigna SR. 1996b. Substance P activation of enteric neurons in response to intraluminal *Clostridium difficile* toxin A in the rat ileum. Gastroenterology 111(5): 1272–1280.
Mapp CE, Miotto D, Braccioni F, Saetta M, Turato G, Maestrelli P, Krause JE, Karpitskiy V, Boyd N, Geppetti P, Fabbri LM. 2000. The distribution of neurokinin-1 and neurokinin-2 receptors in human central airways. Am J Respir Crit Care Med 161(1):207–215.
Marshall KW, Chiu B, Inman RD. 1990. Substance P and arthritis: Analysis of plasma and synovial fluid levels. Arthritis Rheum 33(1):87–90.
Martins MA, Shore SA, Drazen JM. 1991a. Capsaicin-induced release of tachykinins: Effects of enzyme inhibitors. J Appl Physiol 70(5):1950–1956.
Martins MA, Shore SA, Drazen JM. 1991b. Release of tachykinins by histamine, methacholine, PAF, LTD4, and substance P from guinea pig lungs. Am J Physiol 261(6 Pt 1):L449–L455.
Martling CR, Theodorsson-Norheim E, Lundberg JM. 1987. Occurrence and effects of multiple tachykinins: Substance P, neurokinin A, and neuropeptide K in human lower airways. Life Sci 40(16): 1633–1643.
Matis WL, Lavker RM, Murphy GF. 1990. Substance P induces the expression of an endothelial-leukocyte adhesion molecule by microvascular endothelium. J Invest Dermatol 94(4):492–495.

Matsumoto S, Takeda M, Saiki C, Takahashi T, Ojima K. 1997. Effects of tachykinins on rapidly adapting pulmonary stretch receptors and total lung resistance in anesthetized, artificially ventilated rabbits. J Pharmacol Exp Ther 283(3):1026–1031.

Matute-Bello G, Liles WC, Steinberg KP, Kiener PA, Mongovin S, Chi EY, Jonas M, Martin TR. 1999. Soluble Fas ligand induces epithelial cell apoptosis in humans with acute lung injury (ARDS). J Immunol 163(4):2217–2225.

Mazumdar S, Das KM. 1992. Immunocytochemical localization of vasoactive intestinal peptide and substance P in the colon from normal subjects and patients with inflammatory bowel disease. Am J Gastroenterol 87(2):176–181.

McGowan IM, Fairhurst RM, Shanahan F, Anton PA. 1997. Mucosal substance P receptor expression in HIV infection and inflammatory bowel disease. Neuroimmunomodulation 4(2):70–76.

Menkes CJ, Renoux M, Laoussadi S, Mauborgne A, Bruxelle J, Cesselin F. 1993. Substance P levels in the synovium and synovial fluid from patients with rheumatoid arthritis and osteoarthritis. J Rheumatol 20(4):714–717.

Metwali A, Blum AM, Ferraris L, Klein JS, Fiocchi C, Weinstock JV. 1994. Eosinophils within the healthy or inflamed human intestine produce substance P and vasoactive intestinal peptide. J Neuroimmunol 52(1):69–78.

Moore TC, Lami JL, Spruck CH. 1989. Substance P increases lymphocyte traffic and lymph flow through peripheral lymph nodes of sheep. Immunology 67(1):109–114.

Mousli M, Bueb JL, Bronner C, Rouot B, Landry Y. 1990. G protein activation: A receptor-independent mode of action for cationic amphiphilic neuropeptides and venom peptides. Trends Pharmacol Sci 11(9):358–362.

Murris-Espin M, Pinelli E, Pipy B, Leophonte P, Didier A. 1995. Substance P and alveolar macrophages: Effects on oxidative metabolism and eicosanoid production. Allergy 50(4):334–339.

Nadel JA. 1991. Neutral endopeptidase modulates neurogenic inflammation. Eur Respir J 4(6):745–754.

Nadel JA, Borson DB. 1991. Modulation of neurogenic inflammation by neutral endopeptidase. Am Rev Respir Dis 143(3 Pt 2):S33–S36.

Nakagawa N, Sano H, Iwamoto I. 1995. Substance P induces the expression of intercellular adhesion molecule-1 on vascular endothelial cells and enhances neutrophil transendothelial migration. Peptides 16(4):721–725.

Nakanishi S. 1991. Mammalian tachykinin receptors. Annu Rev Neurosci 14:123–136.

Naukkarinen A, Harvima I, Paukkonen K, Aalto ML, Horsmanheimo M. 1993. Immunohistochemical analysis of sensory nerves and neuropeptides, and their contacts with mast cells in developing and mature psoriatic lesions. Arch Dermatol Res 285(6):341–346.

Nawa H, Kotani H, Nakanishi S. 1984. Tissue-specific generation of two preprotachykinin mRNAs from one gene by alternative RNA splicing. Nature 312(5996):729–734.

Nieber K, Baumgarten CR, Rathsack R, Furkert J, Oehme P, Kunkel G. 1992. Substance P and beta-endorphin-like immunoreactivity in lavage fluids of subjects with and without allergic asthma. J Allergy Clin Immunol 90(4 Pt 1):646–652.

Nilsson J, von Euler AM, Dalsgaard CJ. 1985. Stimulation of connective tissue cell growth by substance P and substance K. Nature 315(6014):61–63.

Nilsson G, Alving K, Ahlstedt S, Hokfelt T, Lundberg JM. 1990. Peptidergic innervation of rat lymphoid tissue and lung: Relation to mast cells and sensitivity to capsaicin and immunization. Cell Tissue Res 262(1):125–133.

Numao T, Agrawal DK. 1992. Neuropeptides modulate human eosinophil chemotaxis. J Immunol 149(10):3309–3315.

O'Byrne EM, Blancuzzi V, Wilson DE, Wong M, Jeng AY. 1990. Elevated substance P and accelerated cartilage degradation in rabbit knees injected with interleukin-1 and tumor necrosis factor. Arthritis Rheum 33(7):1023–1028.

O'Connor TM, O'Connell J, O'Brien DI, Bennett MW, Goode T, Bredin CP, Shanahan F. 2003. Upregulation of neurokinin-1 receptor expression in the lungs of patients with sarcoidosis. J Clin Immunol 23(5):425–435.

Ollerenshaw SL, Jarvis D, Sullivan CE, Woolcock AJ. 1991. Substance P immunoreactive nerves in airways from asthmatics and nonasthmatics. Eur Respir J 4(6):673–682.

Pascual DW, Xu-Amano JC, Kiyono H, McGhee JR, Bost KL. 1991. Substance P acts directly upon cloned B lymphoma cells to enhance IgA and IgM production. J Immunol 146(7):2130–2136.

Pauwels R, Joos G, Kips JC. 1993. The genetics of Asthma. In: Marsh DG, Lockhart A, Holgate ST, editors. The genetic control of airway responsiveness in rats. Oxford: Blackwell Scientific Publications. pp 113–120.

Pawlik WW, Konturek SJ, Gustaw P, Czarnobilski K, Sendur R, Jaworek J, Yanaihara N. 1992. Role of tachykinins in the control of pancreatic secretion and circulation. J Physiol Pharmacol 43(1): 43–57.

Payan DG, Goetzl EJ. 1985. Modulation of lymphocyte function by sensory neuropeptides. J Immunol 135(2 Suppl):783s–786s.

Perianin A, Snyderman R, Malfroy B. 1989. Substance P primes human neutrophil activation: A mechanism for neurological regulation of inflammation. Biochem Biophys Res Commun 161(2):520–524.

Pernow B. 1983. Substance P. Pharmacol Rev 35(2):85–141.

Piedimonte G, McDonald DM, Nadel JA. 1990. Glucocorticoids inhibit neurogenic plasma extravasation and prevent virus-potentiated extravasation in the rat trachea. J Clin Invest 86(5):1409–1415.

Piedimonte G, Hoffman JI, Husseini WK, Snider RM, Desai MC, Nadel JA. 1993. NK1 receptors mediate neurogenic inflammatory increase in blood flow in rat airways. J Appl Physiol 74(5):2462–2468.

Piedimonte G, Pickles RJ, Lehmann JR, McCarty D, Costa DL, Boucher RC. 1997. Replication-deficient adenoviral vector for gene transfer potentiates airway neurogenic inflammation. Am J Respir Cell Mol Biol 16(3):250–258.

Piedimonte G, Rodriguez MM, King KA, McLean S, Jiang X. 1999. Respiratory syncytial virus upregulates expression of the substance P receptor in rat lungs. Am J Physiol 277(4 Pt 1):L831–L840.

Pizzichini MM, Pizzichini E, Parameswaran K, Clelland L, Efthimiadis A, Dolovich J, Hargreave FE. 1999. Nonasthmatic chronic cough: No effect of treatment with an inhaled corticosteroid in patients without sputum eosinophilia. Can Respir J 6(4):323–330.

Pothoulakis C, Castagliuolo I, LaMont JT, Jaffer A, O'Keane JC, Snider RM, Leeman SE. 1994. CP-96,345: A substance P antagonist, inhibits rat intestinal responses to *Clostridium difficile* toxin A but not cholera toxin. Proc Natl Acad Sci USA 91(3):947–951.

Rameshwar P, Gascon P. 1995. Substance P (SP) mediates production of stem cell factor and interleukin-1 in bone marrow stroma: Potential autoregulatory role for these cytokines in SP receptor expression and induction. Blood 86(2):482–490.

Rameshwar P, Ganea D, Gascon P. 1993. In vitro stimulatory effect of substance P on hematopoiesis. Blood 81(2):391–398.

Rameshwar P, Poddar A, Zhu G, Gascon P. 1997. Receptor induction regulates the synergistic effects of substance P with IL-1 and platelet-derived growth factor on the proliferation of bone marrow fibroblasts. J Immunol 158(7):3417–3424.

Repke H, Bienert M. 1987. Mast cell activation—a receptor-independent mode of substance P action? FEBS Lett 221(2):236–240.

Roques BP, Noble F, Dauge V, Fournie-Zaluski MC, Beaumont A. 1993. Neutral endopeptidase 24.11: Structure, inhibition, and experimental and clinical pharmacology. Pharmacol Rev 45(1):87–146.

Rosati R, Adil MR, Ali MA, Eliason J, Orosz A, Sebestyen F, Kalemkerian GP. 1998. Induction of apoptosis by a short-chain neuropeptide analog in small cell lung cancer. Peptides 19(9):1519–1523.

Saban R, Nguyen N, Saban MR, Gerard NP, Pasricha PJ. 1999. Nerve-mediated motility of ileal segments isolated from NK(1) receptor knockout mice. Am J Physiol 277(6 Pt 1):G1173–G1179.

Schratzberger P, Reinisch N, Prodinger WM, Kahler CM, Sitte BA, Bellmann R, Fischer-Colbrie R, Winkler H, Wiedermann CJ. 1997. Differential chemotactic activities of sensory neuropeptides for human peripheral blood mononuclear cells. J Immunol 158(8): 3895–3901.

Scicchitano R, Biennenstock J, Stanisz AM. 1988. In vivo immunomodulation by the neuropeptide substance P. Immunology 63(4): 733–735.

Serra MC, Bazzoni F, Della Bianca V, Greskowiak M, Rossi F. 1988. Activation of human neutrophils by substance P. Effect on oxidative metabolism, exocytosis, cytosolic Ca^{2+} concentration, and inositol phosphate formation. J Immunol 141(6):2118–2124.

Shanahan F. 1993. Pathogenesis of ulcerative colitis. Lancet 342(8868):407–411.

Shanahan F, Denburg JA, Fox J, Bienenstock J, Befus D. 1985. Mast cell heterogeneity: Effects of neuroenteric peptides on histamine release. J Immunol 135(2):1331–1337.

Sheldrick RL, Rabe KF, Fischer A, Magnussen H, Coleman RA. 1995. Further evidence that tachykinin-induced contraction of human

isolated bronchus is mediated only by NK2-receptors. Neuropeptides 29(5):281–292.
Sheng M, Dougan ST, McFadden G, Greenberg ME. 1988. Calcium and growth factor pathways of c-fos transcriptional activation require distinct upstream regulatory sequences. Mol Cell Biol 8(7): 2787–2796.
Shore SA, Stimler-Gerard NP, Coats SR, Drazen JM. 1988. Substance P-induced bronchoconstriction in the guinea pig. Enhancement by inhibitors of neutral metalloendopeptidase and angiotensin-converting enzyme. Am Rev Respir Dis 137(2):331–336.
Smith CH, Barker JN, Morris RW, MacDonald DM, Lee TH. 1993. Neuropeptides induce rapid expression of endothelial cell adhesion molecules and elicit granulocytic infiltration in human skin. J Immunol 151(6):3274–3282.
Solway J, Leff AR. 1991. Sensory neuropeptides and airway function. J Appl Physiol 71(6):2077–2087.
Solway J, Kao BM, Jordan JE, Gitter B, Rodger IW, Howbert JJ, Alger LE, Necheles J, Leff AR, Garland A. 1993. Tachykinin receptor antagonists inhibit hyperpnea-induced bronchoconstriction in guinea pigs. J Clin Invest 92(1):315–323.
Sonea IM, Palmer MV, Akili D, Harp JA. 2002. Treatment with neurokinin-1 receptor antagonist reduces severity of inflammatory bowel disease induced by *Cryptosporidium parvum*. Clin Diagn Lab Immunol 9(2):333–340.
Stanisz AM, Befus D, Bienenstock J. 1986. Differential effects of vasoactive intestinal peptide, substance P, and somatostatin on immunoglobulin synthesis and proliferations by lymphocytes from Peyer's patches, mesenteric lymph nodes, and spleen. J Immunol 136(1):152–156.
Stead RH, Tomioka M, Quinonez G, Simon GT, Felten SY, Bienenstock J. 1987. Intestinal mucosal mast cells in normal and nematode-infected rat intestines are in intimate contact with peptidergic nerves. Proc Natl Acad Sci USA 84(9):2975–2979.
Stead RH, Dixon MF, Bramwell NH, Riddell RH, Bienenstock J. 1989. Mast cells are closely apposed to nerves in the human gastrointestinal mucosa. Gastroenterology 97(3):575–585.
Strigas J, Burcher E. 1996. Autoradiographic localization of tachykinin NK2 and NK1 receptors in the guinea-pig lung, using selective radioligands. Eur J Pharmacol 311(2–3):177–186.
Sturiale S, Barbara G, Qiu B, Figini M, Geppetti P, Gerard N, Gerard C, Grady EF, Bunnett NW, Collins SM. 1999. Neutral endopeptidase (EC 3.4.24.11) terminates colitis by degrading substance P. Proc Natl Acad Sci USA 96(20):11653–11658.
Swain MG, Agro A, Blennerhassett P, Stanisz A, Collins SM. 1992. Increased levels of substance P in the myenteric plexus of Trichinella-infected rats. Gastroenterology 102(6):1913–1919.
Takebayashi T, Abraham J, Murthy GG, Lilly C, Rodger I, Shore SA. 1998. Role of tachykinins in airway responses to ozone in rats. J Appl Physiol 85(2):442–450.
Takeda Y, Chou KB, Takeda J, Sachais BS, Krause JE. 1991. Molecular cloning, structural characterization, and functional expression of the human substance P receptor. Biochem Biophys Res Commun 179(3):1232–1240.
Takeyama M, Nagai S, Mori K, Ikawa K, Satake N, Izumi T. 1996. Substance P-like immunoreactive substance in bronchoalveolar lavage fluids from patients with idiopathic pulmonary fibrosis and pulmonary sarcoidosis. Sarcoidosis Vasc Diffuse Lung Dis 13(1): 33–37.
Tatemoto K, Lundberg JM, Jornvall H, Mutt V. 1985. Neuropeptide K: Isolation, structure, and biological activities of a novel brain tachykinin. Biochem Biophys Res Commun 128(2):947–953.
Tiberio IF, Turco GM, Leick-Maldonado EA, Sakae RS, Paiva SO, do Patrocinio M, Warth TN, Lapa e Silva JR, Saldiva PH, Martins MA. 1997. Effects of neurokinin depletion on airway inflammation induced by chronic antigen exposure. Am J Respir Crit Care Med 155(5):1739–1747.
Tomaki M, Ichinose M, Miura M, Hirayama Y, Yamauchi H, Nakajima N, Shirato K. 1995. Elevated substance P content in induced sputum from patients with asthma and patients with chronic bronchitis. Am J Respir Crit Care Med 151(3 Pt 1):613–617.
Trautmann A, Schmid-Grendelmeier P, Kruger K, Crameri R, Akdis M, Akkaya A, Brocker EB, Blaser K, Akdis CA. 2002. T cells and eosinophils cooperate in the induction of bronchial epithelial cell apoptosis in asthma. J Allergy Clin Immunol 109(2):329–337.
Tripp RA, Barskey A, Goss L, Anderson LJ. 2002. Substance P receptor expression on lymphocytes is associated with the immune response to respiratory syncytial virus infection. J Neuroimmunol 129(1–2):141–153.
Uddman R, Hakanson R, Luts A, Sundler F. 1997. Autonomic Control of the Respiratory System. In: Barnes PJ, editor. Distribution of neuropeptides in airways. London: Harvard Academic.
Umeno E, Nadel JA, McDonald DM. 1990. Neurogenic inflammation of the rat trachea: Fate of neutrophils that adhere to venules. J Appl Physiol 69(6):2131–2136.
Van Rensen EL, Hiemstra PS, Rabe KF, Sterk PJ. 2002. Assessment of microvascular leakage via sputum induction: The role of substance P and neurokinin A in patients with asthma. Am J Respir Crit Care Med 165(9):1275–1279.
Van Schoor J, Joos GF, Pauwels RA. 2002. Effect of inhaled fluticasone on bronchial responsiveness to neurokinin A in asthma. Eur Respir J 19(6):997–1002.
Veronesi B, Carter JD, Devlin RB, Simon SA, Oortgiesen M. 1999. Neuropeptides and capsaicin stimulate the release of inflammatory cytokines in a human bronchial epithelial cell line. Neuropeptides 33(6):447–456.
Vieira JE, Warth Mdo P, Leme AS, Maldonado EA, King M, Saldiva PH, Martins MA. 1997. Airway responses to capsaicin in guinea pigs: Role of NK-1 and NK-2 neurokinin receptors. Exp Lung Res 23(1):85–99.
Vishwanath R, Mukherjee R. 1996. Substance P promotes lymphocyte-endothelial cell adhesion preferentially via LFA-1/ICAM-1 interactions. J Neuroimmunol 71(1–2):163–171.
von Euler US, Gaddum JH. 1931. An unidentified depressor substance in certain tissue extracts. J Physiol (London) 72:74–87.
Weihe E, Nohr D, Michel S, Muller S, Zentel HJ, Fink T, Krekel J. 1991. Molecular anatomy of the neuro-immune connection. Int J Neurosci 59(1–3):1–23.
Weinstock JV, Blum A, Walder J, Walder R. 1988. Eosinophils from granulomas in murine schistosomiasis mansoni produce substance P. J Immunol 141(3):961–966.
Wershil BK, Castagliuolo I, Pothoulakis C. 1998. Direct evidence of mast cell involvement in *Clostridium difficile* toxin A-induced enteritis in mice. Gastroenterology 114(5):956–964.
Wu ZX, Satterfield BE, Fedan JS, Dey RD. 2002. Interleukin-1beta-induced airway hyperresponsiveness enhances substance P in intrinsic neurons of ferret airway. Am J Physiol Lung Cell Mol Physiol 283(5):L909–L917.
Ziche M, Morbidelli L, Pacini M, Geppetti P, Alessandri G, Maggi CA. 1990. Substance P stimulates neovascularization in vivo and proliferation of cultured endothelial cells. Microvasc Res 40(2):264–278.