

EDITORIAL

The role of neurotrophins in the pathogenesis of asthma and related diseases: allergy and asthma as prototypic neuro-immune diseases?

Biology of neurotrophins and neurotrophin receptors

The neurotrophins are a family of homologous proteins that so far consist of four members: nerve growth factor (NGF), brain-derived neurotrophic factor (BDNF), neurotrophin-3 (NT-3) and neurotrophin-4/5 (NT-4/5). They are synthesized first as precursor proteins, which then are cleaved intracellularly to mature proteins of about 120 amino acids in length [1]. The mature proteins associate as non-covalent homodimers with a high degree of genomic, structural and biological homology among various species. Additionally, the neurotrophins share a significant level of homology to one another and elicit similar types of biological responses.

Their role in survival, differentiation and maintenance of neurons is well defined [2]. They exhibit partially overlapping but also distinct patterns of expression and cellular targets. In addition to the central nervous system, neurotrophins also effect peripheral afferent and efferent neurons. The trophic (survival promoting) and tropic (directing axon growth) properties of neurotrophins demonstrate their potential for the treatment of central as well as peripheral neuropathies.

However, other molecules such as glia cell line-derived growth factor (GDNF) and leukaemia inhibitory factor (LIF) also have neurotrophic activities but they use different receptor systems as neurotrophins and will not be further discussed in this article.

The biological effects of neurotrophins are mediated by binding to two different receptor types, the tropomyosin-related kinase (trk) family of receptor tyrosine kinases and the p75 receptor, a member of the tumour necrosis factor receptor superfamily [3]. Neurotrophins bind to the trk receptors with high affinity and specificity, trkA is primarily a receptor for NGF, trkB for BDNF and NT-4/5 and trkC for NT-3. In addition, all neurotrophins bind to the low affinity receptor p75NTR. Binding of neurotrophins induces receptor dimerization at the cell surface followed by phosphorylation of receptor tyrosine residues. The phosphorylated tyrosine then recruit intracellular signal transduction proteins. These factors initiate functional changes such as survival, proliferation and differentiation of the corresponding target cell.

Expression of neurotrophins and their receptors in allergic disease

Elevated serum NGF levels have been found in humans with asthma and various other allergic diseases such as rhinoconjunctivitis, urticaria-angioedema and atopic eczema [4, 5]. In

patients with asthma segmental allergen provocation increases BDNF, NGF and NT-3 concentrations in bronchoalveolar fluid [6]. During allergic rhinitis, NGF levels are significantly increased in nasal fluid, and nasal provocation with allergen-increased NGF levels in allergic patients [7]. Since local as well as systemic concentrations of neurotrophins are enhanced in allergic inflammatory diseases, the origin of their production is becoming the focus of clinical and basic research.

The traditional cellular sources of neurotrophins under physiological conditions are nerve-associated cells or neurones themselves [2]. However, a wide range of haematopoietic cells including lymphocytes, macrophages and mast cells are also able to produce these factors [8, 9]. After segmental allergen provocation, BDNF and NGF levels were unchanged 20 min after provocation but markedly increased in the bronchoalveolar lavage fluid after 18 h. These data indicate that neurotrophin production in the lung is associated with late-phase allergic responses but not with early-phase response [6]. In animal models it has been shown that infiltrating immune cells are a major source of NGF and BDNF during airway inflammation. T cells play a central role in orchestrating the allergic inflammation and activated T cells are strong producers of NGF [10] and BDNF [11]. However, alveolar macrophages [12], eosinophils [13] and mast cells [14] are also able to secrete these factors.

In addition to the neurotrophins their receptors have also been analysed in lymphoid tissues during the last years. Thereby expression of the panneurotrophin receptor p75NTR as well as the specific receptors trkA, trkB and trkC were found in various immune cells [15]. In a former study trkB and trkC positive eosinophils were described in human bone marrow [16]. In this issue of *Clinical and Experimental Allergy* Noga et al. [17] further demonstrate the expression of trkA, trkB and trkC by eosinophils purified from human blood. Since eosinophilic infiltration is a hallmark of allergic inflammatory responses, neurotrophin receptor expression on these cells might be an additional pathway regulating inflammatory responses. In addition, Noga et al. demonstrate that all three receptors were functionally active, e.g. NGF treatment of eosinophils resulted in an up-regulation of IL-4 production.

Taken together, neurotrophins and their receptors are widely expressed in various cell types of the adaptive as well as the innate immune system. All these cells are mainly involved in initiation and propagation of the allergic inflammatory response. These data provide evidence for a novel phenotype of the inflammatory response in allergy and asthma. It will now be important to link this phenotype to functional properties of the allergic immune response in these clinical conditions.

Function of neurotrophins in allergic disease

Beyond their originally described effects in regulating neuronal growth, neurotrophins are also involved in other biological processes. During the last decade, it has been recognized that neurotrophins are important players in immune regulation. In allergic inflammatory diseases not only immunological dysregulation but also a complex neuro-immune interaction contributes to the pathogenesis of the disease.

Major pathophysiological events in allergic asthma are: chronic inflammation of the airways, development of airway hyper-reactivity and recurrent airway remodelling. Furthermore, it has been recognized that many types of immune cells are involved in the inflammatory process including mast cells, eosinophils, B/T lymphocytes and antigen-presenting cells. The pathogenesis of asthma is especially characterized by a complex interaction between neurons and inflammatory, epithelial and smooth muscle cells. This suggests that in addition to the immunological dysregulation in allergy and asthma, other aspects need to be considered in the pathology of this disease. A hallmark in allergic asthma is the altered neuronal control of the smooth muscle tone. Sensory and motor neurons innervating the lung exhibit a great degree of functional plasticity in asthma defined as 'neuronal plasticity' [18]. In a guinea-pig model, NGF contributes to hypersensitivity by inducing a phenotypic switch in sensory neurons [19]. Further evidence that neurotrophins mediate sensory nerve plasticity originated from genetically modified animals. Neurotrophin receptor p75 gene-deficient mice exhibit a deficit in sensory innervation [20]. Vice versa, transgenic mice with constitutive overexpression of NGF in the lung epithelia exhibit hyperinnervation in the airways by sympathetic and tachykinin-containing nerve fibres [21].

Increased expression of tachykinins by sensory nerves is apparent in airway inflammation and elevated levels of neuropeptides have been found in the lung of asthmatic patients as well as in bronchoalveolar fluid after allergen challenge [22]. The neuronal tachykinin production is regulated by BDNF and NGF [19, 23] and neuropeptides are involved in several key features of asthma, including airway smooth muscle constriction, vascular dilatation, and augmentation of airway inflammation. Activities of tachykinins on immune cells include a broad range of functional responses from eosinophils, lymphocytes mast cells and macrophages. The neuronal control of the acute inflammatory response resulted in the concept of 'neurogenic inflammation' [9].

Neurotrophins are not only produced by but also exhibit profound effects on immune cells residing in the airways. These effects can be described by the term 'immunological plasticity'. Overexpression of NGF in the lung of transgenic mice mediates an increased neutrophil infiltration after ozone-induced lung inflammation [24]. The release of proinflammatory cytokines, an important mechanism during development of the allergic reaction, is regulated by neurotrophins. NGF augments IL-4 and IL-5 production of allergen-activated lymphocytes and this results in enhanced levels of IgE and IgG1 but not IgG2a antibodies [10]. Furthermore, basophils represent a substantial source of IL-4 and IL-13 in the airways and NGF can directly stimulate IgE-induced IL-13 secretion of basophils [25]. These data clearly indicate that neurotrophins act as amplifiers of the locally occurring immune response.

As discussed above, immune cells are able to express neurotrophin factors together with their corresponding receptors. Therefore one can presume that neurotrophins may act as general autocrine growth or survival factors for immune cells. In fact, NGF is an autocrine survival factor for memory B cells [26, 27], mast cells [28] and monocytes [29]. However, the present data indicate that NGF has a protective role against cell apoptosis rather than a direct growth-promoting effect. Regulation of immune cell apoptosis and survival involves autocrine circuits such as expression of the Fas/Apo1 and its ligand as well as the NGF ligand/receptor system, and both systems are linked by modulating anti-apoptotic intracellular Bcl-2 protein expression through endogenous NGF signalling [26]. The evidence that this autocrine circuit is more 'public' by involving secretion of ligands and their extracellular signalling instead of 'private' or intracrine strongly further suggests a complex network of 'social' control over the fate of a single immune cell.

Thus, for understanding the possible mechanisms of cellular effects the microenvironment of neurotrophin and neurotrophin receptor expression must also be taken into account. The expression of neurotrophins and their receptors are involved in a complex multicellular network to maintain the integrity of the epithelial, immune and nervous system in mucosal tissues. This is most evident in the case of eosinophilic neurotrophin and neurotrophin receptor expression as described by Noga et al. in this issue of *Clinical and Experimental Allergy* [17]. Most of the eosinophils in the human body are distributed in the mucosal tissues in which the epithelial surfaces are exposed to the external environment. The tissue-resident eosinophils, similar to mast cells, communicate with other cells of the tissue compartment by release or response of autocrine and paracrine factors such as cytokines, or even the neurotrophins. In this regard, visceral epithelia are a major source of BDNF in many organs [30]. On the other hand, epithelia that express BDNF lack both the high- and low-affinity receptors for BDNF, trkB and p75^{NTR} [30]. Thus, the main physiologic role of the constitutive epithelial BDNF production may be the regulation of the homeostasis of eosinophils (and other tissue-resident immune cells, including mast cells, dendritic cells) as well as neurons in this tissue compartment. However, during an inflammatory response the additional production of neurotrophins by activated tissue-immune cells may initiate a vicious circle of neurotrophin expression and signalling which contributes to the pathogenesis of allergic airway inflammation as discussed in this article.

Conclusion

There is now growing evidence that neurotrophins contribute to the neuronal as well as immunological dysregulations during allergic airway inflammation. Since both the immune and nervous system share many features including recognition of and reaction to unknown stimuli, it is not surprising that both systems use similar mediators to perform their tasks. Through their widespread expression in immune competent cells, neurotrophins and their receptors are candidate molecules for regulating immune as well as neuroimmune interactions. As discussed above, the immunotrophic function of the neurotrophins in addition to their 'traditional' function as neurotrophic factors is becoming more and more evident. Thus, blocking overexpression of neurotrophins during an allergic

inflammatory process may be an important novel tool of therapeutic intervention in the future.

References

- Chao MV, Bothwell M. Neurotrophins: To cleave or not to cleave. *Neuron* 2002; 33:9–12.
- Lewin GR, Barde YA. Physiology of neurotrophins. *Annu Rev Neurosci* 1996; 19:289–317.
- Kaplan DR, Miller FD. Neurotrophin signal transduction in the nervous system. *Curr Opin Neurobiol* 2000; 10:381–91.
- Bonini S, Lambiasi A, Bonini S et al. Circulating nerve growth factor levels are increased in humans with allergic diseases and asthma. *Proc Natl Acad Sci USA* 1996; 93:10955–60.
- Kupfer J, Gieler U, Braun A, Niemeier V, Huzler C, Renz H. Stress and atopic eczema. *Int Arch Allergy Immunol* 2001; 124:353–5.
- Virchow C, Julius P, Lommatsch M, Luttmann W, Renz H, Braun A. Neurotrophins are increased in bronchoalveolar lavage fluid after segmental allergen provocation. *Am J Respir Crit Care Med* 1998; 158:2002–5.
- Sanico AM, Stanisiz AM, Gleeson TD et al. Nerve growth factor expression and release in allergic inflammatory disease of the upper airways. *Am J Respir Crit Care Med* 2000; 161:11631–5.
- Kerschensteiner M, Gallmeier E, Behrens L et al. Activated human T cells, B cells, and monocyte produce brain-derived neurotrophic factor in vitro and in inflammatory brain lesions: a neuroprotective role of inflammation? *J Exp Med* 1999; 189:865–70.
- Renz H. The role of neurotrophins in bronchial asthma. *Eur J Pharmacol* 2001; 429:231–7.
- Braun A, Appel E, Baruch R et al. Role of nerve growth factor in a mouse model of allergic airway inflammation and asthma. *Eur J Immunol* 1998; 28:3240–51.
- Braun A, Lommatsch M, Mannsfeld A et al. Cellular sources of enhanced brain-derived neurotrophic factor production in mouse model of allergic inflammation. *Am J Respir Cell Mol Biol* 1999; 21:537–46.
- Ricci A, Greco S, Mariotta S, Felici L, Amenta F, Bronzetti E. Neurotrophin and neurotrophin receptor expression in alveolar macrophages: an immunocytochemical study. *Growth Factors* 2000; 18:193–202.
- Kobayashi H, Gleich GJ, Butterfield JH, Kita H. Human eosinophils produce neurotrophins and secrete nerve growth factor on immunologic stimuli. *Blood* 2002; 99:2214–20.
- Skaper SD, Pollock M, Facci L. Mast cells differentially express and release active high molecular weight neurotrophins. *Mol Brain Res* 2001; 97:177–85.
- Braun A, Lommatsch M, Renz H. The role of neurotrophins in allergic bronchial asthma. *Clin Exp Allergy* 2000; 30:178–86.
- Labouyrie E, Dubus P, Groppi A et al. Expression of neurotrophins and their receptors in human bone marrow. *Am J Pathol* 1999; 154:405–15.
- Noga O, Englmann C, Hanf G, Grützkau A, Guhl S, Kunkel G. Activation of the specific neurotrophin receptors TrkA, TrkB and TrkC influences the function of eosinophils. *Clin Exp Allergy* 2002; 32:1348–54.
- Braun A, Nockher WA, Renz H. Control of nerve growth and plasticity. *Curr Opin Pharmacol* 2002; 2:229–34.
- Hunter DD, Myers AC, Udem BJ. Nerve growth factor-induced phenotypic switch in guinea pig airway sensory neurons. *Am J Respir Crit Care Med* 2000; 161:1985–90.
- Lee KF, Huber LJ, Landis SC, Sharpe AH, Chao MV, Jaenisch R. Targeted mutation of the gene encoding the low affinity NGF receptor p75 leads to deficits in the peripheral sensory nervous system. *Cell* 1992; 69:737–49.
- Hoyle GW, Graham RM, Finkelstein JB, Nguyen KPT, Gozal D, Friedman M. Hyperinnervation of the airways in transgenic mice overexpressing nerve growth factor. *Am J Respir Cell Mol Biol* 1998; 18:149–57.
- Heaney LG, Cross LJ, McGarvey LP, Buchanan KD, Ennis M, Shaw C. Neurokinin A is the predominant tachykinin in human bronchoalveolar lavage fluid in normal and asthmatic subjects. *Thorax* 1998; 53:357–62.
- Yao L, Zhang D, Bernd P. Differential regulation of substance P by all members of the nerve growth factor family of neurotrophins in avian dorsal root ganglia throughout development. *Neuroscience* 1997; 79:1197–206.
- Graham RM, Friedman M, Hoyle GW. Sensory nerves promote ozone-induced lung inflammation in mice. *Am J Respir Crit Care Med* 2001; 164:307–13.
- Sin AZ, Roche EM, Togias A, Lichtenstein LM, Schroeder JT. Nerve growth factor or IL-3 induces more IL-13 production from basophils of allergic subjects than from basophils of nonallergic subjects. *J Allergy Clin Immunol* 2001; 108:387–93.
- Torcia M, Bracci-Laudiero L, Lucibello M et al. Nerve growth factor is an autocrine survival factor for memory B lymphocytes. *Cell* 1996; 85:345–56.
- Kronfeld I, Kazimirsky G, Gelfand EW, Brodie C. NGF rescues human B lymphocytes from anti-IgM induced apoptosis by activation of PKC ζ . *Eur J Immunol* 2002; 32:136–43.
- Kanbe N, Kurosawa M, Miyachi Y, Kanbe M, Saitoh H, Matsuda H. Nerve growth factor prevents apoptosis of cord blood-derived human cultured mast cells synergistically with stem cell factor. *Clin Exp Allergy* 2000; 30:1113–20.
- Ia Sala A, Corinti S, Federici M, Saragovi HU, Girolomoni G. Ligand activation of nerve growth factor receptor TrkA protects monocytes from apoptosis. *J Leukoc Biol* 2000; 68:104–10.
- Lommatsch M, Braun A, Mannsfeldt A et al. Abundant production of brain-derived neurotrophic factor by adult visceral epithelia. *Am J Pathol* 1999; 155:1183–93.

W. A. NOCKHER and H. RENZ

*Department of Clinical Chemistry and Molecular Diagnostics
Central Laboratory
Philipps-University Marburg
Baldingerstr
D-35033 Marburg
Germany
E-mail: nockher@med.uni-marburg.de*