

- Guidelines for management of asthma. I. Chronic persistent asthma. *BMJ* 1990;**301**:651–3.
- 9 **Woolcock A**, Rubinfeld AR, Seale JP, *et al*. Thoracic Society of Australia and New Zealand. Asthma management plan, 1989. *Med J Aust* 1989;**151**:650–3.
  - 10 **Wong GWK**, Leung TF, Ko FWS, *et al*. Declining asthma prevalence in Hong Kong Chinese schoolchildren. *Clin Exp Allergy* 2004;**34**:1550–5.
  - 11 **Hong S-J**, Lee M-S, Sohn MH, *et al*. Self-reported prevalence and risk factors of asthma among Korean adolescents: 5-year follow-up study, 1995–2000. *Clin Exp Allergy* 2004;**34**:1556–62.
  - 12 **Braun-Fahrländer C**, Gassner M, Grize L, *et al*. No further increase in asthma, hay fever and atopic sensitisation in adolescents living in Switzerland. *Eur Respir J* 2004;**23**:407–13.
  - 13 **Anderson HR**, Ruggles R, Strachan DP, *et al*. Trends in prevalence of symptoms of asthma, hay fever, and eczema in 12–14 year olds in the British Isles, 1995–2002: questionnaire survey. *BMJ* 2004;**328**:1052–3.
  - 14 **Devenny A**, Wassall H, Ninan T, *et al*. Respiratory symptoms and atopy in children in Aberdeen: questionnaire studies of a defined school population repeated over 35 years. *BMJ* 2004;**329**:489–90.
  - 15 **Toelle BG**, Ng K, Belousova E, *et al*. Prevalence of asthma and allergy in schoolchildren in Belmont, Australia: three cross sectional surveys over 20 years. *BMJ* 2004;**328**:386–7.
  - 16 **Robertson CF**, Roberts MF, Kappers JH. Asthma prevalence in Melbourne schoolchildren: have we reached the peak? *Med J Aust* 2004;**180**:273–6.
  - 17 **Mommers M**, Gielkens-Sijstermans C, Swaen GMH, *et al*. Trends in the prevalence of respiratory symptoms and treatment in Dutch children over a 12 year period: results of the fourth consecutive survey. *Thorax* 2005;**60**:000–0.

## Angiogenesis and VEGF in COPD

# Angiogenesis and vascular endothelial growth factor in COPD

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## A possible role for VEGF in the pathology of asthma and COPD

It has been recognised for several hundred years that the bronchial vasculature is an extensive one with early descriptions by Ruysch and possibly Da Vinci.<sup>1</sup> However, its function and regulation in health and in disease remain poorly understood. Studies a number of years ago suggested an increased number of bronchial vessels in asthma where increased collagen IV staining, a marker of new vessels, was seen in bronchial biopsies of asthmatic airways compared with controls.<sup>2</sup> Subsequent studies by the same group and by Salvato and colleagues have confirmed the presence of angiogenesis in the bronchial circulation in asthma.<sup>3–4</sup> There are a number of candidate angiogenic factors for these changes, perhaps the most important of which are vascular endothelial growth factor (VEGF) and the angiopoietins which are distinct molecules that act together at different stages of angiogenic processes in several biological systems.<sup>5–11</sup> Other molecules with angiogenic potential are fibroblast growth factor,<sup>9</sup> angiogenin,<sup>9</sup> and chemokines such as interleukin (IL)-8<sup>12</sup> and eotaxin.<sup>13</sup> Interestingly, angiogenesis seems to be a feature of inflammatory diseases at a number of sites in the body including the joints<sup>14</sup> and the gut.<sup>15</sup>

The study published in this issue of *Thorax* by Kranenburg and colleagues<sup>16</sup> suggests that bronchial vascular changes may also occur in chronic obstructive pulmonary disease (COPD). The authors measured the cellular

expression pattern of VEGF and its receptors Flt-1 and KDR/Flk-1 by immunohistochemistry in central and peripheral airways obtained from ex-smokers with or without COPD. They found that VEGF Flt-1 and KDR/Flt-1 immunostaining was localised in vascular and airway smooth muscle cells, epithelial cells, and macrophages. In contrast, the endothelial cells did not express VEGF but did express its receptors, consistent with them being effector cells for VEGF to act on rather than an important autocrine source.

VEGF expression on bronchial tissues was higher in patients with COPD than in those without COPD. VEGF exists as at least five gene products A–E with VEGF-A being the most potent.<sup>17</sup> VEGF-A has several spliced variants which are expressed by airway cells.<sup>17</sup> A number of conditions relevant to COPD have been shown to increase VEGF expression and release including cigarette smoke,<sup>18</sup> hypoxia,<sup>19</sup> and cytokines such as IL-1 $\beta$  and transforming growth factor (TGF)- $\beta$ .<sup>20</sup>

What is the significance of VEGF expression patterns in the bronchial vessels in asthma and COPD and how might VEGF contribute to the pathology of these diseases? Increased airway wall thickening would cause enhanced airway narrowing on stimulation with constrictor agents, thereby contributing to bronchial hyperresponsiveness. An increased bronchial vasculature would increase inflammatory cell trafficking

and exudation and transudation of mediators, particularly if vascular permeability was altered. The increased vasculature could also contribute to airway hyperresponsiveness by supporting the increased airway smooth muscle mass which is a feature of both asthma and COPD.<sup>21</sup> Interestingly, the study by Kranenburg *et al*<sup>16</sup> showed increased staining in bronchial smooth muscle consistent with this hypothesis. An alternative hypothesis, however, is that the increase in the bronchial vasculature is a protective mechanism which results in increased clearance of proinflammatory mediators and cytokines from inflamed airways. Studies in mouse models have helped to shed light on this issue. Lee *et al*<sup>22</sup> showed that VEGF was increased in the lungs in a mouse model of asthma and that VEGF receptor inhibitors inhibited cellular influx as well as inhibiting airway hyperresponsiveness and reducing microvascular leakage.

Interestingly, a number of treatments for airway diseases can modify VEGF production. Glucocorticoids reduce VEGF secretion by structural<sup>17</sup> and inflammatory airway cells<sup>23</sup> in vitro and both glucocorticoids and long acting  $\beta$  agonists have been shown to reduce the vascularity of asthmatic airways in vivo.<sup>2,3</sup> The effect of these agents on VEGF expression and angiogenesis in COPD has not yet been studied but would be interesting to determine. This suggests that strategies targeting VEGF may have a beneficial effect on bronchial wall inflammation and remodelling, at least in asthma. The situation may be more complex in COPD, however, due to coexisting emphysema and pulmonary hypertension.

In addition to studying VEGF expression in bronchial and bronchiolar walls, Kranenburg *et al*<sup>16</sup> also looked at VEGF staining in the alveolar spaces and pulmonary vessels. They found that the epithelial and endothelial cells in the alveolar spaces and the distal airways

were intensively positive for VEGF in patients with COPD. They hypothesise that VEGF secreted by these cells may be involved in the establishment and maintenance of the function of the blood-gas interface. Studies in animal models suggest that VEGF and its receptors play a protective role in the development of emphysema. Kasahara *et al*<sup>24</sup> have shown that inhibition of VEGF receptors with the specific receptor inhibitor SU5416 can cause alveolar cell apoptosis and the development of emphysema in rats. These findings are of interest as they suggest that the increased VEGF expression in the distal airspaces seen by Kranenburg *et al*<sup>16</sup> may be a protective compensating mechanism. Collectively, these studies suggest a paradoxical role for VEGF in the bronchi and air spaces in COPD—with a protective role in the alveolus but a detrimental function in the bronchi and bronchioles.

The situation becomes more complex still when changes in the pulmonary circulation are taken into account. Kranenburg *et al*<sup>16</sup> showed that VEGF expression was increased in the pulmonary vessels in COPD, suggesting a potential role in the development of pulmonary hypertension. There is an extensive literature going back over 10 years looking at the role of VEGF and its receptors in pulmonary hypertension. Pulmonary hypertension is characterised by plexiform lesions in pulmonary vessels and VEGF is expressed inside the plexiform lesions as well as in smooth muscle adjacent to the lesions.<sup>25</sup> Data from animal models suggest that VEGF may have a protective role as inhibition of the VEGF receptor II combined with chronic hypoxia causes cell death dependent pulmonary endothelial cell proliferation and severe pulmonary hypertension.<sup>26</sup> Moreover, overexpression of VEGF using cell based gene transfer reduced monocrotaline induced pulmonary hypertension in rats,<sup>27</sup> and similar findings were reported with adenoviral mediated overexpression in a chronic hypoxia rat model.<sup>28</sup>

The paper by Kranenburg *et al* is therefore an important one which raises many questions about VEGF in COPD. VEGF and its receptors are involved in many processes in COPD including bronchial wall remodelling, emphysema, and pulmonary hypertension. The complexity of these roles suggests

that strategies specifically targeting VEGF in COPD would have unpredictable and possibly opposing effects on some of the different processes.

Over the next few years we are likely to find out a great deal more about the importance of the role of angiogenic processes in a number of different lung diseases and the role that VEGF, its receptors, and other angiogenic factors play in several situations.

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#### REFERENCES

- Mitzner W, Wagner E. On the purported discovery of the bronchial circulation by Leonardo da Vinci. *J Appl Physiol* 1992;73:1196–201.
- Orsida BE, Ward C, Li X, *et al*. Effect of a long-acting  $\beta_2$ -agonist over three months on airway wall vascular remodeling in asthma. *Am J Respir Crit Care Med* 2001;164:117–21.
- Orsida BE, Li X, Hickey B, *et al*. Vascularity in asthmatic airways: relation to inhaled steroid dose. *Thorax* 1999;54:289–95.
- Salvato G. Quantitative and morphological analysis of the vascular bed in bronchial biopsy specimens from asthmatic and non-asthmatic subjects. *Thorax* 2001;56:902–6.
- Asai K, Kanazawa H, Kamoi H, *et al*. Increased levels of vascular endothelial growth factor in induced sputum in asthmatic patients. *Clin Exp Allergy* 2003;33:595–9.
- Kanazawa H, Hirata K, Yoshikawa J. Involvement of vascular endothelial growth factor in exercise induced bronchoconstriction in asthmatic patients. *Thorax* 2002;57:885–8.
- McDonald DM. Angiogenesis and remodeling of airway vasculature in chronic inflammation. *Am J Respir Crit Care Med* 2001;164:S39–45.
- Hoshino M, Nakamura Y, Hamid QA. Gene expression of vascular endothelial growth factor and its receptors and angiogenesis in bronchial asthma. *J Allergy Clin Immunol* 2001;107:1034–8.
- Hoshino M, Takahashi M, Aoike N. Expression of vascular endothelial growth factor, basic fibroblast growth factor, and angiogenin immunoreactivity in asthmatic airways and its relationship to angiogenesis. *J Allergy Clin Immunol* 2001;107:295–301.
- Yancopoulos GD, Davis S, Gale NW, *et al*. Vascular-specific growth factors and blood vessel formation. *Nature* 2000;407:242–8.
- Ribatti D, Vacca A, Presta M. The discovery of angiogenic factors: a historical review. *Gen Pharmacol* 2000;35:227–31.
- Tanner JE. Nucleosomes activate NF-kappaB in endothelial cells for induction of the proangiogenic cytokine IL-8. *Int J Cancer* 2004;112:155–60.
- Salcedo R, Young HA, Ponce ML, *et al*. Eotaxin (CCL11) induces in vivo angiogenic responses by human CCR3+ endothelial cells. *J Immunol* 2001;166:7571–8.
- Walsh DA. Angiogenesis in osteoarthritis and spondylosis: successful repair with undesirable outcomes. *Curr Opin Rheumatol* 2004;16:609–15.
- Kanazawa S, Tsunoda T, Onuma E, *et al*. VEGF, basic-FGF, and TGF-beta in Crohn's disease and ulcerative colitis: a novel mechanism of chronic intestinal inflammation. *Am J Gastroenterol* 2001;96:822–8.
- Kranenburg AR, de Boer WJ, Alagappan VKT, *et al*. Enhanced bronchial expression of vascular endothelial growth factor and receptors (Flk-1 and Flt-1) in patients with chronic obstructive pulmonary disease. *Thorax* 2005;60:106–13.
- Knox AJ, Corbett L, Stocks J, *et al*. Human airway smooth muscle cells secrete vascular endothelial growth factor: up-regulation by bradykinin via a protein kinase C and prostanoind-dependent mechanism. *FASEB J* 2001;15:2480–8.
- Wright JL, Tai H, Churg A. Cigarette smoke induces persisting increases of vasoactive mediators in pulmonary arteries. *Am J Respir Cell Mol Biol*, 2004 (epub ahead of print).
- Nilsson I, Shibuya M, Wennstrom S. Differential activation of vascular genes by hypoxia in primary endothelial cells. *Exp Cell Res* 2004;299:476–85.
- Wen FQ, Liu X, Manda W, *et al*. TH2 Cytokine-enhanced and TGF-beta-enhanced vascular endothelial growth factor production by cultured human airway smooth muscle cells is attenuated by IFN-gamma and corticosteroids. *J Allergy Clin Immunol* 2003;111:1307–18.
- Jeffery PK. Remodeling in asthma and chronic obstructive lung disease. *Am J Respir Crit Care Med* 2001;164:S28–38.
- Lee YC, Kwak YG, Song CH. Contribution of vascular endothelial growth factor to airway hyperresponsiveness and inflammation in a murine model of toluene diisocyanate-induced asthma. *J Immunol* 2002;168:3595–600.
- Horiuchi T, Weller PF. Expression of vascular endothelial growth factor by human eosinophils: upregulation by granulocyte macrophage colony-stimulating factor and interleukin-5. *Am J Respir Cell Mol Biol* 1997;17:70–7.
- Kasahara Y, Tudor RM, Taraseviciene-Stewart L, *et al*. Inhibition of VEGF receptors causes lung cell apoptosis and emphysema. *J Clin Invest* 2000;106:1311–9.
- Tudor RM, Chacon M, Alger L, *et al*. Expression of angiogenesis-related molecules in plexiform lesions in severe pulmonary hypertension: evidence for a process of disordered angiogenesis. *J Pathol* 2001;195:367–74.
- Taraseviciene-Stewart L, Kasahara Y, Alger L, *et al*. Inhibition of the VEGF receptor 2 combined with chronic hypoxia causes cell death-dependent pulmonary endothelial cell proliferation and severe pulmonary hypertension. *FASEB J* 2001;15:427–38.
- Campbell AI, Zhao Y, Sandhu R, *et al*. Cell-based gene transfer of vascular endothelial growth factor attenuates monocrotaline-induced pulmonary hypertension. *Circulation* 2001;104:2242–8.
- Partovian C, Adnot S, Raffestin B, *et al*. Adenovirus-mediated lung vascular endothelial growth factor overexpression protects against hypoxic pulmonary hypertension in rats. *Am J Respir Cell Mol Biol* 2000;23:762–71.