CHAPTER 4

Bronchial hyperresponsiveness in athletes: mechanisms for development

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Bronchial hyperresponsiveness (BHR) is an abnormal increase in airflow limitation following exposure to a nonallergenic stimulus [1]. A high prevalence of BHR, relative to the general population, has been reported in athletes. It is now thought that strenuous activity itself may be a contributing factor for the development of BHR in young athletes [2]. However, the airway response to the different provoking stimuli used to measure BHR is not uniform in athletes and the presence or absence of BHR is seldom simply a "yes" or "no" answer. Furthermore, there is normally a substantial overlap in bronchial responsiveness between health and disease. This is demonstrated well in epidemiological studies where a significant percentage of people with BHR have never had any symptoms of asthma [3, 4]. This is probably explained by the high cut-off points (*e.g.* 16 $mg \cdot mL^{-1}$) used to define BHR when a pharmacological agent, such as methacholine or histamine, is used. Before discussing the development of BHR in athletes, it is important to look at the phenomenon of BHR itself.

BHR is a characteristic feature in asthma but is also commonly found in chronic obstructive pulmonary disease (COPD) [5]. Moreover, a high prevalence of BHR has also been documented in other lung disorders, such as sarcoidosis [6], Sjögren's syndrome [7] and rheumatoid arthritis [8]. BHR appears to be a consequence of many different pathophysiological phenomena and its presence only confirms abnormality not cause. In untreated asthmatics with recent disease, BHR in response to direct stimuli, such as methacholine or histamine, seem to be fairly well linked to underlying inflammation in the lower airways. BHR is known to increase after allergen challenge [9]. Moreover, in steroid-naive asthmatics, there is a significant relationship between BHR and induced sputum eosinophils [10]. The severity of BHR also seems to predict the response to inhaled corticosteroids in asthma [11–13]. Moreover, it was shown that adjustment of treatment according to BHR provides better long-term asthma control then adjusting treatment using standard clinical parameters, such as symptoms and lung function [14].

Whilst BHR to stimuli that act directly to cause smooth muscle contraction seems to relate fairly well to underlying disease activity in previously untreated asthma, the relationship is less impressive in chronic asthmatics treated with inhaled steroids. Despite the absence of inflammation in bronchial biopsies from patients treated with inhaled corticosteroids for years, BHR to histamine was still a prominent feature [15]. Thus, BHR is only partly related to airway inflammation in people with chronic asthma. BHR may be explained in part by lower than normal forced expiratory volume in one second (FEV_1) as demonstrated in patients with moderate-to-severe airflow limitation. The resistance in the peripheral or "small airways" [16] is also likely to contribute to BHR. Airway calibre is important because narrowing serves to amplify bronchial smooth

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muscle contraction (fig. 1). There are other factors, including airway remodelling, altered contractile properties of smooth muscle [17, 18] and thickening of the reticular basement membrane [19], which potentially contribute to BHR in the absence of active inflammation. These changes may result from inflammation, but remain after the inflammation has ceased to be active. In patients with COPD, BHR is related to the degree of airway obstruction as measured by FEV_1 [20, 21]. In asthma, evidence of peripheral airway obstruction measured by forced expiratory flow through the mid-portion of the vital capacity ($FEF_{25–75}$) predicts the BHR response to methacholine [22, 23]. Although some suggest $FEF_{25–75}$ predicts exercise-induced bronchoconstriction [24], this was not demonstrated in a formal study [25]. Difference in baseline airway calibre is also thought to partly explain the sex difference, with females having a slightly higher prevalence of BHR to methacholine [26, 27]. In keeping with the concept that baseline calibre could be an important determinant of response to direct stimuli is the finding that a low sensitivity to detect BHR with a direct stimulus occurred in athletes with good lung function and BHR to indirect stimuli [28].

The direct stimuli commonly used are the pharmacological agonists methacholine or histamine. The indirect physical stimuli that are commonly used include exercise, isocapnic hyperventilation, hypertonic saline, mannitol or distilled water (table 1). There are examples of pharmacological agents that act indirectly to stimuli and the most common one is adenosine monophosphate (AMP).

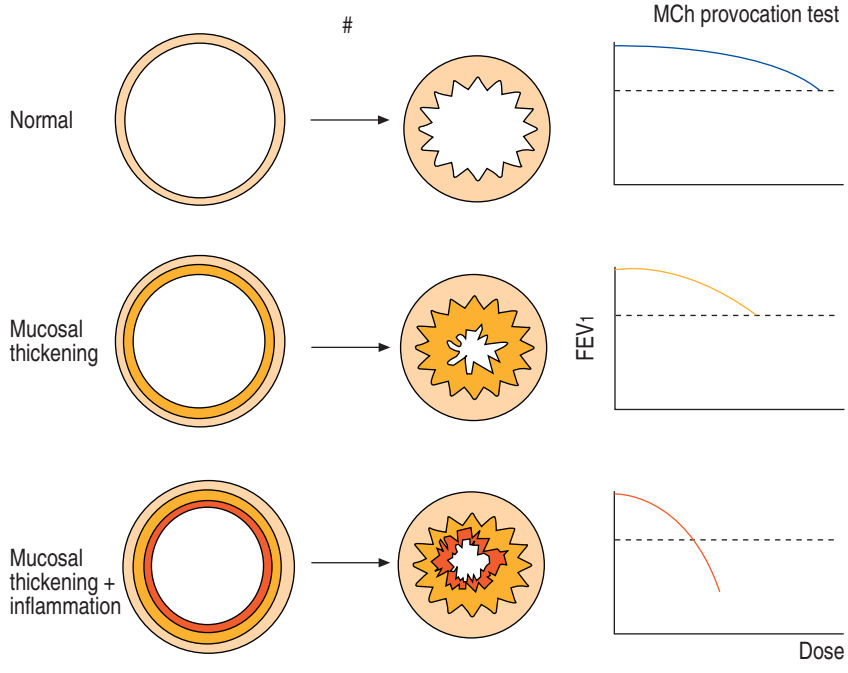

Fig. 1. – Schematic illustration of the mechanisms explaining the degree and slope of bronchial hyperresponsiveness in health and disease. The same degree of muscle contraction (#: 30% narrowing illustrated) induces a different slope response, which is dependent on the degree of mucosal thickening and inflammation. MCh: methacholine.

Table 1. – Direct and indirect stimuli for identifying bronchial hyperresponsiveness

Direct stimuli	Methacholine, histamine, propanolol
Indirect stimuli	Exercise, eucapnic voluntary hyperpnoea, mannitol, hypertonic saline

Although referred to as the presence or absence of BHR, the airway response to the various indirect and direct stimuli varies considerably, and the relationship between the responses is usually poor [29, 30]. In contrast, the responses to histamine and methacholine are more comparable. The reason for this may be that these agents are administered and the response is mediated *via* receptors on the smooth muscle, whereas responses to indirect acting stimuli are dependent on the presence of inflammatory cells and their mediators, in addition to smooth muscle responsiveness. Importantly, the various indirect stimuli share many common characteristics [1]. The response to indirect stimuli can, for example, be inhibited by inhaling sodium cromoglycate and nedocromil sodium, heparin and furosemide [31–33]. Another common characteristic of indirect stimuli is the refractoriness that follows the initial challenge such that the response is less than half following a second challenge. This refractoriness, sometimes called tachyphylaxis, occurs in ~50% of subjects. Cross refractoriness to the indirect stimuli has also been documented [34].

Bronchial hyperresponsiveness in athletes

The prevalence of BHR has been reported to be higher in athletes than in the general population. Most of the studies have been performed using methacholine or histamine and the prevalence of BHR amongst athletes has been reported to be between 1.5–2-times higher than matched controls. The prevalence of exercise-induced bronchospasm (EIB) is also increased relative to the general population, with reported prevalence rates of 11–50% depending on sport activity and the cut-off values used for fall in FEV_1 (15 or 10%) [35, 36]. However, these studies have been limited by failure to include control subjects. LANGDEAU *et al*. [37] investigated the Canadian Olympic Team and found that nearly 50% were positive to methacholine compared with 18% of the healthy controls. However, there was no defined border between the presence and absence of BHR, but more a continuous distribution with a tendency for the athletes to be more responsive [37] (fig. 2).

There are some common risk factors to all athletes and also some risk factors that are specific for the individual sporting activity (table 2). While hyperpnoea of dry, cold air may be a dominating risk factor in cross-country skiers, cyclists are exposed to road dust. As with marathon runners, cyclists are exposed to airborne allergens. Swimmers are exposed to chlorine gas and skaters to ozone and oxides of nitrogen (table 2).

Risk factors for development of BHR in athletes: possible mechanisms

Immune suppression: respiratory tract infections

One common risk factor is the increased vulnerability to respiratory tract infections in all athletes who perform strenuous physical activity. Of nearly 1,300 marathon runners attending the Los Angeles (CA, USA) marathon in 1987, 12.9% reported that they had a respiratory tract infection within 1 week after the run! This value compared with only

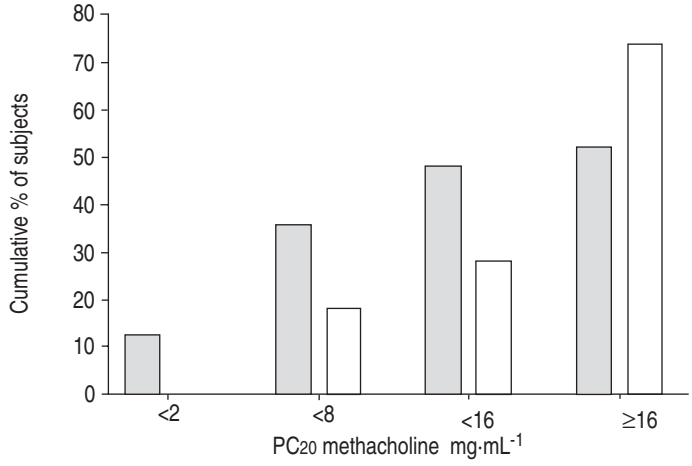

Fig. 2. – Methacholine provocation test amongst 100 high-level athletes from Canada (▒) compared with 50 healthy non-athletic controls (□). A general trend towards increased hyperresponsiveness was seen amongst the athletes. PC_{20}: provocative concentration of methacholine causing a 20% reduction in forced expiratory volume in one second. Reproduced with permission from [37].

Table 2. – Risk factors for development of bronchial hyperresponsiveness in athletes

Irritants
Cyclists (*e.g.* road dust, diesel exhaust)
Swimmers (*e.g.* chloramines)
Skaters (*e.g.* N_2O, ozone)
Cold, dry air
Cross-country skiers
Skaters
Allergens
Marathon runners
Cyclists
Viral infections
All strenuous sport activities

2.2% for the control group [38]. There is evidence that immunosuppression occurs in relation to strenuous activity and is greatest in the hours immediately following strenuous activity. The term "open window theory" is used to refer to this period of suppressed immunity and it occurs between 3–72 h after heavy exercise. During this time there is an increased susceptibility for viral or bacterial infections that might be clinical or subclinical [39]. Potential important immunological features are decreases in: T-cell-mediated immune responses (including reduced proliferative response to lectins [40]); delayed type hypersensitivity reactions [41]; phagocytic activity and oxidative burst amongst macrophages and neutrophils [42]; and NK cell activity [43] (fig. 3).

Another important immunological feature is the decreased humoral response documented after strenuous exercise. A decrease in immunoglobulin (Ig)A concentration in nasal secretions by 70% was observed for $\geq$18 h after racing 31 km [44]. Following strenuous prolonged exercise, salivary secretion rates fall, decreasing the level of IgA-mediated immune protection at the mucosal surface [45, 46]. Moreover, nasal mucociliary transit time is significantly prolonged for several days after a marathon

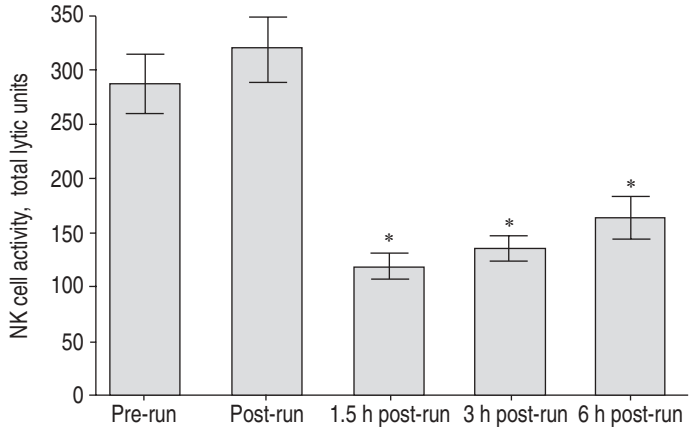

Fig. 3. – Natural killer (NK) cell activity response to 2.5 h of intensive running in 62 marathon runners. Data reproduced with permission from [43]. *: $p<0.05$.

and is caused in part by abnormally functioning ciliated cells [47]. This is presumably due to dehydration of the nasal surface fluid layer making the cilia less efficient. In a recent study of marathon runners competing in the Western States Endurance Run, 26% of those completing the run reported an upper respiratory tract infection (URTI) within 2 weeks of the race. The best predictor of getting an infection was a low serum IgA secretion rate at mid-race after 90 km [48]. Interestingly, lymphoid aggregates mimicking bronchus associated lymphoid tissue (BALT) is commonly found in cross-country skiers with BHR [49] (fig. 4). Even though the pathogenesis is unclear, the present authors can speculate that these changes are an indication of immune response to repeated clinical or subclinical infections [50, 51]. In keeping with this speculation is the finding that training with an URTI induces a long-lasting ($\geq$6 weeks), increased BHR to histamine [52]. Thus, immune suppression may play a role in the development of BHR, at least in endurance athletes.

Influence by (cold) dry air hyperventilation

Airway injury following hyperpnoea of cold, dry air is also likely to be an important risk factor for the development of BHR in cross-country skiers. Repeated dry, cold air hyperpnoea in dogs has been shown to increase resistance in the peripheral airways [53]. This increased resistance is associated with increased production of pro-inflammatory mediators (leukotriene (LT)B_4 and LTC_4) and an increased number of inflammatory cells (neutrophils and eosinophils) in bronchoalveolar lavage (BAL) fluid [53]. The same increase in peripheral airway response has been reported in humans breathing cold, dry air [54]. Dry air represents an osmotic stress to the respiratory mucosa with dehydration [55] leading to shrinkage and sloughing of epithelial cells [56]. Epithelial cells and mast cells are believed to be of special importance, releasing inflammatory mediators, such as histamine, leukotrienes and prostaglandins (PGs), in response to the osmotic effects of dehydration [57, 58]. Elevated levels of urinary LTE_4 have been documented after EIB [59] and increased concentrations of leukotrienes have also been measured in BAL fluid after dry air hyperpnoea [60]. The role of leukotrienes is to sustain the airway narrowing provoked by exercise and this has been demonstrated by pre-medication with a leukotriene receptor antagonist enhancing recovery from EIB [61]. In particular, PGD_2 (measured as the metabolite $9\alpha,11\beta PGF_2$) is also an important mediator and significant

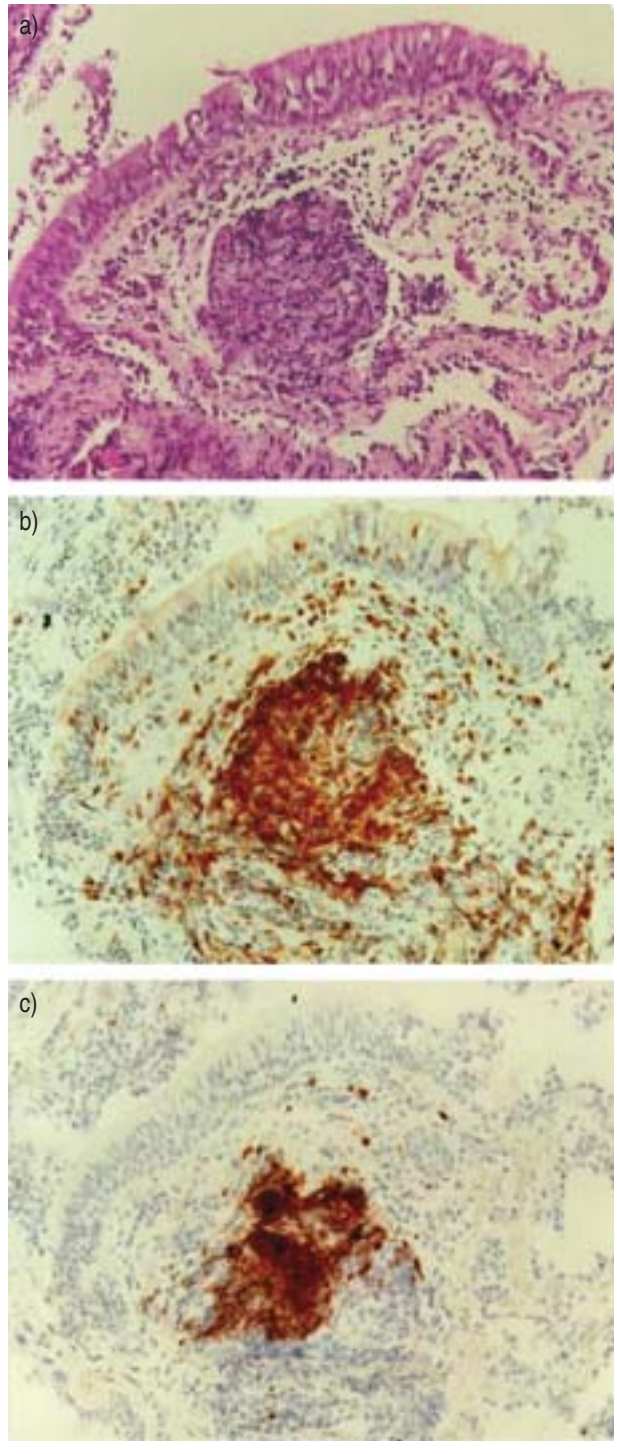

Fig. 4. – Lymphoid aggregates in bronchial mucosa from a skier with asthma symptoms and bronchial hyperresponsiveness. Haematoxylin-eosin staining (a). CD3 stains all T-cells and CD20 all B-cells (b and c). Adapted with permission from [49].

increases in this metabolite have been noted in the urine after exercise [57, 62]. The cyclooxygenase inhibitors, such as flurbiprofen [63] and indomethacin [64], have been shown to partly inhibit the exercise-induced airway response. However, total inhibition of PG synthesis is not necessarily positive because some PGs are protective. For example, epithelial cells synthesise PGE_2 and this plays an important role in refractoriness to exercise, preventing EIB after repeated challenges [65]. Osmotic stimulus to epithelial cells *in vitro* induces release of interleukin (IL)-8 [66]. IL-8 promotes neutrophil chemotaxis, an event that has been reported *in vivo* during EIB [67].

Indeed, it may be the loss of protective PGs that makes the difference in the airway response to exercise between asthmatic and healthy subjects who have BHR. There are several important pieces of information that have come together recently that may help to explain how an athlete may become responsive to exercise. The first is the finding of mast cells in healthy subjects close to the airway surface [68, 69]. The second is the finding that healthy subjects do release mediators, such as PGD_2 and LTs, in response to dehydration stress. Thus, MICKLEBOROUGH *et al*. [57] and CAILLAUD *et al*. [58] found strenuous exercise, and BRANNAN [70] found inhalation of mannitol in healthy fit subjects, was associated with increased urinary excretion of the metabolite of PGD_2 and LTC_4. If the mediators are present in sufficient concentrations then it only remains for the airway smooth muscle to become responsive for the airways of otherwise healthy people to respond. It is the events that might make the airway smooth muscle of the elite athletes sensitive that are of interest. Several mechanisms suggested by different investigators are illustrated in figure 5 [2]. They include airway injury in response to excessive dehydration stress causing exudation of plasma and repeated exposure to circulating substances in the repair process.

Studies in skiers. Skiers from mid-Norway and Sweden aspiring to be elite were investigated with methacholine provocation test, bronchoscopy and BAL. While the skiers in Sweden had their training in a cold, dry climate, the Norwegian skiers trained in a coastal climate that was less cold. The difference in temperature (usually -20 *versus* -5°C) is a possible reason why the prevalence of BHR was higher in Sweden *versus* Norway (45 *versus* 15%). Bronchoscopy and BAL of these skiers revealed evidence of airway remodelling, shown by increased deposition of tenascin and collagen close to the basement membrane. Interestingly, the thickening was the same for both those with and without current BHR to methacholine, indicating that the structural changes were a general consequence of chronic hyperpnoea of cold, dry air [71]. Bronchoscopy and BAL also revealed a pattern of inflammation different from the one usually seen in non-athletic asthmatics. With the exception of a few atopic subjects there was no evidence of eosinophil activation. A slight increase of neutrophils, tumour necrosis factor (TNF)-α and myeloperoxidase was measured in the skiers, as well as increased numbers of mast cells and lymphocytes [72]. However, no significant differences could be seen in the inflammatory pattern between those with or without current BHR. The subjects were also investigated using noninvasive markers of inflammation, which included bronchial provocation with AMP [73] and the measurement of exhaled nitric oxide (eNO). Slightly elevated levels of eNO and increased responsiveness to AMP were measured, but only in a few subjects, all of whom were atopic [74].

A trial of 3 months' treatment with the inhaled corticosteroid budesonide in skiers with exercise-induced "asthma" symptoms and BHR was also conducted. Interestingly, with the exception of a slight improvement in FEV_1 in the budesonide treatment group, no beneficial effect could be measured. Thus, there was no effect of treatment on BHR, airway remodelling or inflammatory indices measured in the biopsies or BAL. In contrast, most of the changes seem to be related to intensity of "dehydration" stress to the

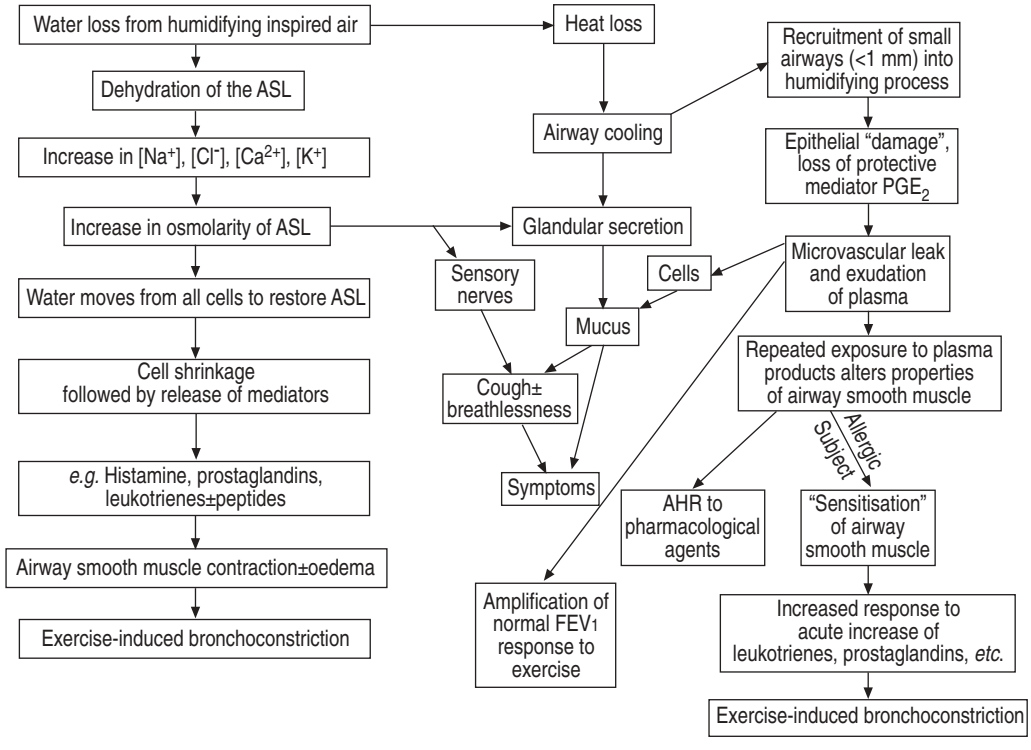

Fig. 5. – Schematic presentation of mechanisms related to hyperpnoea of cold, dry air and effect on the airway mucosa. ASL: airway surface liquid; FEV_1: forced expiratory volume in one second; PGE_2: prostaglandin E_2; AHR: airway hyperresponsivenes. Reproduced with permission from [2].

airways. For example, there was a clear improvement documented in the placebo group when the skiers went from a high intensity to a less intense period of training [75]. This is in keeping with other observations in skiers [76]. A recent report in elite swimmers shows similar findings, with spontaneous recovery occurring in those reducing their exposure following retirement from high-level sport activities [77].

Thus, it seems as if cold, air hyperpnoea in humans produces asthma-like symptoms that in many circumstances differ from those usually seen in asthmatics who are not elite athletes. The role of sensory nerve stimulation in response to increased osmolarity may serve to explain some of these symptoms in the absence of airway narrowing [78, 79].

For winter athletes, chronic dehydration or other stress to the airways may result in exposure to circulating substances with the potential to induce airway remodelling and an increase in contractility of the smooth muscle leading to BHR [2]. In contrast, in summer athletes, the inflammatory response is more characterised by increased numbers of lymphocytes, mast cells and neutrophils, while eosinophils are only seen in those who are also atopic.

Allergen exposure

The hyperpnoea of exercise, especially during the summer, increases the allergen load and thereby the risk of being sensitised to airborne allergens. Under resting conditions, pollen allergens (>10 μm) are usually filtered out by the nose and have the potential to

cause allergic rhinitis. During hyperpnoea, there is a shift from nose to mouth breathing so that there is an increased amount of allergen that enters the lower airways, despite the relatively large size. Many patients with rhinitis have BHR to methacholine or histamine [80]. Bronchoscopies and studies of induced sputum have indicated asymptomatic inflammation in the lower airways in people with allergic rhinitis. This suggests that rhinitis is an intermediate stage and indicates a high risk of developing asthma later. In the 2000 Olympic and Paralympic Games in Sydney (Australia) 56% gave a symptom history consistent with allergic rhinoconjunctivitis, 41% had symptoms of allergic rhinoconjunctivitis and a positive test response to any one allergen, and 29% had seasonal allergic rhinoconjunctivitis (a positive history and at least one positive skin-prick test response to a seasonal allergen) [81]. In elite athletes from Finland, the highest prevalence of asthma was found in swimmers. However, in elite runners, asthma symptoms were closely related to those sensitised to airborne allergens [82]. This finding contrasts with the reported low prevalence of allergy in skiers [83] who are less likely to be exposed to massive amounts of airborne allergens compared with summer athletes.

Influence by irritants

Several studies report a high prevalence of BHR to methacholine or histamine in swimmers [84–87]. However, while 60% had a provocative concentration of methacholine causing a 20% reduction in FEV_1 <8 $mg \cdot mL^{-1}$, only 20% of the swimmers in the Canadian Olympic Team had exercise-induced wheeze or dyspnoea [37]. The same discrepancy between BHR and prevalence of symptoms has been reported by other investigators [88]. In a study on Finnish swimmers, active elite swimmers were compared with those who had retired from active swimming. The group was followed for 5 yrs. The prevalence of atopy in the group that were actively swimming was 56% at baseline and increased to 69% at follow-up. Among the retired swimmers, the atopy prevalence was 46% on both occasions. The prevalence of BHR to histamine was 44% in the active group, increasing to 50% at follow-up, compared with 31% amongst the retired swimmers, decreasing to 12% ($p<0.05$) at follow-up. The change in BHR was associated with evidence of airway inflammation as measured by a slight increase in sputum lymphocytes and eosinophils [77, 89].

Chlorinated pools and asthma development

Chloramine is formed when chlorine from the water reacts with protein from the airways. Increased exposure to chlorine is thought to contribute to the development of BHR in swimmers. Although the concentration of chlorine in the air may not be high, the high ventilation rates of exercise mean that the actual amount of chlorine inhaled during periods of intense training may result in a high total load of chlorine [90].

Further prolonged stays in swimming halls may also induce problems in non-athletes [91]. Reduced plasma Clara cell protein (CC16) levels have not only been reported in swimmers but also in pool attendants who are repeatedly exposed to the chlorine in swimming pools. CC16 is a protein important for mucosal defence and is produced by the Clara cells in the airways. CC16 has been found to be related to BHR in asthmatic children [92], and induction of allergic inflammation in the lower airway in asthmatics is associated with reduced serum CC16 levels [93]. The important role of CC16 in mucosal defence was further explored in a recent study, which indicated an interaction between chlorine and ozone exposure on Clara cell function measured as expression of CC16 [94]. The degree of exposure to natural ozone was measured in children spending 4 h

outdoors. The children were divided in two groups according to those who frequently visited indoor pools and those who seldom visited indoor pools. In the group who frequently visited indoor pools, significantly lower plasma CC16 levels were found. Moreover, during outdoor stay and ozone exposure, there was a tendency of further decrease in CC16 levels in this same group while those who infrequently visited the pool showed an opposite pattern.

Skating and asthma

Increased prevalence of BHR is also reported in skaters [88, 95–97], as well as in ice hockey players [98, 99]. Levy *et al*. [100] measured levels of nitrogen dioxide (NO_2) in different ice halls and found mean daily concentrations of 37–206 parts per billion (ppb) [100]. Exposure to 4 parts per million of NO_2 for 20 min is known to induce inflammation in the lower airways [101]. Exercising at high ventilation for hours in ice halls, with NO_2 concentrations >100 ppb, could result in a cumulative dose above the level that is known to induce inflammation in previously healthy subjects. Interestingly, treatment with montelukast in these ice hockey players with asthma symptoms and presence of BHR did not prove to be beneficial. This is in contrast to what is known from treating exercise-induced asthma in nonatopic subjects and indicates that the phenomenon seen in ice hockey players represents a different pathophysiology. However, a recent study reports a benefit from montelukast when exercise is performed in an environment where there is a high concentration of particles [102]. What is of great concern is the report of a faster than normal decline in FEV1 in young female skaters training in facilities where the ice resurfacing machines are driven by diesel fuel [103].

Cycling and asthma

Cyclists are repeatedly exposed to irritants, as well as allergens, and this group had the highest frequency of recorded use of asthma medication (50%) amongst 1996 USA summer Olympians [95]. Road dust and diesel exhaust are two important trigger factors for airway inflammation, and motor vehicle emissions, especially from diesel engines, are a major source of airborne pollutants. The combustion of fossil fuels produces a number of unhealthy substances, including carbon monoxide, nitrogen oxides, benzene, sulphur dioxides and particulate matter [102, 104]. Experimental studies exposing healthy humans to diesel exhaust particles (DEP), in concentrations that can be expected in daily life situations, has shown increased airway resistance and increased numbers of inflammatory cells, mainly lymphocytes and neutrophils, in the airways. Similar exposure of mild asthmatics also induced increased airway resistance. Exposure to DEP has also been shown to increase responsiveness to methacholine and to increase the IL-6 concentration in sputum [105]. In addition, DEPs can interact with allergen to augment allergen-induced responses, so that allergen-specific IgE levels are up to 50-fold greater in allergic subjects challenged with DEPs plus allergen than in those receiving allergen alone. There is also evidence that DEP exposure can drive the allergen-induced response towards the T-helper cell type-2 pathway, possibly through direct stimulation of mast cells and basophils in the airways [106, 107].

Summary

Development of bronchial hyperresponsiveness (BHR) is complex, although there are common risk factors for all athletes. These include: 1) effort-induced immunosuppression with increased vulnerability to respiratory tract infections and 2) exercise-induced hyperpnoea causing the airways to be exposed to higher than normal levels of allergens, fine particles and gases, and to be subjected to dehydration stress from conditioning of large volumes of cold and dry air.

Whilst exposure to airborne allergens is important in cyclists and runners, it is the irritants and gases that are important in swimmers and skaters. The bronchoscopy findings in skiers suggest that airway injury can occur simply from the dehydration stress. One potential outcome of dehydration stress is exudation of bulk plasma to restore the airway surface liquid. If the smooth muscle is repeatedly exposed to plasma products that have the potential to alter its contractile properties, then it is likely to become more sensitive to circulating mediators, such as leukotrienes and prostaglandins. In a winter athlete, this could lead to nonspecific BHR and in an atopic athlete the smooth muscle could become passively sensitised and develop BHR to allergens. Nonspecific BHR in athletes should not be necessarily interpreted as an indicator of asthma. Skiers and skaters have failed to benefit from treatment with either inhaled corticosteroids or leukotriene antagonists. Other strategies are required and these should include a reduction in environmental levels of potentially offending agents *e.g.* chloride content in swimming pools, fine particles and nitrogen dioxide in ice hockey halls.

Winter athletes may benefit from using heat-exchange devices and summer athletes from masks that capture allergens. Due to the increased vulnerability to respiratory tract infections, hard training and competition in close relation to a recent upper respiratory tract infectious episode should be discouraged. These interventions may lead to a reduction in the prevalence of BHR in elite athletes.

Keywords: Aeroallergens, bronchial hyperresponsiveness, fine particles, gases, infection, injury.

References

1. Joos GF, O'Connor B, Anderson SD, *et al.* ERS Task Force. Indirect airway challenges. *Eur Respir J* 2003; 21: 1050–1068.
2. Anderson SD, Kippelen P. Exercise-induced bronchoconstriction: pathogenesis. *Curr Allergy Asthma Rep* 2005; 5: 116–122.
3. Woolcock AJ, Peat JK, Salome CM, *et al.* Prevalence of bronchial hyperresponsiveness and asthma in a rural adult population. *Thorax* 1987; 42: 361–368.
4. Pattemore PK, Asher MI, Harrison AC, Mitchell EA, Rea HH, Stewart AW. The interrelationship among bronchial hyperresponsiveness, the diagnosis of asthma, and asthma symptoms. *Am Rev Respir Dis* 1990; 142: 549–554.
5. Sciurba FC. Physiologic similarities and differences between COPD and asthma. *Chest* 2004; 126: 117S–124S; discussion 159S–161S.
6. Shorr AF, Torrington KG, Hnatiuk OW. Endobronchial involvement and airway hyperreactivity in patients with sarcoidosis. *Chest* 2001; 120: 881–886.
7. Ludviksdottir D, Janson C, Bjornsson E, *et al.* Different airway responsiveness profiles in atopic

asthma, nonatopic asthma, and Sjogren's syndrome. BHR Study Group. Bronchial hyperresponsiveness. *Allergy* 2000; 55: 259–265.
8. Hassan WU, Keaney NP, Holland CD, Kelly CA. Bronchial reactivity and airflow obstruction in rheumatoid arthritis. *Ann Rheum Dis* 1994; 53: 511–514.
9. Cavigioli G, Mastropasqua B, Pelucchi A, Marazzini L, Foresi A. Reproducibility of allergen-induced asthma and associated increase in bronchial responsiveness to methacholine in asthmatic children. *Ann Allergy* 1993; 70: 411–417.
10. Louis R, Sele J, Henket M, *et al.* Sputum eosinophil count in a large population of patients with mild to moderate steroid-naive asthma: distribution and relationship with methacholine bronchial hyperresponsiveness. *Allergy* 2002; 57: 907–912.
11. Juniper EF, Kline PA, Vanzieleghem MA, Ramsdale EH, O'Byrne PM, Hargreave F. Effect of long-term treatment with inhaled corticosteroids on airway hyperresponsiveness and clinical asthma in non-steroid dependent asthmatics. *Am Rev Respir Dis* 1990; 142: 832–836.
12. Rodwell LT, Anderson SD, Seale JP. Inhaled steroids modify bronchial responses to hyperosmolar saline. *Eur Respir J* 1992; 5: 953–962.
13. Brannan JD, Koskela H, Anderson SD, Chan H-K. Budesonide reduces sensitivity and reactivity to inhaled mannitol in asthmatic subjects. *Respirology* 2002; 7: 37–44.
14. Sont JK, Willems LN, Bel EH, van Krieken JH, Vandenbroucke JP, Sterk PJ. Clinical control and histopathologic outcome of asthma when using airway hyperresponsiveness as an additional guide to long-term treatment. *Am J Respir Crit Care Med* 1999; 159: 1043–1051.
15. Lundgren R, Söderberg M, Hörstedt P, Stenling R. Morphological studies of bronchial mucosal biopsies from asthmatics before and after ten years of treatment with inhaled steroids. *Eur Respir J* 1988; 1: 883–889.
16. Wagner EM, Bleecker ER, Permutt S, Liu MC. Direct assessment of small airways reactivity in human subjects. *Am J Respir Crit Care Med* 1998; 157: 447–452.
17. Johnson PR, Burgess JK, Underwood PA, *et al.* Extracellular matrix proteins modulate asthmatic airway smooth muscle cell proliferation *via* an autocrine mechanism. *J Allergy Clin Immunol* 2004; 113: 690–696.
18. Johnson PR, Burgess JK. Airway smooth muscle and fibroblasts in the pathogenesis of asthma. *Curr Allergy Asthma Rep* 2004; 4: 102–108.
19. Amin K, Ludviksdottir D, Janson C, *et al.* Inflammation and structural changes in the airways of patients with atopic and nonatopic asthma. BHR Group. *Am J Respir Crit Care Med* 2000; 162: 2295–2301.
20. Koyama H, Nishimura K, Ikeda A, Sakai N, Mishima M, Izumi T. Influence of baseline airway calibre and pulmonary emphysema on bronchial responsiveness in patients with chronic obstructive pulmonary disease. *Respir Med* 1996; 90: 323–328.
21. Ramsdale EH, Morris MM, Roberts RS, Hargreave FE. Bronchial responsiveness to methacholine in chronic bronchitis: relationship to airflow limitation and cold air responsiveness. *Thorax* 1984; 39: 912–918.
22. Parker AL, Abu-Hijleh M, McCool FD. Ratio between forced expiratory flow between 25% and 75% of vital capacity and FVC is a determinant of airway reactivity and sensitivity to methacholine. *Chest* 2003; 124: 63–69.
23. Ownby DR, Peterson EL, Johnson CC. Factors related to methacholine airway responsiveness in children. *Am J Respir Crit Care Med* 2000; 161: 1578–1583.
24. Rundell KW, Spiering BA, Evans TM, Baumann JM. Baseline lung function, exercise-induced bronchoconstriction, and asthma-like symptoms in elite women ice hockey players. *Med Sci Sports Exerc* 2004; 36: 405–410.
25. Dickinson JW, Whyte GP, McConnell AK, Harries M. The role of FEF50 in the diagnosis of exercise-induced asthma in elite athletes. *Proc Am Thorac Soc* 2005; 2 (abstract issue): A774.
26. Wassmer G, Jorres RA, Heinrich J, Wjst M, Reitmeir P, Wichmann HE. The association between baseline lung function and bronchial responsiveness to methacholine. *Eur J Med Res* 1997; 2: 47–54.

27. Henriksen AH, Holmen TL, Bjermer L. Gender differences in asthma prevalence may depend on how asthma is defined. *Respir Med* 2003; 97: 491–497.
28. Holzer K, Anderson SD, Douglass J. Exercise in elite summer athletes: Challenges for diagnosis. *J Allergy Clin Immunol* 2002; 110: 374–380.
29. Pauwels R, Joos G, Van Der Straten M. Bronchial hyperresponsiveness is not hyperresponsiveness is not bronchial asthma. *Clin Allergy* 1988; 18: 317–321.
30. Smith CM, Anderson SD. Inhalational challenge using hypertonic saline in asthmatic subjects: a comparison with responses to hyperpnoea, methacholine and water. *Eur Respir J* 1990; 3: 144–151.
31. Anderson SD, Brannan JD, Leuppi JD, Koskela H. Monitoring airway hyper-responsiveness: Indirect stimuli-exercise, hypertonic saline mannitol and adenosine monophosphate. *In*: Gibson PG, ed. Monitoring Asthma. Boca Raton, Francis & Taylor, 2005; pp. 275–323.
32. Anderson SD. Single dose agents in the prevention of exercise-induced asthma. A descriptive review. *Treat Respir Med* 2004; 3: 365–379.
33. Leuppi JD, Brannan JD, Anderson SD. Bronchial provocations tests: The rationale for using inhaled mannitol as a test for airway hyperresponsiveness. *Swiss Med Wkly* 2002; 132: 151–158.
34. Belcher NG, Rees PJ, Clark TJM, Lee TH. A comparison of the refactory periods induced by hypertonic airway challenge and exercise in bronchial asthma. *Am Rev Respir Dis* 1987; 135: 822–825.
35. Mannix ET, Roberts MA, Dukes HJ, Magnes CJ, Farber MO. Airways hyperresponsiveness in high school athletes. *J Asthma* 2004; 41: 567–574.
36. Rundell KW, Jenkinson DM. Exercise-induced bronchospasm in the elite athlete. *Sports Med* 2002; 32: 583–600.
37. Langdeau J-B, Turcotte H, Bowie DM, Jobin J, Desgagné P, Boulet L-P. Airway hyperresponsiveness in elite athletes. *Am J Respir Crit Care Med* 2000; 161: 1479–1484.
38. Nieman DC, Johanssen LM, Lee JW, Arabatzis K. Infectious episodes in runners before and after the Los Angeles Marathon. *J Sports Med Phys Fitness* 1990; 30: 316–328.
39. Nieman DC. Special feature for the Olympics: effects of exercise on the immune system: exercise effects on systemic immunity. *Immunol Cell Biol* 2000; 78: 496–501.
40. Nieman DC, Simandle S, Henson DA, *et al.* Lymphocyte proliferative response to 2.5 hours of running. *Int J Sports Med* 1995; 16: 404–409.
41. Bruunsgaard H, Hartkopp A, Mohr T, *et al. In vivo* cell-mediated immunity and vaccination response following prolonged, intense exercise. *Med Sci Sports Exerc* 1997; 29: 1176–1181.
42. Nieman DC, Fagoaga OR, Butterworth DE, *et al.* Carbohydrate supplementation affects blood granulocyte and monocyte trafficking but not function after 2.5 h or running. *Am J Clin Nutr* 1997; 66: 153–159.
43. Nieman DC, Henson DA, Garner EB, *et al.* Carbohydrate affects natural killer cell redistribution but not activity after running. *Med Sci Sports Exerc* 1997; 29: 1318–1324.
44. Müns G, Liesen H, Riedel H, Bergmann K-C. EinfluB von langstreckenlauf auf den IgA-gehalt in nasensekret und speichel. [Influence of marathon running on IgA-levels in nasal and mouth secretion]. *Deut Zeit Sportmed* 1989; 40: 63–65.
45. Steerenberg PA, van Asperen IA, van Nieuw Amerongen A, Biewenga A, Mol D, Medema GJ. Salivary levels of immunoglobulin A in triathletes. *Eur J Oral Sci* 1997; 105: 305–309.
46. Gleeson M, Hall ST, McDonald WA, Flanagan AJ, Clancy RL. Salivary IgA subclasses and infection risk in elite swimmers. *Immunol Cell Biol* 1999; 77: 351–355.
47. Muns G, Singer P, Wolf F, Rubinstein I. Impaired nasal mucociliary clearance in long-distance runners. *Int J Sports Med* 1995; 16: 209–213.
48. Nieman DC, Henson DA, Fagoaga OR, *et al.* Change in salivary IgA following a competitive marathon race. *Int J Sports Med* 2002; 23: 69–75.
49. Sue-Chu M, Karjalainen E-M, Altraja A, *et al.* Lymphoid aggregates in endobronchial biopsies from young elite cross-country skiers. *Am J Respir Crit Care Med* 1998; 158: 597–601.
50. Tschernig T, Pabst R. Bronchus-associated lymphoid tissue (BALT) is not present in the normal adult lung but in different diseases. *Pathobiology* 2000; 68: 1–8.

51. Delventhal S, Hensel A, Petzoldt K, Pabst R. Effects of microbial stimulation on the number, size and activity of bronchus-associated lymphoid tissue (BALT) structures in the pig. *Int J Exp Pathol* 1992; 73: 351–357.
52. Heir T, Aanestad G, Carlsen K-H, Larsen S. Respiratory tract infection and bronchial responsiveness in elite athletes and sedentary control subjects. *Scand J Med Sci Sports* 1995; 5: 94–99.
53. Davis MS, Freed AN. Repeated dry air hyperventilation causes airway inflammation, edema, and mucosal squamous metaplasia. *Am J Respir Crit Care Med* 1998; 157: A672.
54. Kaminsky DA, Bates JH, Irvin CG. Effects of cool, dry air stimulation on peripheral lung mechanics in asthma. *Am J Respir Crit Care Med* 2000; 162: 179–186.
55. Anderson SD, Daviskas E. The mechanism of exercise-induced asthma is ……. *J Allergy Clin Immunol* 2000; 106: 453–459.
56. Hallstrand TS, Moody MW, Wurfel MM, Schwartz LB, Henderson WR, Aitken ML. Inflammatory basis of exercise-induced bronchoconstriction. *Am J Respir Crit Care Med* 2005; 172: 679–686.
57. Mickleborough TD, Murray RL, Ionescu AA, Lindley MR. Fish oil supplementation reduces severity of exercise-induced bronchoconstriction in elite athletes. *Am J Respir Crit Care Med* 2003; 168: 1181–1189.
58. Caillaud C, Le Creff C, Legros P, Denjean A. Strenuous exercise increases plasmatic and urinary leukotriene E4 in cyclists. *Can J Appl Physiol* 2003; 28: 793–806.
59. Kikawa Y, Hosoi S, Inoue Y, *et al.* Exercise-induced urinary excretion of leukotriene E4 in children with atopic asthma. *Pediatr Res* 1991; 29: 455–459.
60. Pliss LB, Ingenito EP, Ingram RH, Pichurko B. Assessment of bronchoalveolar cell and mediator response to isocapnic hyperpnea in asthma. *Am Rev Respir Dis* 1990; 142: 73–78.
61. Leff JA, Busse WW, Pearlman D, *et al.* Montelukast, a leukotriene-receptor antagonist, for the treatment of mild asthma and exercise-induced bronchoconstriction. *N Engl J Med* 1998; 339: 147–152.
62. O'Sullivan S, Roquet A, Dahlén B, Dahlén S-E, Kumlin M. Urinary excretion of inflammatory mediators during allergen-induced early and late phase asthmatic reactions. *Clin Exp Allergy* 1998; 228: 1332–1339.
63. Finnerty JP, Holgate ST. Evidence for the roles of histamine and prostaglandins as mediators in exercise-induced asthma: the inhibitory effect of terfenadine and flurbiprofen alone and in combination. *Eur Respir J* 1990; 3: 540–547.
64. Shimizu T, Mochizuki H, Shigeta M, Morikawa A. Effect of inhaled indomethacin on exercise-induced bronchoconstriction in children with asthma. *Am J Respir Crit Care Med* 1997; 155: 170–173.
65. Ishii Y, Kitamura S. Hyperventilation stimulates the release of Prostaglandin I2 and E2 from lung in humans. *Prostaglandins* 1990; 39: 685–691.
66. Hashimoto S, Matsumoto K, Gon Y, Nakayama T, Takeshita I, Horie T. Hyperosmolarity-induced interleukin-8 expression in human bronchial epithelial cells through p38 mitogen-activated protein kinase. *Am J Respir Crit Care Med* 1999; 159: 634–640.
67. Lee TH, Nagakura T, Cromwell O, Brown MJ, Causon R, Kay AB. Neutrophil chemotactic activity and histamine in atopic and nonatopic subjects after exercise-induced asthma. *Am Rev Respir Dis* 1984; 129: 409–412.
68. Brightling CE, Bradding P, Symon FA, Holgate ST, Wardlaw AJ, Pavord ID. Mast cell infiltration of airway smooth muscle in asthma. *N Engl J Med* 2002; 346: 1699–1705.
69. Carroll NG, Mutavdzic S, James AL. Distribution and degranulation of airway mast cells in normal and asthmatic subjects. *Eur Respir J* 2002; 19: 879–885.
70. Brannan JD, Gulliksson M, Anderson SD, Chew N, Kumlin M. Evidence of mast cell activation and leukotriene release after mannitol inhalation. *Eur Respir J* 2003; 22: 491–496.
71. Karjalainen E-M, Laitinen A, Sue-Chu M, Altraja A, Bjermer L, Laitinen LA. Evidence of airway

inflammation and remodeling in ski athletes with and without bronchial hyperresponsiveness to methacholine. *Am J Respir Crit Care Med* 2000; 161: 2086–2091.
72. Sue-Chu M, Larsson L, Moen T, Rennard SI, Bjermer L. Bronchoscopy and bronchoalveolar lavage findings in cross-country skiers with and without "ski asthma". *Eur Respir J* 1999; 13: 626–632.
73. Sue-Chu M, Brannan JD, Anderson SD, Chew N, Bjermer L. Airway responsiveness to methacholine (Mch), adenosine 5-monophosphate (AMP), mannitol (Man), eucapnic voluntary hyperpnea (EVH) and sport specific field exercise challenge (Ex) in cross country ski athletes. *Eur Respir J* 2002; 20: Suppl. 38, 410s.
74. Sue-Chu M, Henriksen AH, Bjermer L. Non-invasive evaluation of lower airway inflammation in hyper-responsive elite cross-country skiers and asthmatics. *Respir Med* 1999; 93: 719–725.
75. Sue-Chu M, Karjalainen E-M, Laitinen A, Larsson L, Laitinen LA, Bjermer L. Placebo-controlled study of inhaled budesonide on indices of airways inflammation in bronchoalveolar lavage fluid and bronchial biopsies in cross country skiers. *Respiration* 2000; 67: 417–425.
76. Heir T, Larsen S. The influence of training intensity, airway infections and environmental conditions on seasonal variations in bronchial responsiveness in cross-country skiers. *Scand J Med Sci Sports* 1995; 5: 152–159.
77. Helenius I, Rytilä P, Sarna S, *et al.* Effect of continuing or finishing high-level sports on airway inflammation, bronchial hyperresponsiveness, and asthma: A 5-year prospective follow-up study of 42 highly trained swimmers. *J Allergy Clin Immunol* 2002; 109: 962–968.
78. Pisarri TE, Jonson A, Coleridge HM, Coleridge JCG. Intravenous injection of hypertonic NaCl solutions stimulates pulmonary C-fibres in dogs. *Am J Physiol* 1991; 260: Suppl. 5 Pt 2, H1522–H1530.
79. Sont JK, Booms P, Bel EH, Vandenbroucke JP, Sterk PJ. The severity of breathlessness during challenges with inhaled methacholine and hypertonic saline in atopic asthmatic subjects. The relationship with deep breath-induced bronchodilation. *Am J Respir Crit Care Med* 1995; 152: 38–44.
80. Polosa R, Ciamarra I, Mangano G, *et al.* Bronchial hyperresponsiveness and airway inflammation markers in nonasthmatics with allergic rhinitis. *Eur Respir J* 2000; 15: 30–35.
81. Katelaris CH, Carrozzi FM, Burke TV, Byth K. A springtime olympics demands special consideration for allergic athletes. *J Allergy Clin Immunol* 2000; 106: 260–266.
82. Helenius IJ, Tikkanen HO, Sarna S, Haahtela T. Asthma and increased bronchial responsiveness in elite athletes: atopy and sport event as risk factors. *J Allergy Clin Immunol* 1998; 101: 646–652.
83. Sue-Chu M, Larsson L, Bjermer L. Prevalence of asthma in young cross-country skiers in central Scandinavia: differences between Norway and Sweden. *Respir Med* 1996; 90: 99–105.
84. Helenius I, Haahtela T. Allergy and asthma in elite summer sport athletes. *J Allergy Clin Immunol* 2000; 106: 444–452.
85. Zwick H, Popp W, Budik G, Wanke T, Rauscher H. Increased sensitization to aeroallergens in competitive swimmers. *Lung* 1990; 168: 111–115.
86. Potts JE. Adverse respiratory health effects of competetive swimmers: the prevalence of symptoms, illnesses, and bronchial responsiveness to methacholine and exercise. MD Thesis 1994, University of British Columbia, Canada.
87. Potts J. Factors associated with respiratory problems in swimmers. *Sports Med* 1996; 21: 256–261.
88. Turcotte H, Langdeau JB, Bowie DM, Boulet LP. Are questionnaires on respiratory symptoms reliable predictors of airway hyperresponsiveness in athletes and sedentary subjects? *J Asthma* 2003; 40: 71–80.
89. Helenius IJ, Rytilä P, Metso T, Haahtela T, Venge P, Tikkanen HO. Respiratory symptoms, bronchial responsiveness, and cellular characteristics of induced sputum in elite swimmers. *Allergy* 1998; 53: 346–352.
90. Drobnic F, Freixa A, Casan P, Sanchis J, Guardino X. Assessment of chlorine exposure in swimmers during training. *Med Sci Sports Exerc* 1996; 28: 271–274.

91. Thickett KM, McCoach JS, Gerber JM, Sadhra S, Burge PS. Occupational asthma caused by chloramines in indoor swimming-pool air. *Eur Respir J* 2002; 19: 827–832.
92. Sengler C, Heinzmann A, Jerkic SP, *et al.* Clara cell protein 16 (CC16) gene polymorphism influences the degree of airway responsiveness in asthmatic children. *J Allergy Clin Immunol* 2003; 111: 515–519.
93. Lensmar C, Nord M, Gudmundsson GH, *et al.* Decreased pulmonary levels of the anti-inflammatory Clara cell 16 kDa protein after induction of airway inflammation in asthmatics. *Cell Mol Life Sci* 2000; 57: 976–981.
94. Lagerkvist BJ, Bernard A, Blomberg A, *et al.* Pulmonary epithelial integrity in children: relationship to ambient ozone exposure and swimming pool attendance. *Environ Health Perspect* 2004; 112: 1768–1771.
95. Weiler J, Layton T, Hunt M. Asthma in United States Olympic athletes who participated in the 1996 Summer Games. *J Allergy Clin Immunol* 1998; 102: 722–726.
96. Anderson SD, Fitch K, Perry CP, *et al.* Responses to bronchial challenge submitted for approval to use inhaled beta2 agonists prior to an event at the 2002 Winter Olympics. *J Allergy Clin Immunol* 2003; 111: 44–49.
97. Wilber RL, Rundell L, Szmedra L, Jenkinson DM, Im J, Drake SD. Incidence of exercise-induced bronchospasm in Olympic Winter Sport athletes. *Med Sci Sports Exerc* 2000; 32: 732–737.
98. Lumme A, Haahtela T, Öunap J, *et al.* Airway inflammation, bronchial hyperresponsiveness, and asthma in elite ice hockey players. *Eur Respir J* 2003; 22: 113–117.
99. Leuppi JD, Kuln M, Comminot C, Reinhart WH. High prevalence of bronchial hyperresponsiveness and asthma in ice hockey players. *Eur Respir J* 1998; 12: 13–16.
100. Levy JI, Lee K, Yanagisawa Y, Hutchinson P, Spengler JD. Determinants of nitrogen dioxide concentrations in indoor ice skating rinks. *Am J Public Health* 1998; 88: 1781–1786.
101. Sandstrom T, Stjernberg N, Eklund A, *et al.* Inflammatory cell response in bronchoalveolar lavage fluid after nitrogen dioxide exposure of healthy subjects: a dose-response study. *Eur Respir J* 1991; 4: 332–339.
102. Rundell KW. High levels of airborne ultrafine and fine particulate matter in indoor ice arenas. *Inhal Toxicol* 2003; 15: 237–250.
103. Rundell K. Pulmonary function decay in women ice hockey players: is there a relationship to ice rink air quality? *Inhal Toxicol* 2004; 16: 117–123.
104. Riedl M, Diaz-Sanchez D. Biology of diesel exhaust effects on respiratory function. *J Allergy Clin Immunol* 2005; 115: 221–228; quiz 229.
105. Nordenhall C, Pourazar J, Ledin MC, Levin JO, Sandstrom T, Adelroth E. Diesel exhaust enhances airway responsiveness in asthmatic subjects. *Eur Respir J* 2001; 17: 909–915.
106. Devouassoux G, Saxon A, Metcalfe DD, *et al.* Chemical constituents of diesel exhaust particles induce IL-4 production and histamine release by human basophils. *J Allergy Clin Immunol* 2002; 109: 847–853.
107. Nemmar A, Hoet PH, Vermylen J, Nemery B, Hoylaerts MF. Pharmacological stabilization of mast cells abrogates late thrombotic events induced by diesel exhaust particles in hamsters. *Circulation* 2004; 110: 1670–1677.