

## 3rd International Immunonutrition Workshop

### Session 6: Role of physical activity on immune function Physical activity, immunity and infection

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During the last few decades, scientific evidence has confirmed a wide range of health benefits related to regular physical activity. How physical activity affects the immune function and infection risk is, however, still under debate. Commonly, intensive exercise suppresses the activity and levels of several immune cells, while other immune functions may be stimulated by moderate physical activity. With this knowledge, the understanding of the relationship between different levels of physical activity on the immune function has been raised as a potential tool to protect health not only in athletes but also in the general population; the mechanisms that translate a physically active lifestyle into good health continue to be investigated. Reviewing the literature, although several outcomes (i.e. the mechanisms by which different levels and duration of physical activity programmes affect numerous cell types and responses) remain unclear, given that the additional benefits encompass healthy habits including exercise, the use of physical activity programmes may result in improved health of elderly populations. Moderate physical activity or moderate-regulated training may enhance the immune function mainly in less fit subjects or sedentary population and the pre-event fitness status also seems to be an important individual factor regarding this relationship. Although adequate nutrition and regular physical activity habits may synergistically improve health, clinical trials in athletes using nutritional supplements to counteract the immune suppression have been inconclusive so far.

Further research is necessary to find out to what extent physical activity training can exert an effect on the immune function.

#### Physical activity: Immune function: Infection: Nutrition

The term 'physical activity' is often used interchangeably with 'exercise' and although both have a number of common elements, it is important to point out the differences. While physical activity is defined as any body movement produced by skeletal muscular action that increases energy expenditure, exercise is planned, structured, and repetitive physical activity<sup>(1)</sup>. On the other hand, physical fitness is the capacity to perform physical activity, and refers to a full range of physiological and psychological qualities<sup>(1)</sup>. The intensity of physical activity may be classified using a standardized classification based on the energy costs of specific physical activities expressed as metabolic-equivalent scores (where 1 metabolic-equivalent is the energy

needed per kg of body weight per hour of activity divided by the energy needed per kg per hour at rest)<sup>(2)</sup>. Normally, physical activity levels are classified as not active, light, or moderate intensity (metabolic-equivalent scores <6) and vigorous intensity (metabolic-equivalent scores ≥6).

The worldwide beneficial effects of a regular moderate physical active lifestyle on a number of physiological processes (cardiovascular function, insulin sensitivity, dyslipidemia, visceral adiposity and hypertension) are well documented<sup>(3–7)</sup>. While observational studies have pointed out that light-to-moderate physical activity is associated with a reduction of total mortality risk<sup>(8)</sup>, early sedentary lifestyle is considered to be an independent risk factor for

**Abbreviations:** CHO, carbohydrate; CRP, C-reactive protein; Th, T helper; URTI, upper respiratory tract infection.

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**Table 1.** Main effects of acute *v.* moderate exercise on both innate and adaptive immunity

|   | Innate immunity  | Adaptive immunity (cellular immunity)  |
|---|--|--|
| Acute exercise (90 min or longer)             | <ul style="list-style-type: none"> <li>↑ Number of circulating NK cells<sup>(11)</sup></li> <li>↓ NK cell cytotoxic activity and NK cells counts after exercise<sup>(28)</sup></li> <li>↑ Number of circulating neutrophils and monocytes<sup>(52)</sup></li> <li>↑ Monocyte and neutrophil phagocytosis<sup>(38)</sup></li> <li>↓ Nasal neutrophil phagocytic capacity (transient suppression)<sup>(27)</sup></li> <li>↓ Expression of toll-like receptors<sup>(35)</sup></li> <li>↑ Cortisol production</li> <li>↑ CRP level<sup>(35)</sup></li> </ul> | <ul style="list-style-type: none"> <li>↑ Number of circulating lymphocytes<sup>(35)</sup></li> <li>↓ Proliferative response of lymphocytes to mitogens<sup>(19)</sup></li> <li>↓ Number of circulating lymphocytes and T-lymphocyte</li> <li>↑ CD4+/CDB+ ratio after strenuous and prolonged exercise</li> <li>↑ Levels of pro-inflammatory cytokine IL-6<sup>(35,38)</sup></li> <li>↑ Levels of anti-inflammatory cytokines IL-10 und IL-1ra</li> <li>↓ Levels of TNF<math>\alpha</math></li> <li>Lipolysis induction</li> <li>↓ Th1/Th2 ratio<sup>(150)</sup></li> <li>↓ Nasal and salivary IgA level<sup>(27)</sup></li> <li>↓ Macrophage major histocompatibility complex class II expression<sup>(27,35,151)</sup></li> <li>↑ T-memory (CD45RO+) and T-naïve (CD45RA+) cell counts<sup>(154,156)</sup></li> <li>↑ CD45RO+/CD45RA+ ratio<sup>(154,156)</sup></li> <li>↓</li> <li>- Alteration of regulatory mechanism<sup>(155)</sup></li> <li>- Suppression of cellular immunity → 'open window'</li> </ul> |
| Moderate exercise (anti-inflammatory effects) | <ul style="list-style-type: none"> <li>↓ Levels of biomarkers that are used to assess systemic inflammation (mitogen-stimulated inflammatory cytokine production, skeletal muscle inflammatory protein content, adipokine production, serum levels at CRP)<sup>(35)</sup></li> </ul>   | <ul style="list-style-type: none"> <li>↓ Exercise-induced IL-6 mRNA response after 10 weeks of training (untrained young men)<sup>(157)</sup></li> <li>↓ Systemic concentrations of IL-6 in elderly people at risk for disability<sup>(158)</sup></li> <li>Not suppress the salivary IgA response or alter cortisol secretion<sup>(159)</sup></li> </ul>   |

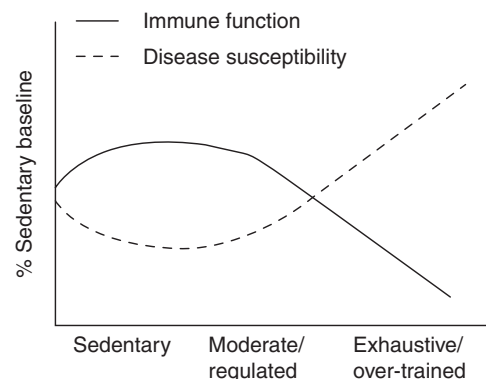
CRP, C-reactive protein; NK, natural killer; Th, T helper; ↑, increase; ↓, decrease.

several chronic diseases in adulthood<sup>(9)</sup>. Nowadays, physical activity therapy has received remarkable scientific attention for several reasons: effectiveness and logistical advantages over other therapies, relatively easy to implement, little cost or equipment and performance in either clinical or home settings<sup>(10)</sup>.

Physical activity has been shown to induce considerable physiological change on the immune system; however, their global effect on the immune function and their consequences on infection risk are still under debate.

Moreover, the mechanisms that explain a physically active lifestyle in terms of good health continue to be investigated<sup>(11)</sup>. Commonly, although intensive exercise, such as endurance training, could lead to the suppression of the activity and levels of several immune cells, other immune functions may be stimulated by moderate physical activity<sup>(12–15)</sup>.

This outcome has (at least partially) been explained by the relationship between physical activity and upper respiratory tract infections (URTI). While regular physical activity is inversely associated with URTI, several studies provide evidence supporting that athletes and sportsmen are at an increased risk of infections during periods of heavy training (the so-called 'open window' of impaired immunity)<sup>(16,17)</sup>. Several years ago, these findings suggested the 'Inverted J Hypothesis' in exercise immunology (Fig. 1), where disease susceptibility is increased in sedentary and over-trained subjects in comparison with regulated, moderate training<sup>(18)</sup>. There is no doubt that this hypothesis is influenced by several factors including the



**Fig. 1.** 'Inverted J Hypothesis' adapted from Woods<sup>(18)</sup>.

intensity, type and duration of physical activity<sup>(19,20)</sup>. In Table 1, the main effects of acute *v.* moderate exercise on both innate and adaptive immunity are summarized.

Since age-related decline in the immune function has been documented, the interest of the beneficial impact of a regular moderate physical activity on immune competence among the elderly has increased. On the other hand, there is a great interest in the possible effect of nutritional supplements on the incidence of infections after endurance training periods.

The present review summarizes evidence about the influence of physical activity on immunity and infection, taking into account ageing and nutritional aspects.

## Immunity in response to physical activity

### *Acute–chronic–intense physical activity*

Previous studies have reported an immune–physiologic adaptation to an acute bout of exercise leading to an increase of neutrophils together with a decrease of eosinophils, most probably influenced by plasma volume changes<sup>(21–25)</sup>. The possible mechanism seems to be associated with the migration of cells from endothelial tissues, as well as a phagocytic and inflammatory response to exercise-induced tissue damage<sup>(26)</sup>. Nevertheless, which different subsets of leucocytes migrate differentially into different tissues after exercise seems to be dependent on other factors such as the duration and intensity of the exercise<sup>(27)</sup>. A suppression of the respiratory burst and/or phagocytic capacity from neutrophils immediately after acute exercise has also been observed<sup>(26)</sup>.

Although the global health significance of transient changes in innate immunity cells such as natural killer cell subsets remains unclear<sup>(11)</sup>. After high-intensity exercise periods, a period of natural killer cell function depression has been described to be partly responsible of an increased susceptibility to the infection stage<sup>(28)</sup>. Some *in vitro* cross-sectional studies of the immune function have suggested enhanced natural killer cell cytotoxicity<sup>(29,30)</sup> and T-lymphocyte proliferation among highly trained athletes *v.* untrained subjects<sup>(19,21,22,31,32)</sup>. With regard to immune cell counts, the results are less consistent<sup>(33–35)</sup>.

Acute and chronic intense exercise are also known to impair cell-mediated immunity by decreasing the expression of toll-like receptors and increasing cortisol and IL-6 production, leading to a state of inflammation, via a decreased macrophage and T-helper (Th) 1 cell cytokine production<sup>(35)</sup>. Furthermore, intense training seems to be associated with an imbalanced immunity response by an enhanced release of pro-inflammatory cytokines such as TNF $\alpha$ , IL-1 $\beta$  and IL-6, followed by anti-inflammatory cytokines release, such as IL-10 and IL-1ra<sup>(36,37)</sup>, and also suppressing cellular immunity, leading to an increased infections risk susceptibility<sup>(38)</sup>.

A number of factors including exercise duration and intensity, and sex and age seem to influence the changes produced in the immune response and further studies are needed to identify additional moderating factors (e.g. training and nutritional status) to understand the mechanisms<sup>(11)</sup>.

*Moderate–regulated physical activity.* The immune adaptation to physical activity depends on the intensity, duration and type of exercise, as well as environment temperature, nutrition, and hydration status, body composition, and concentration of hormones and cytokines<sup>(41)</sup>.

Several studies have reported that moderate regular physical activity can enhance the immune response. Animal studies have shown that exercise training may have a beneficial effect in natural cytotoxicity and T-lymphocyte proliferation reducing stress-induced antibody formation<sup>(40)</sup>, as well as enhanced counts of T-cells, B-cells and Ig<sup>(41–44)</sup>. Thus, according to this outcome, if regular physical activity can enhance cell-mediated adaptive immune responses in young healthy animals, perhaps participation in regular physical activity would benefit other populations (i.e. the

elderly) whose immune system is compromised to a greater extent<sup>(45)</sup>.

Exercise training has recently been suggested to up-regulate Th-cell-mediated immune functions and be helpful to reduce infection risk and autoimmune diseases in elderly people<sup>(46)</sup>. Moreover, regular moderate-intensity endurance exercise has shown to increase lymphocyte proliferation, IL-2 production and T-cell subsets in this population<sup>(47,48)</sup>.

Regarding inflammation, while both moderate and intense exercise stimulate the phagocytic process of microbial, bacterial and viral pathogens in sedentary women, the profile of pro-/anti-inflammatory cytokine release seems to be enhanced following the moderate exercise<sup>(49)</sup>. Exercise has been shown to produce a short-term, inflammatory response with increased acute phase reactants, whereas both cross-sectional comparisons and longitudinal exercise training studies demonstrate a long-term ‘anti-inflammatory’ effect<sup>(50)</sup>. The attenuated lowered expression of some toll-like receptors seems to offer a new mechanism to explain some of the effects of the health benefits of an active lifestyle by reducing cardiovascular and metabolic disease risk which are commonly associated with elevated levels of inflammation markers including cytokines<sup>(35)</sup>.

On the other hand, cross-sectional and longitudinal studies have shown that athletes show a higher incidence of illness than ‘amateur’ sportsmen or sedentary individuals<sup>(51)</sup>, although the results are still controversial<sup>(52,53)</sup>, and some explanation for this conflicting point of view may be related with an inadequate recovery or prior history of illness<sup>(51,54)</sup> or by individual determinants such as physical training level, age, nutritional status, ageing and health status.

## Physical activity, immunity and ageing

### *Immune-senescence*

Ageing is a continuous multi-dimensional process of physiological changes including several immunological alterations characterized by increased susceptibility to infectious and autoimmune diseases that are collectively called immune-senescence<sup>(55,56)</sup>. Older adults have been shown to exhibit an increased incidence of infections as compared with young or middle-aged adults<sup>(39, 57–59)</sup>. This clinical evidence enhances morbidity and mortality risk in this population group. Alternatively, the elderly population also frequently presents a higher risk of developing nutritional disorders caused by the ageing process, which may modify the dietary habits as well as physical activity habits<sup>(60–62)</sup>.

With regard to immune alterations associated with increased infectious risk, ageing is associated with declines in T-cell subset counts as well as their function<sup>(63–65)</sup>. The factors leading to this T-cell imbalance include a decrease in naive T-cells or Th0 cells<sup>(64–68)</sup>, age-associated changes in surface molecule expression<sup>(69,70)</sup>, alterations in intracellular signalling<sup>(71,72)</sup>, increased rates of apoptosis<sup>(73,74)</sup> and decreased proliferative capacity<sup>(75,76)</sup>.

Ageing has also been associated with alterations in the Th1–Th2 balance<sup>(19)</sup>, but with contradictory outcomes. While some studies have documented an age-associated

shift towards Th2 cytokine as IL-4, IL-6 and IL-10 production in human subjects<sup>(77,78)</sup>, others have reported a trend towards increased Th1 (IL-2 and interferon- $\gamma$ )<sup>(79,80)</sup>. These conflicting findings are probably due to experimental factors such as techniques and *in vitro* stimulus used<sup>(81)</sup>.

A decrease in B-cell numbers and function has also been observed in the elderly population<sup>(82–84)</sup>. An incomplete stimulation from other immune cells and a diminished differentiation capacity has been suggested as possible biological mechanisms to explain the age-related immunodepression<sup>(85–87)</sup>.

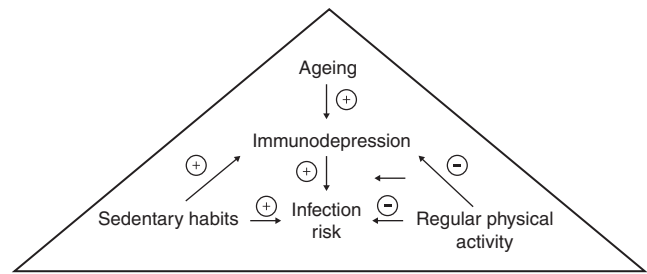
#### *Physical activity counteracting age-induced immunodepression*

Since the decline in immune response with age is well documented, the interest of the impact of physical activity on immune competence among the elderly has increased. On the other hand, sedentary habits seem to be associated with increased infection risk in ageing. Leveille *et al.* observed that physical inactivity in women aged 55–80 (followed for a period of 6 years) was associated with an increased risk of hospitalization largely due to infections<sup>(88)</sup>.

Other studies addressing the impact of physical activity in elderly populations have suggested that moderate exercise may counteract the effects of immune senescence<sup>(39, 89–91)</sup>. Some studies have also suggested that exercise may reduce low-grade inflammation by reduced levels of C-reactive protein (CRP)<sup>(92)</sup>. A decrease in lipopolysaccharide-stimulated IL-6, IL-1 $\beta$  and TNF $\alpha$  production in the active elderly have also been shown when compared with the sedentary elderly<sup>(92)</sup>. According to a retrospective and prospective study carried out to examine the relationship between physical activity and URTI in elderly subjects (aged 66–84 years), the greatest protection from URTI seems to be associated with a specific level of physical activity (equivalent to jogging 7 km/d or walking 90 min/d at a speed of about 19–20 min/mile)<sup>(93)</sup>.

Besides, the duration of the intervention studies seems to be a determining factor to debate about the final outcome. While a 10-week moderate-to-vigorous exercise trial was not associated with decreased URTI in older women<sup>(94)</sup>, longer intervention trials (10 months) have reported a decrease of URTI symptoms after regular moderate intensity exercise intervention<sup>(91)</sup>. These different effects could be partially due to the relatively small number of subjects and the short exercise intervention of the former study<sup>(94)</sup>.

The intensity of the activity is also important. Moderate exercise training has been associated with the improvement of expression of CD28 on Th cells and Th1–Th2 balances in the elderly. Exercise training could up-regulate Th cell-mediated immune functions and be helpful for a decrease in the risk of infections and autoimmune diseases in elderly people<sup>(46)</sup>. Nevertheless, a 12-month moderate resistance training programme has been reported to increase the muscle strength of previously sedentary, clinically healthy, elderly women, although no changes were found on immune phenotypic or functional parameters, such as natural killer cell cytotoxic activity, lymphoproliferative response to the mitogen phytohemagglutinin, CD3, CD4, CD8,



**Fig. 2.** Proposed model of the relationships between ageing, infection risk and physical activity.

CD19, CD56 cell counts, as well as CD25, CD28, CD45RA, CD45RO, CD69, CD95, human leucocyte antigen (HLA)-DR cellular expression<sup>(95)</sup>.

Finally, it seems important to highlight that the combination of physical activity programmes added to diet interventions may be promising. Several studies on middle-aged adults have found that physical activity and diet interventions resulting in weight loss reduced serum CRP, IL-6 and IL-18, whereas exercise training alone reduced serum CRP, IL-6, and blood mononuclear cell production of TNF $\alpha$  and IL-1 $\alpha$  in patients at risk for heart disease<sup>(95–98)</sup>.

The mechanisms by which different levels and duration of physical activity programmes affect numerous cell types and responses remain unclear. But there is enough evidence supporting the regular physical activity benefits counteracting the immune-senescence and the infection risk of elderly populations (Fig. 2).

Further studies of how physical activity impacts on immunosenescence in older adults are warranted and very promising in order to improve the healthcare of geriatric populations.

#### **Nutritional countermeasures of immunodepression**

Nutrition and physical activity have strong, simultaneous and continuous influences on health and disease<sup>(99–101)</sup>. Numerous nutrients are known to be involved in energy metabolism and protein synthesis modulating the immune function. Some potential mechanisms of this modulation include the ability of the immune cells to proliferate and differentiate, the production of proteins with specific immune-related functions, phagocytic activity and immune cell-mediated immunity<sup>(102)</sup>.

The concept of 'immunonutrition' refers to the ability of certain nutrients to modulate the immune status if they are provided in sufficient amounts, in general, far more often in excess of the usual nutritional requirements<sup>(17,103)</sup>. Lacking adequate nutrition, the immune system is undoubtedly deprived of the components needed to generate an effective immune response<sup>(104)</sup>. Vitamins (A,  $\beta$ -carotene, folic acid, B6, B12, C, E and riboflavin) and minerals (Fe, Zn and Se) play a pivotal role in maintaining the antioxidant/oxidant balance in immune cells and in protecting them from oxidative stress and preserving their adequate function<sup>(104–107)</sup>.



### Endurance athletes

Considerable evidence indicates that inadequate nutrition and psychological stress increase the negative effects of exertion (i.e. exercise periods) upon the immune system<sup>(104)</sup>. A number of non-systematic review articles have recommended that athletes consume a balanced diet ensuring energy and nutrient requirements<sup>(108,109)</sup>. In this sense, several studies have evaluated the efficacy of nutritional supplements on the incidence infection risk after exercise periods. We briefly describe the main studies reporting effects on nutritional supplements used in physical activity performance.

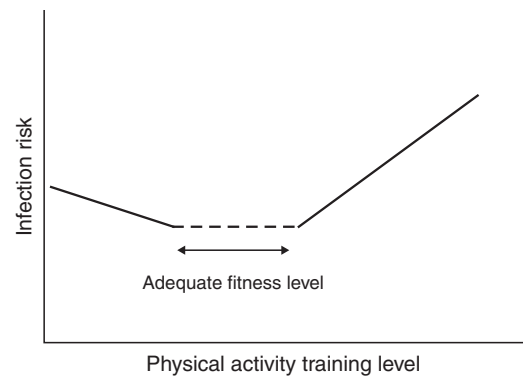
Several studies have pointed to the benefits of carbohydrate (CHO) ingestion before and during exercise for maintaining immune competence (attenuated reduction in functional responses in a number of immune cells and mediators including lymphocytes, neutrophils and inflammatory cytokines)<sup>(110–120)</sup>. In fact, several studies have reported larger stress hormones (cortisol and adrenaline) and plasma cytokine (IL-1ra, IL-6 and IL-10) responses in subjects performing exercise on diets with less than 10% of energy intake from CHO, as compared to those on normal or high CHO diets<sup>(114,121)</sup>.

The amount of CHO consumed in the pre-exercise meal has recently been suggested to probably be the most important influencing factor rather than the type of CHO in modifying the immune response to prolonged exercise (approximately 112 min at 70%  $\text{VO}_{2\text{max}}$  for the first hour and 76%  $\text{VO}_{2\text{max}}$  for the last 52 min) reflected in less perturbation of the circulating numbers of leucocytes, neutrophils and T lymphocyte subsets and in decreased elevation of the plasma IL-6 concentrations immediately after exercise<sup>(122)</sup>. Bishop *et al.* showed that ingestion of CHO-rich sports drink by cyclists undertaking 2 h of moderate intensity exercise decreased the salivary IgA concentration, with no overall effect on the salivary IgA secretion rate<sup>(114)</sup>. Other studies have suggested that CHO beverage supplementation (about 1 litre/h of 6% CHO) may attenuate the serum cortisol increases in both runners and cyclists<sup>(110,116–118,123–126)</sup>. Summarizing, besides the number of studies supporting the effects of CHO supplementation on the maintenance of the immune response after exercise, further studies are needed to understand the clinical significance.

High-intensity physical exercise disrupts the balance between oxidants and antioxidant defences contributing to free radical generation<sup>(127)</sup>. Thus, although the cellular antioxidant mechanism adapts efficiently to exercise, extreme physical exercise causes oxidative damage to athletes, which may be associated with immune alterations<sup>(128–130)</sup>.

Vitamin C supplementation ( $\geq 1000$  mg/d) augmented the increase in lymphocyte counts after exercise, attenuated the serum cortisol increase in ultramarathon runners and attenuated the increase of inflammatory cytokines<sup>(131–135)</sup>. No significant differences in the self-reported incidence of URTI after exercise have been found when the subjects consumed 1000 mg vitamin C daily for 2 months prior to and 1 month after the marathon<sup>(136)</sup>.

An inhibitory effect of combined vitamin C and vitamin E supplementation (for 4 weeks) on plasma IL-6 and



**Fig. 3.** Proposed model of the association between physical activity training level and infection risk.

cortisol responses to prolonged exercise has been reported in physically active non-athletes<sup>(137)</sup>. Acute supplementation with Zn and vitamin E did not have an effect on the cortisol response to exercise in eumenorrhic runners<sup>(138)</sup>.

Restoring glutamine levels after prolonged exercise to physiological levels have been reported to be useful in immune response recovery<sup>(17)</sup>. However, a large number of studies evaluating the glutamine supplementation effect have not found consistent effects on the immune response (lymphocyte and neutrophil counts, salivary IgA levels, oxidative burst activity, or plasma IL-6 concentrations) after exercise<sup>(36,134,139–142)</sup>. Consequently, the available literature does not provide evidence to support the beneficial effect of glutamine supplementation during exercise.

The fact of increasing the dietary fat intake of athletes to 42%, while maintaining energy intake equal to expenditure, has been reported to improve endurance exercise performance at 60–80% of  $\text{VO}_{2\text{max}}$  in cyclists, soldiers and runners<sup>(143)</sup>. Other studies have found no effects on immunity after supplementation of *n*-3 PUFA<sup>(144)</sup>. Further studies are needed to determine whether the beneficial effects of higher fat diets or fatty acid supplementation in athletes are able to reduce their rate of infections.

Soyabean supplementation with moderate-intensity endurance exercise in rats may be useful in the prevention of the impairment of T-cell immunity caused by soyabean supplementation through increases in interferon- $\gamma$  production and changes to the CD4+ :CD8+ ratio<sup>(145)</sup>.

Probiotics could positively affect athletic performance through enhanced recovery from fatigue, an improved immune function, and maintenance of a healthy gastrointestinal tract function<sup>(146)</sup>. There is emerging evidence of the humoral and cell-mediated immune systems with probiotic enhancement<sup>(147)</sup>, although the effects of probiotics on the immune function in highly trained athletes remain unclear<sup>(17,148)</sup>.

Increased salivary IgA levels have been observed among a cohort of athletes (35 distance runners; 15 female, 20 male; 35–58 years) following colostrum supplementation for 12 weeks<sup>(149)</sup>.

Ginseng (1125 mg/d) consumption has also been evaluated in 10 healthy sedentary males for 35 d after a moderate-exercise protocol. No effects were found in the total leucocyte, neutrophil, monocyte, or lymphocyte

(CD3+, CD4+, CD16+ and CD20+) counts, lymphocyte proliferation or neutrophil oxidative burst<sup>(150)</sup>.

### Ageing

Nutrition supplementation in ageing has unclear benefits. In middle-aged adults, several studies have reported weight loss and reduced serum CRP, IL-6 and IL-18, whereas exercise training alone is able to reduce serum CRP, IL-6, and blood mononuclear cell production of TNF $\alpha$  and IL-1 $\alpha$  in patients at risk for heart disease<sup>(96–98)</sup>.

Hammett *et al.* performed a randomized controlled trial to assess the effects of 6 months' regular exercise training on serum CRP levels in a healthy elderly population (60–85 years). The training intensity was increased gradually so that by the fourth month, participants were training for 45 min at a heart rate of 80% of their current estimated VO<sub>2max</sub>. Although an improvement of the cardiorespiratory fitness of these subjects was reported, no significant reduction was found in their serum CRP levels<sup>(151)</sup>.

Anyway, since a longer-term intervention (10 months) in older adults found that aerobic exercise training reduced serum IL-6, CRP, IL-18 and aerobic exercise plus flexibility–balance–resistance exercise training were associated with reduced TNF $\alpha$ <sup>(152)</sup>, the type and intensity of exercise may be an important determinant in reducing inflammatory markers.

The effects of 17 weeks physical exercise and micronutrient enriched foods were evaluated on cellular immune response in the elderly population of 112 independently living, frail elderly men and women (mean age 79.2 (sd 5.9)). While the authors found that exercise may prevent or slow the age-related decline in the immune response measured by the delayed-type hypersensitivity skin test response and micronutrient enriched foods showed no effect<sup>(153)</sup>.

### Summary and perspectives

Numerous studies have shown an inverse relationship between heavy exercise and immune response, while moderate physical activity or moderate–regulated training may enhance the immune function (Fig. 3). The pre-event fitness status also seems to be an important individual factor regarding this relationship<sup>(98)</sup>. In any case, physical activity is an important health-related variable, albeit difficult to measure.

The amount and type of physical activity that is needed to decrease the risk substantially seems to be moderate in both absolute and relative terms<sup>(99)</sup>. The burden to population health caused by physical inactivity is huge and the potential to improve population health by increased physical activity is promising. An additional number of factors including age, health status, exercise duration, intensity and specific nutrient intake (i.e. CHO, glutamine, vitamins, etc.) seem to influence the response of the immune system to physical activity<sup>(11,102)</sup>.

Further research is needed to identify additional moderating factors and to understand the mechanisms of immune cell mobilization and resistance to infections. Additional

research is also necessary to find out to what extent physical activity training can exert an effect on the immune function and inflammatory markers in a global healthy population.

Finally, although adequate nutrition and regular physical activity habits may synergistically improve health, the clinical trials using nutritional supplements tried in athletes to counteract the immune suppression have been so far inconclusive. Future studies are required to clarify issues such as the appropriate specific dietary supplementation dosage, duration and ingestion patterns.

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### References

1. Caspersen CJ, Powell KE & Christenson GM (1985) Physical activity, exercise, and physical fitness: definitions and distinctions for health-related research. *Public Health Rep* **100**, 126–131.
2. Ainsworth BE, Haskell WL, Leon AS *et al.* (1993) Compendium of physical activities, classification of energy costs of human physical activities. *Med Sci Sports Exerc* **25**, 71–80.
3. Hambrecht R, Wolf A, Gielen S *et al.* (2000) Effect of exercise on coronary endothelial function in patients with coronary artery disease. *N Engl J Med* **342**, 454–460.
4. Blair SN, Cheng Y & Holder JS (2001) Is physical activity or physical fitness more important in defining health benefits? *Med Sci Sports Exerc* **33**, Suppl. 6, S379–S399 discussion S419–S420.
5. Thompson PD, Buchner D & Pina IL *et al.* (2003) Exercise and physical activity in the prevention and treatment of atherosclerotic cardiovascular disease: a statement from the Council on Clinical Cardiology (Subcommittee on Exercise, Rehabilitation, and Prevention) and the Council on Nutrition, Physical Activity, and Metabolism (Subcommittee on Physical Activity). *Circulation* **107**, 3109–3116.
6. Melzer K, Kayser B & Pichard C (2004) Physical activity: the health benefits outweigh the risks. *Curr Opin Clin Nutr Metab Care* **7**, 641–647.
7. Braith RW & Stewart KJ (2006) Resistance exercise training: its role in the prevention of cardiovascular disease. *Circulation* **113**, 2642–2650.
8. Manini TM, Everhart JE, Patel KV *et al.* (2006) Daily activity energy expenditure and mortality among older adults. *JAMA* **296**, 171–179.
9. Booth FW, Gordon SE, Carlson CJ *et al.* (2000) Waging war on modern chronic diseases: primary prevention through exercise biology. *J Appl Physiol* **88**, 774–787.
10. Senchina DS & Kohut ML (2007) Immunological outcomes of exercise in older adults. *Clin Interv Aging* **2**, 3–16.
11. Timmons BW & Cieslak T (2008) Human natural killer cell subsets and acute exercise: a brief review. *Exerc Immunol Rev* **14**, 8–23.
12. Brolinson PG & Elliott D (2007) Exercise and the immune system. *Clin Sports Med* **26**, 311–319.

13. Klentrou P, Cieslak T, McNeil M *et al.* (2002) Effect of moderate exercise on salivary IgA and infection risk in humans. *Eur J Appl Physiol* **87**, 153–158.
14. Matthews CE, Ockene IS, Freedson PS *et al.* (2002) Moderate to vigorous physical activity and risk of upper-respiratory tract infection. *Med Sci Sports Exerc* **34**, 1242–1248.
15. Nieman DC (1998) Influence of carbohydrate on the immune responses to intensive, prolonged exercise. *Exerc Immunol Rev* **4**, 64–76.
16. Chubak J, McTiernan A, Sorensen B *et al.* (2006) Moderate-intensity exercise reduces the incidence of colds among postmenopausal women. *Am J Med* **119**, 937–942.
17. Moreira A, Kekkonen RA, Delgado L *et al.* (2007) Nutritional modulation of exercise-induced immunodepression in athletes: a systematic review and meta-analysis. *Eur J Clin Nutr* **61**, 443–460.
18. Woods JA, Davis JM, Smith JA *et al.* (1999) Exercise and cellular innate immune function. *Med Sci Sports Exerc* **31**, 57–66.
19. MacKinnon LT (1999) *Advances in Exercise Immunology*. p. 159. Champaign, IL: Human Kinetics.
20. Mooren FC, Bloming D, Lechtermann A *et al.* (2002) Lymphocyte apoptosis after exhaustive and moderate exercise. *J Appl Physiol* **93**, 147–153.
21. MacKinnon LT (1994) Current challenges and future expectations in exercise immunology: back to the future. *Med Sci Sports Exerc* **26**, 191–194.
22. Pedersen BK & Nieman DC (1998) Exercise and immunology: integration and regulation. *Immunol Today* **19**, 204–206.
23. Lim CL, Byrne C, Chew SA *et al.* (2005) Leukocyte subset response during exercise under heat stress with carbohydrate or water intake. *Aviat Space Environ Med* **76**, 726–732.
24. Walsh NP & Whitham M (2006) Exercising in environmental extremes a greater threat to immune function? *Sports Med* **36**, 941–976.
25. Romeo J, Jiménez-Pavón D, Cervantes-Borunda M *et al.* (2008) Immunological changes after a single bout of moderate-intensity exercise in a hot environment. *J Physiol Biochem* **64**, 197–204.
26. Pyne DB (1994) Regulation of neutrophil function during exercise. *Sports Med* **17**, 245–258.
27. Nieman DC & Pedersen BK (1999) Exercise and immune function. Recent developments. *Rev Sports Med* **27**, 73–80.
28. Pedersen BK & Ullum H (1994) NK cell response to physical activity: possible mechanisms of action. *Med Sci Sports Exerc* **26**, 140–146.
29. Nieman DC, Henson DA, Gusewitch G *et al.* (1993) Physical activity and immune function in elderly women. *Med Sci Sports Exerc* **25**, 823–831.
30. Starkie RL, Hargreaves M, Rolland J *et al.* (2005) Heat stress, cytokines, and the immune response to exercise. *Brain Behav Immun* **19**, 404–412.
31. Kusaka Y, Kondou H & Morimoto K (1992) Healthy lifestyles are associated with higher natural killer cell activity. *Prev Med* **21**, 602–615.
32. Fehrenbach E & Schneider ME (2006) Trauma-induced systemic inflammatory response versus exercise-induced immunomodulatory effects. *Sports Med* **36**, 373–384.
33. Pedersen B (1996) *Immune Responses to Acute Exercise*. pp. 79–92. New York: CRC Press.
34. Pedersen BK & Hoffman-Goetz L (2000) Exercise and the immune system: regulation, integration, and adaptation. *Physiol Rev* **80**, 1055–1081.
35. Gleeson M (2007) Immune function in sport and exercise. *J Appl Physiol* **103**, 693–699.
36. Castell LM, Poortmans JR, Leclercq R *et al.* (1997) Some aspects of the acute phase response after a marathon race, and the effects of glutamine supplementation. *Eur J Appl Physiol Occup Physiol* **75**, 47–53.
37. Nieman DC, Henson DA, Smith LL *et al.* (2001) Cytokine changes after a marathon race. *J Appl Physiol* **91**, 109–114.
38. Suzuki K, Nakaji S, Yamada M *et al.* (2002) Systemic inflammatory response to exhaustive exercise. Cytokine kinetics. *Exerc Immunol Rev* **8**, 6–48.
39. Malaguarnera L, Cristaldi E, Vinci M *et al.* (2008) The role of exercise on the innate immunity of the elderly. *Eur Rev Aging Phys Act* **5**, 43–49.
40. Moraska A & Fleshner M (2001) Voluntary physical activity prevents stress-induced behavioral depression and anti-KLH antibody suppression. *Am J Physiol Regul Integr Comp Physiol* **281**, R484–R489.
41. Elphick GF, Greenwood BN, Campisi J *et al.* (2003) Increased serum nIgM in voluntarily physically active rats: a potential role for B-1 cells. *J Appl Physiol* **94**, 660–667.
42. Elphick GF, Wieseler-Frank J, Greenwood BN *et al.* (2003) B-1 cell (CD5+/CD11b+) numbers and nIgM levels are elevated in physically active vs. sedentary rats. *J Appl Physiol* **95**, 199–206.
43. MacNeil B & Hoffman-Goetz L (1993) Chronic exercise enhances in vivo and in vitro cytotoxic mechanisms of natural immunity in mice. *J Appl Physiol* **74**, 388–395.
44. MacNeil B & Hoffman-Goetz L (1993) Effect of exercise on natural cytotoxicity and pulmonary tumor metastases in mice. *Med Sci Sports Exerc* **25**, 922–928.
45. Rogers CJ, Zaharoff DA, Hance KW *et al.* (2008) Exercise enhances vaccine-induced antigen-specific T-cell responses. *Vaccine* **26**, 5407–5415.
46. Shimizu K, Kimura F, Akimoto T *et al.* (2008) Effect of moderate exercise training on T-helper cell subpopulations in elderly people. *Exerc Immunol Rev* **14**, 24–37.
47. Nasrullah I & Mazzeo RS (1992) Age-related immunosenescence in Fischer 344 rats: Influence of exercise training. *J Appl Physiol* **73**, 1932–1938.
48. Chen P, Li H, Xu F *et al.* (2003) Heavy load exercise induced dysfunction of immunity and neuroendocrine responses in rats. *Life Sci* **72**, 2255–2262.
49. Giraldo E, Garcia JJ, Hinchado MD *et al.* (2009) Exercise intensity-dependent changes in the inflammatory response in sedentary women: role of neuroendocrine parameters in the neutrophil phagocytic process and the pro-/anti-inflammatory cytokine balance. *Neuroimmunomodulation* **16**, 237–244.
50. Kasapis C & Thompson PD (2005) The effects of physical activity on serum C-reactive protein and inflammatory markers: a systematic review. *J Am Coll Cardiol* **45**, 1563–1569.
51. Brenner IKM, Shek PN & Shephard RJ (1994) Infection in athletes. *Sports Med* **17**, 86–107.
52. Pyne DB & Gleeson M (1998) Effects of intensive exercise training on immunity in athletes. *Int J Sports Med* **19**, 183–191.
53. Shephard RJ & Shek PN (1999) Exercise, immunity and susceptibility to infection. A J-shaped relationship? *Phys Sportsmed* **27**, 47–71.
54. Reid VL, Gleeson M, Williams N *et al.* (2004) Clinical investigation of athletes with persistent fatigue and/or recurrent infections. *Br J Sports Med* **38**, 42–45.
55. Castle SC (2000) Clinical relevance of age-related immune dysfunction. *Clin Infect Dis* **31**, 578–585.



56. Bauer ME, Jeckel CM, & Luz C (2009) The role of stress factors during aging of the immune system. *Ann N Y Acad Sci* **1153**, 139–152.
57. Bender BS (2003) Infectious disease risk in the elderly. *Immunol Allergy Clin North Am* **23**, 57–64.
58. Falsey AR & Walsh EE (2005) Respiratory syncytial virus infection in elderly adults. *Drugs Aging* **22**, 577–587.
59. High KP, Bradley S, Loeb M *et al.* (2005) A new paradigm for clinical investigation of infectious syndromes in older adults: Assessment of functional status as a risk factor and outcome measure. *Clin Infect Dis* **40**, 114–122.
60. Meunier N, Feillet-Coudray C, Rambeau M *et al.* (2005) Impact of micronutrient dietary intake and status on intestinal zinc absorption in late middle-aged men: the ZENITH study. *Eur J Clin Nutr* **59**, S48–S52.
61. Oldewage-Theron WH, Samuel FO & Venter CS (2008) Zinc deficiency among the elderly attending a care centre in Sharpeville, South Africa. *J Hum Nutr Diet* **21**, 566–574.
62. Unkiewicz-Winiarczyk A, Bagniuk A, Gromysz-Kałkowska K *et al.* (2009) Calcium, magnesium, iron, zinc and copper concentration in the hair of tobacco smokers. *Biol Trace Elem Res* **128**, 152–160.
63. Solana R & Pawelec G (1998) Molecular and cellular basis of immunosenescence. *Mech Aging Dev* **102**, 115–129.
64. Franceschi C, Bonafe M & Valensin S (2000a) Human immunosenescence: The prevailing of innate immunity, the failing of clonotypic immunity, and the filling of the immunological space. *Vaccine* **18**, 1717–1720.
65. Linton P & Thoman ML (2001) T-cell senescence. *Front Biosci* **6**, D248–D261.
66. Tannahill GM, Elliott J, Barry AC *et al.* (2005) 'SOCS2 can enhance interleukin-2 (IL-2) and IL-3 signaling by accelerating SOCS3 degradation'. *Mol Cell Biol* **25**, 9115–9126.
67. Hong MS, Dan JM, Choi J-Y *et al.* (2004) Age-associated changes in the frequency of naive, memory, and effector CD8+ T cells. *Mech Aging Dev* **125**, 615–618.
68. Naylor K, Li G, Vallejo AN *et al.* (2005) The influence of age on T cell generation and TCR diversity. *J Immunol* **174**, 7446–7452.
69. Schwab R, Russo C & Weksler ME (1992) Altered major histocompatibility complex-restricted antigen recognition by T cells from elderly humans. *Eur J Immunol* **22**, 2989–2993.
70. Xu X, Beckman I, Dimopoulos K *et al.* (1992) Age-related changes in the expression of T cell activation antigens following phytohaemagglutinin stimulation. *Exp Clin Immunogenet* **9**, 203–211.
71. Miller RA, Jacobson B, Weil G *et al.* (1987) Diminished calcium influx in lectin-stimulated T cells from old mice. *J Cell Physiol* **132**, 337–342.
72. Ohkusu K, Du J, Isobe K-I *et al.* (1997) Protein kinase C $\alpha$ -mediated chronic signal transduction for immunosenescence. *J Immunol* **159**, 2082–2084.
73. Gupta S (2000) Molecular steps of cell suicide: an insight into immune senescence. *J Clin Immunol* **20**, 229–239.
74. McLeod JD (2000) Apoptotic capability in aging T cells. *Mech Aging Dev* **121**, 151–159.
75. Froelich CJ, Burkett JS, Guiffaut S *et al.* (1988) Phytohemagglutinin induced proliferation by aged lymphocytes: Reduced expression of high affinity interleukin-2 receptors and interleukin-2 secretion. *Life Sci* **43**, 1583–1590.
76. Castle SC, Uyemara K, Crawford W *et al.* (1999) Age-related impaired proliferation of peripheral blood mononuclear cells is associated with an increase in IL-10 and IL-12. *Exp Gerontol* **34**, 243–252.
77. Castle S, Uyemara K, Wong W *et al.* (1997) Evidence of enhanced type 2 immune response and impaired upregulation of a type 1 response in frail elderly nursing home residents. *Mech Aging Dev* **94**, 7–16.
78. Rink L, Cakman I & Kirchner H (1998) Altered cytokine production in the elderly. *Mech Aging Dev* **102**, 199–209.
79. Fagiolo U, Cossarizza A, Scala E *et al.* (1993) Increased cytokine production in mononuclear cells of healthy elderly people. *Eur J Immunol* **23**, 2375–2378.
80. Riancho JA, Zarrabeitia MT, Amado JA *et al.* (1994) Age-related differences in cytokine secretion. *Gerontology* **40**, 8–12.
81. Gardner EM & Murasko DM (2002) Age-related changes in Type 1 and Type 2 cytokine production in humans. *Biogerontol* **3**, 271–289.
82. Whisler RL & Newhouse YG (1985) Immunosenescence of the human B cell system: Impaired activation/proliferation in response to autologous monocytes pulsed with Staph protein A and the effects of interleukins 1 and 2 compared to interferon. *Lymphokine Res* **4**, 331–337.
83. Zheng B, Han S, Takahashi Y *et al.* (1997) Immunosenescence and germinal center reaction. *Immunol Rev* **160**, 63–77.
84. Frasca D, Riley RL, & Blomberg BB (2005) Humoral immune response and B cell functions including immunoglobulin class switch are downregulated in aged mice and humans. *Semin Immunol* **17**, 378–384.
85. Ennist DL, Jones KH, St Pierre RL *et al.* (1986) Functional analysis of the immunosenescence of the human B cell system: Dissociation of normal activation and proliferation from impaired terminal differentiation into IgM immunoglobulin-secreting cells. *J Immunol* **136**, 99–105.
86. Spencer NFL & Daynes RA (1997) IL-12 directly stimulates expression of IL-10 by CD5+ B cells and IL-6 by both CD5+ and CD5- B cells: Possible involvement in age-associated cytokine dysregulation. *Int Immunol* **9**, 745–754.
87. Whisler RL & Grants IS (1993) Age-related alterations in the activation and expression of phosphotyrosine kinases and protein kinase C (PKC) among human B cells. *Mech Aging Dev* **71**, 31–46.
88. Leveille SG, Gray S, LaCroix AZ *et al.* (2000) Physical inactivity and smoking increase risk for serious infections in older women. *J Am Geriatr Soc* **48**, 1582–1588.
89. Bruunsgaard H & Pedersen BK (2000) Effects of exercise on the immune system in the elderly population. *Immunol Cell Biol* **78**, 523–531.
90. Drela N, Kozdron E & Szczypiorski P (2004) Moderate exercise may attenuate some aspects of immunosenescence. *BMC Geriatr* **29**, 4–8.
91. Kohut ML & Senchina DS (2004) Reversing age-associated immunosenescence via exercise. *Exerc Immunol Rev* **10**, 6–41.
92. McFarlin BK, Flynn MG, Campbell WW *et al.* (2006) Physical activity status, but not age, influences inflammatory biomarkers and toll-like receptor 4. *J Gerontol A Biol Sci Med Sci* **61**, 388–393.
93. Kostka T, Berthouze SE, Lacour J-R *et al.* (2000) The symptomatology of upper respiratory tract infections and exercise in elderly people. *Med Sci Sports Exerc* **32**, 46–51.
94. Fahlman M, Boardley D, Flynn MG *et al.* (2000) Effects of endurance training on selected parameters of immune function in elderly women. *Gerontology* **46**, 97–104.
95. Raso V, Benard G, Da Silva Duarte AJ *et al.* (2007) Effect of resistance training on immunological parameters of healthy elderly women. *Med Sci Sports Exerc* **39**, 2152–2159.
96. Smith JK, Dykes R, Douglas JE *et al.* (1999) Long-term exercise and atherogenic activity of blood mononuclear



- cells in persons at risk of developing ischemic heart disease. *JAMA* **281**, 1722–1727.
97. Esposito K, Pontillo A, Di Palo C *et al.* (2003) Effect of weight loss and lifestyle changes on vascular inflammatory markers in obese women: a randomized trial. *JAMA* **289**, 1799–1804.
  98. Goldhammer E, Tanchilevitch A, Maor I *et al.* (2005) Exercise training modulates cytokines activity in coronary heart disease patients. *Int J Cardiol* **10**, 93–99.
  99. Vuori IM (2001) Health benefits of physical activity with special reference to interaction with diet. *Public Health Nutr* **4**, 517–528.
  100. Romeo J, Wärnberg J, Gómez-Martínez S *et al.* (2008) Neuroimmunomodulation by nutrition in stress situations. *Neuroimmunomodulation* **15**, 165–169.
  101. Wärnberg J, Gomez-Martinez S, Romeo J *et al.* (2009) Nutrition, inflammation, and cognitive function. *Ann N Y Acad Sci* **1153**, 164–175.
  102. Moreira A, Delgado L, Moreira P *et al.* (2009) Does exercise increase the risk of upper respiratory tract infections? *Br Med Bull* **90**, 111–131.
  103. Grimble RF (2001) Nutritional modulation of immune function. *Proc Nutr Soc* **60**, 389–397.
  104. Marcos A, Nova E & Montero A (2003) Changes in the immune system are conditioned by nutrition. *Eur J Clin Nutr* **57**, Suppl. 1, S66–S69.
  105. Grimble RF (1997) Effect of antioxidative vitamins on immune function with clinical applications. *Int J Vitam Nutr Res* **67**, 312–320.
  106. Victor VM & De la Fuente M (2002) N-acetylcysteine improves in vitro the function of macrophages from mice with endotoxin-induced oxidative stress. *Free Radic Res* **36**, 33–45.
  107. Chandra RK (2002) Nutrition and the immune system from birth to old age. *Eur J Clin Nutr* **56**, S73–S76.
  108. Gleeson M (2000) The scientific basis of practical strategies to maintain immunocompetence in elite athletes. *Exerc Immunol Rev* **6**, 75–101.
  109. Gleeson M, Nieman DC & Pedersen BK (2004) Exercise, nutrition and immune function. *J Sports Sci* **22**, 115–125.
  110. Bishop NC, Blannin AK, Robson PJ *et al.* (1999) The effects of carbohydrate supplementation on immune responses to a soccer-specific exercise protocol. *J Sports Sci* **17**, 787–796.
  111. Bishop NC, Blannin AK, Walsh NP *et al.* (2001) Carbohydrate beverage ingestion and neutrophil degranulation responses following cycling to fatigue at 75%  $\text{VO}_{2\text{max}}$ . *Int J Sports Med* **22**, 226–231.
  112. Bishop NC, Walsh NP, Haines DL *et al.* (2001) Pre-exercise carbohydrate status and immune responses to prolonged cycling: I. Effect on neutrophil degranulation. *Int J Sport Nutr Exerc Metab* **11**, 490–502.
  113. Bishop NC, Walsh NP, Haines DL *et al.* (2001c) Pre-exercise carbohydrate status and immune responses to prolonged cycling: II. Effect on plasma cytokine concentration. *Int J Sport Nutr Exerc Metab* **11**, 503–512.
  114. Bishop NC, Blannin AK, Armstrong E *et al.* (2000) Carbohydrate and fluid intake affect the saliva flow rate and IgA response to cycling. *Med Sci Sports Exerc* **32**, 2046–2051.
  115. Henson DA, Nieman DC, Parker JC *et al.* (1998) Carbohydrate supplementation and the lymphocyte proliferative response to long endurance running. *Int J Sports Med* **19**, 574–580.
  116. Nieman DC, Henson DA, Garner EB *et al.* (1997c) Carbohydrate affects natural killer cell redistribution but not activity after running. *Med Sci Sports Exerc* **29**, 1318–1324.
  117. Nieman DC, Davis JM, Henson DA *et al.* (2003) Carbohydrate ingestion influences skeletal muscle cytokine mRNA and plasma cytokine levels after a 3-h run. *J Appl Physiol* **94**, 1917–1925.
  118. Nieman DC, Davis JM, Brown VA *et al.* (2004) Influence of carbohydrate ingestion on immune changes after 2 h of intensive resistance training. *J Appl Physiol* **96**, 1292–1298.
  119. Starkie RL, Angus DJ, Rolland J *et al.* (2000) Effect of prolonged, submaximal exercise and carbohydrate ingestion on monocyte intracellular cytokine production in humans. *J Physiol* **528**, 647–655.
  120. Starkie RL, Arkinstall MJ, Koukoulas I *et al.* (2001) Carbohydrate ingestion attenuates the increase in plasma interleukin-6, but not skeletal muscle interleukin-6 mRNA, during exercise in humans. *J Physiol* **533**, 585–591.
  121. Mitchell JB, Pizza FX, Paquet A *et al.* (1998) Influence of carbohydrate status on immune responses before and after endurance exercise. *J Appl Physiol* **84**, 1917–1925.
  122. Chen YJ, Wong SH, Wong CK *et al.* (2008) The effect of a pre-exercise carbohydrate meal on immune responses to an endurance performance run. *Br J Nutr* **100**, 1260–1268.
  123. Koch AJ, Pottetiger JA, Chan MA *et al.* (2001) Minimal influence of carbohydrate ingestion on the immune response following acute resistance exercise. *J Sport Nutr Exerc Metab* **11**, 149–161.
  124. Bacurau RF, Bassit RA, Sawada L *et al.* (2002) Carbohydrate supplementation during intense exercise and the immune response of cyclists. *Clin Nutr* **21**, 423–429.
  125. McAnulty SR, McAnulty LS, Nieman DC *et al.* (2003) Influence of carbohydrate ingestion on oxidative stress and plasma antioxidant potential following a 3 h run. *Free Radic Res* **37**, 835–840.
  126. Chan MA, Koch AJ, Benedict SH *et al.* (2003) Influence of carbohydrate ingestion on cytokine responses following acute resistance exercise. *Int J Sport Nutr Exerc Metab* **13**, 454–465.
  127. Packer L, Cadenas E, Davies KJ (2008) Free radicals and exercise: an introduction. *Free Radic Biol Med* **44**, 123–125.
  128. Mena P, Maynar M, Gutierrez JM *et al.* (1991) Erythrocyte free radical scavenger enzymes in bicycle professional racers. Adaptation to training. *Int J Sports Med* **12**, 563–566.
  129. Tauler P, Aguilo A, Fuentespina E *et al.* (2002) Diet supplementation with vitamin E, vitamin C and beta-carotene cocktail enhances basal neutrophil antioxidant enzymes in athletes. *Pflugers Arch* **443**, 791–797.
  130. Radak Z, Pucsuk J, Boros S *et al.* (2000) Changes in urine 8-hydroxydeoxyguanosine levels of super-marathon runners during a four-day race period. *Life Sci* **66**, 1763–1767.
  131. Nieman DC, Henson DA, Butterworth DE *et al.* (1997b) Vitamin C supplementation does not alter the immune response to 2.5 hours of running. *Int J Sport Nutr* **7**, 173–184.
  132. Nieman DC, Peters EM, Henson DA *et al.* (2000b) Influence of vitamin C supplementation on cytokine changes following an ultramarathon. *J Interferon Cytokine Res* **20**, 1029–1035.
  133. Nieman DC, Henson DA, McAnulty SR *et al.* (2002b) Influence of vitamin C supplementation on oxidative and immune changes after an ultramarathon. *J Appl Physiol* **92**, 1970–1977.
  134. Peters EM, Anderson R, Nieman DC *et al.* (2001a) Vitamin C supplementation attenuates the increases in circulating cortisol, adrenaline and anti-inflammatory polypeptides

- following ultramarathon running. *Int J Sports Med* **22**, 537–543.
135. Peters EM, Anderson R, & Theron AJ (2001b) Attenuation of increase in circulating cortisol and enhancement of the acute phase protein response in vitamin C-supplemented ultramarathoners. *Int J Sports Med* **22**, 120–126.
136. Himmelstein S, Robergs R, Koehler K *et al.* (1998) Vitamin C supplementation and upper respiratory tract infections in marathon runners. *J Exerc Physiol* (online) **1**, 1–17.
137. Fischer CP, Hiscock NJ, Penkowa M *et al.* (2004) Supplementation with vitamins C and E inhibits the release of interleukin-6 from contracting human skeletal muscle. *J Physiol* **558**, 633–645.
138. Singh A, Papanicolaou DA, Lawrence LL *et al.* (1999) Neuroendocrine responses to running in women after zinc and vitamin E supplementation. *Med Sci Sports Exerc* **31**, 536–542.
139. Krzywkowski K, Petersen EW, Ostrowski K *et al.* (2001a) Effect of glutamine supplementation on exercise-induced changes in lymphocyte function. *Am J Physiol Cell Physiol* **281**, C1259–C1265.
140. Krzywkowski K, Petersen EW, Ostrowski K *et al.* (2001b) Effect of glutamine and protein supplementation on exercise-induced decreases in salivary IgA. *J Appl Physiol* **91**, 832–838.
141. Walsh NP, Blannin AK, Bishop NC *et al.* (2000) Effect of oral glutamine supplementation on human neutrophil lipopolysaccharide-stimulated degranulation following prolonged exercise. *Int J Sport Nutr Exerc Metab* **10**, 39–50.
142. Hiscock N, Petersen EW, Krzywkowski K *et al.* (2003) Glutamine supplementation further enhances exercise-induced plasma IL-6. *J Appl Physiol* **95**, 145–148.
143. Venkatraman JT, Leddy J & Pendergast D (2000) Dietary fats and immune status in athletes: clinical implications. *Med Sci Sports Exerc* **32**, Suppl. 7, S389–S395.
144. Toft AD, Thorn M, Ostrowski K *et al.* (2000) N-3 polyunsaturated fatty acids do not affect cytokine response to strenuous exercise. *J Appl Physiol* **89**, 2401–2406.
145. Kwon DK, Hwang KH, Kim YK *et al.* (2008) Effects of swimming exercise and soybean supplementation on the immune functions of rats fed a high-fat diet. *Clin Exp Pharmacol Physiol* **35**, 638–642.
146. Nichols AW (2007) Probiotics and athletic performance: a systematic review. *Curr Sports Med Rep* **6**, 269–273.
147. Erickson KL & Hubbard NE (2000) Probiotics immunomodulation in health and disease. *J Nutr* **130**, Suppl. 2S, 403S–409S.
148. Kekkonen RA, Vasankari TJ, Vuorimaa T *et al.* (2007) The effect of probiotics on respiratory infections and gastrointestinal symptoms during training in marathon runners. *Int J Sport Nutr Exerc Metab* **17**, 352–363.
149. Crooks CV, Wall CR, Cross ML *et al.* (2006) The effect of bovine colostrum supplementation on salivary IgA in distance runners. *Int J Sport Nutr Exerc Metab* **16**, 47–64.
150. Biondo PD, Robbins SJ, Walsh JD *et al.* (2008) A randomized controlled crossover trial of the effect of ginseng consumption on the immune response to moderate exercise in healthy sedentary men. *Appl Physiol Nutr Metab* **33**, 966–975.
151. Hammett CJ, Oxenham HC, Baldi JC *et al.* (2004) Effect of six months' exercise training on C-reactive protein levels in healthy elderly subjects. *J Am Coll Cardiol* **44**, 2411–2413.
152. Kohut ML, McCann DA, Russell DW *et al.* (2006) Aerobic exercise, but not flexibility/resistance exercise, reduces serum IL-18, CRP, and IL-6 independent of beta-blockers, BMI, and psychosocial factors in older adults. *Brain Behav Immun* **20**, 201–209.
153. Chin A, Paw MJ, de Jong N *et al.* (2000) Immunity in frail elderly: a randomized controlled trial of exercise and enriched foods. *Med Sci Sports Exerc* **32**, 2005–2011.
154. Lancaster GI, Halson SL, Khan Q *et al.* (2004) Effects of acute exhaustive exercise and chronic exercise training on type 1 and type 2 T lymphocytes. *Exerc Immunol Rev* **10**, 91–106.
155. Ceddia MA & Woods JA (1999) Exercise suppresses macrophage antigen presentation. *J Appl Physiol* **87**, 2253–2258.
156. Gannon GA, Rhind S, Shek PN *et al.* (2002) Naive and memory T cell subsets are differentially mobilized during physical stress. *Int J Sports Med* **23**, 223–229.
157. Fischer CP, Plomgaard P, Hansen AK *et al.* (2004) Endurance training reduces the contraction-induced interleukin-6 mRNA expression in human skeletal muscle. *Am J Physiol Endocrinol Metab* **287**, E1189–E1194.
158. Nicklas BJ, Hsu FC, Brinkley TJ *et al.* (2008) Exercise training and plasma C-reactive protein and interleukin-6 in elderly people. *J Am Geriatr Soc* **56**, 2045–2052.
159. Sari-Sarraf V, Reilly T & Doran DA (2006) Salivary IgA response to intermittent and continuous exercise. *Int J Sports Med* **27**, 849–855.