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Exercise and Immunity in Athletic Performance and a Healthy Life

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Book of Abstracts

Foreword

This booklet contains abstracts of all the talks and posters that were presented at the 10th ISEI Symposium in Oxford, 10-13 July 2011

S.1 - S.13 : Invited speaker presentations

O.1 - O.18 : Free oral communications

P.1 - P.91 : Free poster communications

Abstracts of poster communications that were submitted but not presented are not included in this document

For the sake of consistency, the numbering of oral and poster communications used at the symposium (according to the original Abstract Booklet) has been retained

Symposium Abstracts by Invited Speakers

S.1. Understanding the mechanisms that control host defence responses and genetic programming of the immune response

PS Foster

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Host defence against infection is regulated by the dynamic interplay between innate host defence pathways and the adaptive immune response. In the last 20 years significant advances have been made in understanding how the host senses infection and discriminates the type of infection, which leads to the programming of a specific adaptive (T and B cell response) for elimination of the pathogen. In this presentation I will describe how Toll Like Receptors (TLRs) sense different pathogens that lead to the activation of professional antigen presenting, which programme T cells for generation of the appropriate adaptive immune response. The role of microRNA (miRNA), small non-coding RNA species, in regulating translational mechanism is now established in mammalian cells. However, their function in regulating the genetic programmes in the immune system that regulate host defence and immunity is only beginning to emerge. In a limited number of studies TLR activation has been linked to alterations in miRNA expression and the control transcription circuits that regulate the subsequent immune response. The integration of both systems (TLR and miRNA) for the maintenance of health and induction of disease will also be discussed.

S.2. Physical inactivity, inflammation and metabolism

BK Pedersen

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Type 2 diabetes, cardiovascular diseases, colon cancer, breast cancer, dementia, and depression constitute a cluster of diseases, which defines “a diseasome of physical inactivity”. Both physical inactivity and abdominal adiposity, reflecting accumulation of visceral fat mass, are associated with the occurrence of the diseases within the diseasome. Physical inactivity appears to be an independent and strong risk factor for accumulation of visceral fat, which again is a source of systemic inflammation. Chronic inflammation is involved in the pathogenesis of insulin resistance, atherosclerosis, neurodegeneration, and tumour growth. Evidence suggests that the protective effect of exercise may to some extent be ascribed to the anti-inflammatory effect of regular exercise, which can be mediated via a reduction in visceral fat mass and/or by induction of an anti-inflammatory environment with each bout of exercise.

S.3. The anti-inflammatory potential of exercise: implications for health and disease

BK McFarlin, WL Breslin, KC Carpenter & K Strohacker

Health & Human Performance, University of Houston, USA

Regular physical activity is known to exert anti-inflammatory effects. Such effects are manifested via changes in various aspects of innate immune system function. In this presentation we will explore the various known and novel anti-inflammatory actions. Also, the role of physical activity in disease prevention will be discussed in the context of anti-inflammatory actions.

S.4. Diet, genetics, and the mammalian microbiome

R Knight

Howard Hughes Medical Institute and Department of Chemistry & Biochemistry, University of Colorado, USA

Advances in sequencing technology have led to an unprecedented ability to understand the trillions of symbionts each of us harbors. Here I discuss recent technological advances that allow us to collect dense spatial series and timeseries data and to relate them to key external parameters. Using the ob mutant and TLR5 knockout mouse models as examples, together with long timeseries collected in humans, together with the influence of diet in humans and other mammals, I discuss implications of the microbiome for behavior and prospects for future studies exploring the relationships between diet, genetics and exercise.

S.5. Exercise and neural inflammatory diseases

EJ Downer, RM O'Callaghan, EW Griffin, AM Kelly & MA Lynch

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There has been a major drive to understand the mechanisms underlying the beneficial effects of exercise since it was reported that voluntary exercise induced adult hippocampal neurogenesis. Evidence indicates that exercise induces the neurotrophin, brain-derived neurotrophic factor (BDNF), and BDNF-associated signalling, with positive outcomes on cognitive function [1,2]. However the fact that exercise also induced neurogenesis in hippocampus of aged animals stimulated an interest in investigating its possible effects in neurodegenerative diseases. It has been suggested that exercise may decrease the risk of developing Alzheimer's disease (AD) and Parkinson's disease (PD) with evidence of beneficial effects of exercise on cognitive function in AD patients and improvements in balance and musculoskeletal function in patients with PD. Data from animal models suggests that exercise decreases pathology in transgenic mouse models of AD and protect dopaminergic neurons from neurotoxic insults in models of PD [3,4]. Although the mechanisms underlying these changes are not fully understood, it is known that exercise ameliorates the oxidative changes which accompany loss of dopaminergic neurons in models of PD and attenuates the glial activation which is a feature of the brain in animal models of AD.

In addition to the beneficial effects of exercise in these conditions, data from recent studies have indicated that the age-related deficit in synaptic plasticity which accompanies the decrease in expression of BDNF is attenuated in aged rats which were placed on an exercise regime for 8 months [2]. Data will be presented which indicate that the age-related increase in inflammatory cytokines, and in signalling events which are indicative of inflammatory changes, were attenuated by exercise, which are consistent with the widely-accepted view that both acute and chronic exercise affect the function of several immune cells.

References:
[1] Cotman CW & Berchtold *Trends Neurosci* 2002: 25: 295-301
[2] O'Callaghan RM et al. *Hippocampus* 2009:, 1019-1029.
[3] Parachikova A et al. *Neurobiol Dis* 2008: 30: 121–129.
[4] Ang E-T et al. *Front Aging Neurosci* 2010: 2: 25.

S.6. Exercise as a means of reducing acute and chronic inflammation: Impact on health

J Woods, S Martin, B Pence, M Cook & R Greene

Department of Kinesiology and Community Health, University of Illinois at Urbana-Champaign, USA

Exaggerated and chronic inflammation is increasingly associated with the pathophysiology of many diseases. New pharmaceuticals are being developed to treat inflammation but there are risks of negative side-effects. The potentially beneficial role that behavioral interventions such as regular exercise play in modulating inflammatory responses and their consequences is beginning to emerge. Our lab has been interested in determining whether exercise can reduce exaggerated and/or chronic inflammation and improve sickness behaviours including fatigue and depression [1], obesity-related comorbidities [2,3], cutaneous wound healing [4], inflammatory bowel disease, and risk and recovery from influenza infection [5]. We have found that regularly performed exercise when administered prior or during inflammatory insult, in most cases, reduces the inflammatory response and improves physiological and behavioural consequences of inflammation. For example, we have found that exercise can improve cutaneous wound healing in aged and obese; an effect associated with reduced wound inflammation in aged. Exercise, when applied early after infection before symptoms, also reduces mortality to influenza virus infection; an effect associated with altered cytokine profiles within infected lungs. We have also demonstrated that modest amounts of treadmill running can reduce inflammation within visceral adipose tissue of high fat diet-fed obese mice; an effect related to improved metabolic derangements and reduced systemic inflammation. Furthermore, exercise training prior to systemic LPS administration can attenuate some, but not all, sickness behaviors in aged mice that manifest exaggerated and chronic responses to such treatment. In contrast, exercise training prior to administration of dextran sodium sulphate (DSS which induces inflammatory colitis) actually exacerbates colon inflammation in mice, while not affecting sickness behaviour. Given the exercise effects across such a wide range of models, it is likely that there are multiple mechanisms responsible for these effects. Our lab is working towards a deeper understanding of these mechanisms including whether peripheral to brain communication is altered.

References:

1. Valentine RJ et al. *Brain Behav Immun* 2009: 23(5): 643-648.
2. Vieira VJ et al. *Brain Behav Immun* 2009: 23(4): 485-491.
3. Vieira VJ et al. *Am J Physiol* 2009: 296(5): E1164-E1171.
4. Keylock KY et al. *Am J Physiol* 2008: 294(1): R179-R184.
5. Lowder T et al. *Exerc Immunol Rev* 2006: 12: 97-111.

S.7. Immunodepression and intensive exercise: the evidence

PC Calder

School of Medicine, University of Southampton, UK

Evidence for an impact of intensive exercise on different aspects of the immune response will be presented. Possible dietary strategies to intervene will be discussed and supporting evidence evaluated.

S.8. Immune depression in response to intense exercise - the practical implications for athletes and coaches

S Bermon

Monaco Institute for Sports Medicine and Surgery, Monaco

Several studies have suggested that athletes are at increased risk of respiratory tract infections (URTI). Exercise-induced suppression of some immune functions after intense and/or prolonged exercise and during strenuous training periods may explain this increased susceptibility. Regular sharing of the same training and living facilities within a team may also contribute to this increased frequency or duration of URTI. Moreover, the increased exposure to new pathogens while travelling put the athlete at a higher risk of gastrointestinal infections. Thus, infectious episodes are the most common reason for presenting to a sports medicine clinic and for not to train.

There is no single intervention that completely eliminates the risk of contracting an infection, but there are several effective ways of reducing the number, duration and severity of infectious episodes incurred over a period. The presented practical implications are primary or secondary (after infection onset) preventive measures. They are mostly driven by common sense, and can be understood by athletes and their entourage, keeping in mind the contagious nature of viruses, bacteria and fungi.

S.9. The effect of psychological stress and exercise on immunity: Qualitatively distinct or just a question of magnitude?

VE Burns & JA Bosch

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Psychological stress and exercise have profound influences on immune function, with considerable evidence that these effects are mediated by neuroendocrine pathways. Despite considerable overlap in observations and underlying mechanisms, there are also some key distinctions between the immunological responses to these different stimuli. To date, though, there has been relatively little cross-talk between psycho-neuro-immunologists and exercise immunologists. We believe that by enhancing communication and cooperation between these research fields, we will strengthen both fields and enhance our understanding of how these stress-induced changes may affect human health.

Our laboratory has been using both exercise and psychological stress paradigms to study behavioural influences on immune function. This talk will present data from two research programmes that have been our focus in recent years. The first examines stress-induced lymphocytosis and lymphopenia, and their neuro-endocrine and microbiological determinants. The second programme of research investigates the use of psychological and exercise stress as adjuvants to enhance vaccination responses. Similarities and distinctions between the immunological effects of the different stressor types will be highlighted, their implications discussed, and proposals made for further research bringing together stress and exercise immunology.

S.10. Exercise produces stress resistance: Benefits for mental and physical health

M Fleshner, K Speaker & BN Greenwood

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The cascade of responses that comprise the acute stress response is adaptive under most circumstances. Increases in respiration, heart rate, blood pressure, pupil dilation, energy mobilization, focused attention and immunity, for example, function in concert to promote successful fight or flight responses and improve one's chances for survival. It is important to emphasize, therefore, that stress resistance does not imply the absence of the stress response. Instead, I suggest that optimal stress resistance delays the "tipping point" from adaptive to maladaptive responses, and increases the duration and/or intensity of stressor exposure needed to cross over. In other words, individuals with high levels of stress resistance are able to endure a great deal of stress before experiencing negative effects.

Physically active people and animals are more resistant to the negative consequences of stressor exposure on mental and physical health. Using an animal model, my laboratory has demonstrated that compared with sedentary rats, rats living with running wheels for six weeks prior to exposure to an intense acute stressor are protected against a variety of negative stressor consequences including affective dysregulation (i.e., anxiety and depression) and immune modulation. I will present evidence from a series of studies that exercise produces changes in central stress-responsive neurocircuitry. Specifically, voluntary freewheel running produces changes in the brain that modulate dopamine reward pathways [1], reduce stress-induced activation of the dorsal raphe (DRN) 5HT system [2,3] and reduce central sympathetic nervous system (SNS) activation [4]. Constraint of DRN 5HT contributes to the benefits of exercise on stress-induced affective dysregulation [4]; whereas constraint in central SNS drive contributes to the protection against stress-induced, suppression of T cell dependent antibody responses [6,7], and activation of systemic sterile inflammatory responses [8]. Thus regular moderate physical activity promotes stress resistance by inducing plasticity in stress-responsive neurocircuitry.

References:

1. Greenwood BN et al. *Behavioral Brain Research* 2011: 217: 354-362.
2. Greenwood BN et al. *J Neurosci* 2003: 23: 2889-2898.
3. Greenwood BN et al. *Brain Res* 2005: 1033: 164-178.
4. Greenwood BN et al. *Neuroscience* 2003: 120: 269-281.
5. Greenwood N & Fleshner M. *Exerc Sci Sport Rev* 2011: 39.
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7. Fleshner M et al. In *The Neuroimmunological Basis of Behavior and Mental Disorders,* Springer Publishing, New York, NY 2009: pp 87-107.
8. Johnson JD et al. *Neuroscience* 2005: 135: 1295-1307.

S.11. Optimizing immune function: do we know what to measure and how to interpret?

R Albers

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The immune system has evolved as a defense mechanism against infections and tissue damage and is thus crucial to the maintenance of health on a day-to-day basis. The immune system comprises structural and cellular elements that are dispersed throughout the body. Inappropriate immune regulation can aggravate tissue damage, lead to chronic inflammation and development of allergic and autoimmune diseases. Exogenous factors such as exercise or nutrition can influence immune functioning in many ways and at many levels. It is therefore important to consider the immunological relevance of effects observed. Specific *in vivo* measures such as response to vaccination can for instance indicate resistance to infection, but due to the complexities of the system, no single assay or marker allows conclusions to be drawn about the immune system as a whole. Current approaches and developments in the assessment of the impact of exogenous factors on immune function will be discussed, focussing on the impact of nutrition.

S.12. Stress, immunity and ageing

JM Lord

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Life expectancy in the developed world is increasing at a rate of 2 years per decade and there are now more people aged over 60 years than under 16 in the UK [1]. However there is evidence that healthy life expectancy (years free of ill health) is not keeping pace [2] and considerable research effort in now being placed upon understanding the detrimental effects of the ageing process in order to develop interventions to maintain people in good health for longer.

Most systems in the body undergo a functional decline with age and the immune system is a key victim of the ageing process. The age-related decline in immune function, termed immunesenescence, is evidenced by increased susceptibility to infection with age and increased mortality associated with infections [3], vaccination responses are also significantly reduced in older adults [4]. Immunesenescence is the consequence of changes to both the adaptive and innate arms of the immune system: reduced production of T cells by the thymus and reduced proliferation of memory T cells results in poor responses to new pathogens and reduced ability to deal with pathogens previously encountered [4]. In addition reduced neutrophil bactericidal function [5] increases susceptibility to bacterial infections such as pneumonia. Ageing is also accompanied by an increased basal level of pro-inflammatory cytokines such as TNFα and IL6, termed inflammaging [6], which is thought to contribute to age-related pathology including metabolic syndrome, cardiovascular disease and sarcopenia.

The immune system does not operate in isolation and is influenced significantly by the endocrine environment, which also changes with age. This paper will discuss changes in the hypothalamic-pituitary-adrenal (HPA) axis with ageing [7] and how this impacts upon the response to both physical stress (hip fracture), including the key role played by cortisol and dehydroepiandrosterone sulphate in modulating neutrophil responses. The paper will finish by discussing evidence for the benefits of physical activity for maintaining immune function into old age, including epidemiological data suggesting a positive association between a high level of physical activity, the HPA axis and inflammaging.

References:

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2. House of Lords Science and Technology committee report July 2005. Ageing: Scientific aspects.
3. Laupland KB et al. *J Infect Dis* 2003: 187(9): 1452-1459.
4. Pawelec G et al. *Immunological Rev* 2005: 205: 257-268.
5. Shaw AC et al. *Current Opinion Immunol* 2010: 22: 507-513.
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Abstracts of Free Oral Communications

Orals: Exercise, Metabolism and Inflammation

O.1. Metabolite clusters of the exercise and recovery phase detected in a non-hypothesis-driven metabolomics approach

E Kenar[1], X Zhao[2], J Hansen[3], H-U Häring[4], O Kohlbacher[1], G Xu[2], R Lehmann[4], BK Pedersen[3], C Weigert[4] & P Plomgaard[3]

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Regularly performed exercise has notable health benefits. On the other hand, it poses an extreme challenge for the human body affecting various metabolic pathways and network interactions. The complete understanding of the health-promoting metabolic networks as well as the identity of the players is still under investigation. To this end, we aimed in a non-targeted metabolomics study to identify clusters of metabolites with similar time kinetics thereby gaining novel insights in the metabolic physiology of exercise and recovery.

Eight healthy human subjects completed a two hour knee-extension exercise bout followed by a three hour recovery phase under standardized conditions. Plasma samples obtained prior to, at 60, 120, 150, 180, and 300 minutes of the experiment were analyzed by a non-targeted metabolomics approach applying ultra-performance liquid chromatography (UPLC)-qTOF-mass spectrometry. For the data evaluation, we developed a novel bioinformatics approach using fuzzy c-means clustering to extract metabolite masses showing similar kinetics. Following that, the identity of characteristic masses was obtained by UPLC-MS/MS.

The time kinetics of more than 5000 metabolite ion masses led to nine different clusters containing solely masses reflecting the cluster-specific dynamic profile in at least 5 individuals. The strongest cluster showed a peak increase of metabolite ion masses at the cessation of exercise. Other clusters indicated an early and transient increase or decrease of certain metabolite masses during exercise, but also a transient increase or decrease after the exercise bout were observed. Up to 50 metabolite masses per cluster represented the dominating metabolite species. More than 90% of the metabolite ion masses did not show a specific or comparable kinetics in at least 5 individuals. Among the dominating metabolite masses, we have identified medium chain acyl-carnitines as characteristic exercise-induced biomarkers. Further identified metabolites were lysophosphatidylcholines, diacylglycerols, bilirubin, and amino acids. Based on potential identifications from mass queries of metabolite databases, the nine characteristic clusters were mapped onto pathways to elucidate the metabolic networking of exercise and to support further the identification of unknown metabolite masses.

In summary, we present here a novel approach to reveal deeper insights in the complex metabolic regulation and networking during an exercise bout and the recovery phase.

O.2. H_2O_2 release in exhaled breath condensate as a parameter for oxidative stress in exercise (ECRA)

E Marek, J Volke, K Mückenhoff, W Marek & P Platen

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Background: Exhaled breath condensate (EBC) contains numerous mediators of oxidative stress (NO, H_2O_2). Exercise is characterised by an increase of reactive oxygen species (ROS) in blood, which may also be found in EBC. Building of H_2O_2 can be induced by ROS. In order to get inside into the correlation of H_2O_2 release in EBC and exercise intensities, we investigated H_2O_2 release at rest and at different levels of exercise in a group of young and healthy subjects.

Methods: 16 healthy subjects, 7 females and 9 males (23.3±1.5 years, 175±8 cm, 68.9±9.2 kg), were investigated, during resting conditions as well as at 60%, 75%, and 90% of maximal work capacity(pmax) (each lasting 5 minutes) on a cycle ergometer. 100 L exhaled air along with capillary blood samples from the earlobe were collected under stationary load conditions. EBC was obtained by cooling the exhaled air volume to -20°C. H_2O_2 measurements in EBC were performed using an enzym-sensor (ECo-Check, FILT). The rates of expiratory release of H_2O_2 in pmol/min were calculated from the concentrations of H_2O_2 in the EBC, the EBC volume and the time of collection.

Results: At rest H_2O_2 concentration in EBC was 216±52 nmol/L, H_2O_2 release in the collected EBC was 115±45 pmol/min. Oxygen consumption at rest was 5.3±1.1 ml/min/kg and increased to 28.3±4.5, 34.8±8.8 and 40.8±4.5 ml/min/kg at 90% (pmax) ($p<0.001$).At 60%, 75% and 90% of pmax, H_2O_2 concentration in EBC increased to 288±80, 322±71, 334 ± 95 nmol/L respectively ($p<0.01$). Taking into account the theoretical water volumes of 4.4 ml EBC derived from 100 L exhaled air, H_2O_2 release increased to 160±75, 250±88 and 357±162 pmol/min ($p<0.001$). The correlation of H_2O_2 release and ventilation can be described by $r = 0.8$.

Conclusions: In healthy subjects, a nearly 3-fold increased of H_2O_2 release in EBC was found during exhausting exercise. The elevated levels of H_2O_2 may be interpreted as an increase of ROS during exhausting exercise. In further experiments it will to be proven whether there is a correlation between H_2O_2 release in EBC and typical profiles of exercise situations.

O.3. Altered metabolic response to a single bout of exercise in mice fed a vitamin E and C- enriched diet

M Hoene, AK Pohl, R Lehmann, H-U Häring, E Schleicher & C Weigert

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The increased energy demand associated with prolonged physical exercise causes pronounced changes in metabolite fluxes and directly affects the liver, as the key regulator of energy homeostasis. In the liver, endurance exercise provokes a strong and acute response not only of metabolic, but also of stress-sensitive signalling pathways [1]. Antioxidant intake with the aim of reducing oxidative stress has a high prevalence in the general population and among endurance athletes. However, excess intake of vitamin C and E could impair the adaptational response of the muscle to physical exercise as well as beneficial effects on whole-body insulin sensitivity [2]. We attempted to clarify firstly, whether the underlying mechanisms are related to anti-oxidative or direct metabolic effects of these vitamins and secondly, if the adaptational response of the liver is affected as well.

Mice were supplemented during 4 weeks with a vitamin E and C- enriched diet before being subjected to 1h of treadmill running at non-exhaustive conditions. The acute increase in circulating fatty acids after physical exercise, and the induction of metabolic regulators like PGC-1alpha and PDK-4 in the liver were blunted with the treatment. In contrast, the induction of stress-responsive immediate early genes in the liver was not affected.

Thus, antioxidants impair the adaptive response to exercise not only in the muscle, but also in the liver, and this could be due to metabolic rather than anti-oxidative effects. Possibly, vitamin E, which is closely linked to fat turnover in the body, might directly modify energy metabolism and thus affect the hepatic response to physical exercise.

References:
1. Hoene M & Weigert C. *Exerc Immunol Rev* 2010: 16:163-183.
2. Ristow M et al. *PNAS* 2009: 106: 8665-8670.

Orals: Exercise and the Intestinal Microbiota: Function and Immunity

O.4. Response to exercise in the heat following a period of probiotics supplementation

CM Shing, JM Peake, F Lim, D Briskey & L Vitetta

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Intense exercise training and/or heat stress lead to various immune and gastrointestinal disturbances. These disturbances in immune function and gastrointestinal barrier have been associated with increases in systemic lipopolysaccharide (LPS) [1]. In view of the purported benefits of probiotics on gastrointestinal immune function, this study investigated the influence of probiotics supplementation on gastrointestinal permeability and systemic inflammation when exercising in the heat.

Twelve trained, male runners (Height: 178 ± 7cm, Body Mass: 72.6 ± 7.6, VO_2max: 62.9 ± 6.8 kg/ml/min, Age: 29 ± 7 years) completed the double-blind, placebo controlled, randomised cross-over trial. Runners exercised to fatigue at a speed corresponding to 80% ventilatory threshold in 35°C, 40% humidity. This treadmill run was repeated under the same conditions following four weeks of supplementation with either probiotics or a placebo. Following a three week washout period runners completed the cross over. Five grams each of lactulose and rhamnose were administered prior to each run and urine collected for a five hour period to assess gastrointestinal permeability. Venous blood samples were collected pre-, post-, and 1hr post-exercise for the determination of full blood counts, LPS and cytokines. Core temperature was monitored during each run via an ingested temperature sensor telemetry system.

Four weeks of supplementation with probiotics significantly reduced lactulose-to-rhamnose ratio following running in the heat (pre-exercise: 0.030 +/- 0.018, post-exercise: 0.018 +/- 0.008), when compared to a placebo (pre-exercise: 0.022 +/- 0.008, post-exercise: 0.021 +/- 0.009) ($p=0.032$). Probiotics supplementation was associated with a reduction in one hour post-exercise IL-10 ($p=0.038$) and IL-6 concentration ($p<0.005$). There was a significant effect of trial (pre- to post-supplementation) x group for LPS concentration ($p=0.02$), with post-exercise values lower following probiotics supplementation. Area under the curve for core temperature was not significantly different between groups ($p=0.34$).

Probiotics supplementation reduces gastrointestinal permeability when running in the heat, which may influence circulating inflammatory cytokines and LPS concentration.

References:
1. Lim C et al. *App Phys Nut Met* 2009: 34: 616-624.
2. Selkirk G et al. *Am J Physiol Regul Integr Comp Physiol* 2008: 296: R611-623.

O.5. The role of the gut microbiota in the acute stressor evoked sterile inflammatory response

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Exposure to acute physical or psychological stressors produces a sterile inflammatory response as evidenced by increases in inflammatory proteins in the blood [1]. The signals mediating the stress-induced cytokine release remain unknown. Previous *in vitro* studies suggest that both **P**athogen **A**ssociated **M**olecular **P**atterns (PAMPs) and **D**anger Associated **M**olecular **P**atterns (DAMPs) signal the immune system to release cytokines. Bailey et al. [2] recently reported that reducing the gut microbiota, the primary source of PAMPs in a healthy animal, reduces chronic stress-induced increases in plasma cytokines. While either signal is sufficient to release many cytokines, it has been recently reported that a subset of cytokines require both DAMP and PAMP signals for maximal release [3]. The commensal flora, therefore, may be necessary for stress to release maximal levels of dual-signal requiring cytokines. We hypothesize that stripping the gut flora should reduce stress-induced release of dual-signal requiring but not single-signal requiring, cytokines.

Male F344 rats (n=16/group) were either treated for 4 days with antibiotics in the drinking water or served as controls. Gut microbiota depletion was verified by via fecal agar bacterial growth. Rats were exposed to a well-characterized stressor (100, 5s, 1.5mA tail-shocks) or remained in their home cage. Single- (IL-6, IL-10, IL-12) and dual-signal requiring cytokines (IL-1β, IL-1α, IL-18), DAMPs (eHsp72, HMGB-1, ATP, uric acid, and blood glucose), PAMPs (peptidoglycans), and indicators of the stress response (corticosterone, splenic weight) were measured in plasma immediately after stressor termination.

Both single- and dual-signal requiring cytokines were released by stressor exposure. DAMPs were similarly stress responsive. Antibiotics did not reduce the hormonal stress response, nor did they reduce the increase in single-signal requiring cytokine production, or DAMPs. Only the release of dual-signal requiring cytokines was reduced by antibiotic administration.

These data suggest that the commensal bacteria are necessary for stress to maximally increase dual-signal requiring plasma cytokines. The absence of an effect of antibiotics on other cytokines suggest that the presence of a second signal in addition to the PAMPs derived from the commensal flora, can induce cytokine release. In the context of the stress response, the second signal may be endogenous DAMPs.

References:

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O.6. Gastrointestinal barrier dysfunction, endotoxin neutralizing capacity and immuno-inflammatory mediators following repeated exertional heat stress exposure (ECRA)

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Recent evidence in humans has shown a greater endotoxin leakage in sedentary untrained (UT) compared with endurance trained (TR) males at core temperatures below those normally assumed to be essential for the progression of heat illness or heat stroke during acute exertional heat stress (EHS) exposure [1]. Minimal data exists examining the role of gastrointestinal (GI) barrier function, circulating immuno-inflammatory mediators and physiological heat tolerance that accompanies repeated EHS exposures and whether the same cytoprotective responses observed in TR individuals [2] may be induced during repeated EHS in UT. The purpose of this work was to examine the immuno-inflammatory cascades accompanying classical physiological adaptations in untrained males following repeated EHS exposures.

Nine UT males (Age = 27 ± 2 years, $\dot{V}O_{2peak}$ = 52 ± 2 mL·kgLBM^{-1}·min^{-1}, 17 ± 2% body fatness) walked to exhaustion (EXH) at 4.5 km·h^{-1} with 2% elevation in a climatic chamber (40°C, 30% R.H.) while wearing encapsulating protective clothing on 9 separate days. Venous whole blood samples were collected at baseline (PRE), 38.0°C, 38.5°C, 39.0°C and EXH on Days 1 and 9, while PRE/EXH samples were drawn on Days 5 and 6. Samples were analyzed for endogenous endotoxin, endotoxin neutralizing capacity (ENC) and for circulating immuno-inflammatory mediators macrophage inflammatory protein (MIP)-1β, IL-6, IL-10, lipopolysaccharide binding protein (LBP), soluble (s) CD14 and extracellular (e) HSP72 by ELISA. In addition, intracellular (i) HSP72 was examined in circulating leukocyte subsets by flow cytometry.

Classical physiological adaptations, including reduced HR and increased tolerance time, delta core temperature tolerated, plasma volume and sweat rates were observed during repeated EHS exposure. Mild endotoxemia (21.3 ± 5 pg/mL) was observed at EXH on Days 1, 5, 6, 9 as well as a 14% reduction in ENC (12014 ± 717 vs. 10292 ± 929 ENU/mL) from PRE to EXH on Day 1. Accompanying the changes in endogenous endotoxin and thermoregulatory adaptations were enhanced circulating levels of IL-6, IL-10, LBP, sCD14 as well as cytoprotective increases in both eHSP72 and iHSP72.

These findings suggest that GI barrier dysfunction may be an important mechanism of immuno-inflammatory modulation and cytoprotective adaptations observed following repeated EHS exposures.

References:

1. Selkirk GA et al. *Am J Physiol Regul Integr Comp Physiol* 2008: 295: R611-R623.
2. Selkirk GA et al. *Am J Physiol Regul Integr Comp Physiol* 2009: 296: R575-R586.

Orals: Exercise and Neural Inflammatory Diseases

O.7. Acute exercise modulates the pro-BDNF and BDNF content in PBMCs in an intensity–related manner: novel cellular response to induced-stress and/or adaptation?

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The neurotrophin Brain Derived Neurotrophic Factor (BDNF) exerts its major action within the CNS, but its production and biological functions are also extended to other peripheral tissues where it displays regulatory effects on energy homeostasis and immune functions [1,2]. Moreover although several studies showed that acute exercise can transiently increase circulating BDNF, possibly contributing to benefits at neurobiological level [3], only few of them have investigated its biological sources, other the neurons, that may contribute to these systemic changes [4].

The aim of our study was therefore to verify whether an exercise-induced stress could modulate the content of BDNF and its precursor form, pro-BDNF, also within the PBMCs population.

We examined the effect of two cycling sessions, at exhaustive (MAX) and moderate intensity (IAT), of ten healthy young adults. Physiological and biological parameters were measured before and after the sessions. In addition to the endogenous expression of BDNF, pro-BDNF and their low-affinity receptor p75NTR, also the expression of anti-apoptotic (Bcl-xL) and stress response proteins (hsp 27, hsp 70, hsp 90, αB christallin), as well as the level of oxidative stress (HNE adducts) were quantified. Further analysis included hematocrit, serum and plasma BDNF, VEGF, PDGF-BB, bFGF, pro and anti-inflammatory cytokines.

Besides confirming that MAX exercise increased the serum concentration of BDNF, a novel major finding was that both exercise sessions modulated the BDNF and pro-BDNF content within PBMCs during recovery. While the pro-BDNF tended to increase after both sessions, the BDNF quantity changed in different manner according the intensity of exercise: it decreased after MAX, indicating a possible release with autrocrine destination (p75NTR also increased) and, in an opposite way, it increased after the IAT condition, indicating a possible adaptive mechanism of BDNF accumulation in response to a only moderate stress. These speculations were supported by the fact that stress response and anti apoptotic factors, but not HNE, within PBMCs were significantly regulated by the MAX condition, remaining instead stable after the IAT exercise.

These data indicate that also PBMCs could be partially responsible for the transient changes of systemic BDNF, and introduce a possible novel role of this neurotrophin in the inflammatory-like response activated by exhaustive exercise.

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Orals: Exercise and Health: Disease Prevention and Treatment

O.8. Acute exercise enhancement vaccination response: A model of weaker and stronger immune responses

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Vaccination is a remarkable medical achievement, but many vaccines elicit poor responses, which limits efficacy. Exercise has been identified as a possible behavioural adjuvant; brief muscle damaging exercise immediately prior to vaccination enhances antibody responses with effects mostly confined to strains showing weaker control responses [1,2]. To test the hypothesis that exercise enhances weaker, but not stronger immune responses, we tested the effect of exercise on the response to either a full- or half-dose Pneumococcal (Pn) vaccination. The exercise task was developed for clinical applicability, minimizing equipment, and inducing less muscle damage.

Subjects were 132 young healthy adults (75 women; age: 22±2.7years; BMI: 23±3.8Kg/m^2), who were randomized to one of four groups: Exercise or control task, receiving a full- or half-dose Pn vaccination. Prior to vaccination, exercise groups completed a 15min task using resistance bands involving 30s of arm and shoulder exercise and 30s rest alternations. Control subjects rested quietly during this time. Antibody levels to 10 Pn strains were evaluated at baseline and 1 month.

To assess overall effect of exercise, a multivariate ANOVA was performed with change scores (1 month–baseline) for all 10 Pn strains, controlling for gender and race. A significant effect of group showed an overall greater change in antibody levels among all strains in the exercise groups ($F(10,86)=1.94, p=.05, \eta^2=.18$). We then compared the effect of exercise in full-dose and half-dose responses, and found that the exercise group had significantly larger responses in the half-dose group ($F(10,37)=2.12, p=.035, \eta^2=.36$), but in the full-dose groups there were no differences.

The current data showed an overall effect of enhanced response among participants who completed the exercise task prior to vaccination compared to resting controls. Importantly, this effect was significant in the half-dose group, while no differences were seen in the full-dose group. The current study adds to data indicating the effectiveness of exercise as a vaccine adjuvant, particularly in weaker responses. Our model of weaker responses provides evidence that testing in at risk populations should be pursued given the public health benefits of no-cost behavioural approach to give greater levels of protection to those most at risk.

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O.9. Antigen-stimulated monocyte and T-lymphocyte activation and systemic inflammatory cytokine concentration in chronic kidney disease: effect of regular moderate intensity aerobic exercise (ECRA)

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Introduction: Chronic kidney disease (CKD) patients suffer from very high cardiovascular risk associated with chronic systemic inflammation. Regular exercise has been shown to exert anti-inflammatory effects in various disease states, but its role in modifying inflammatory status in CKD is unclear. The aim of this study was to investigate the effects of 6-months of regular moderate intensity aerobic exercise on activation markers in superantigen-stimulated monocytes and T-lymphocytes and systemic inflammatory cytokine levels in CKD patients.

Methods: Eighteen CKD3-5 pre-dialysis patients (11M, 7F), age 62±8 years, exercised for 30 min, 5 times/week, for 6-months. The exercise programme consisted of brisk walking at a Rating of Perceived Exertion (RPE) of 12-14. A further 14 CKD3-5 pre-dialysis patients (8M, 6F) age 56±15 years, acted as non-exercise controls. Renal function was comparable between groups (eGFR, ml/min/1.73m^2: exercise, 25.3±7.9, control, 29.3±5.5). Resting venous blood samples were collected at baseline and after 6-months. SEB-activated monocytes ($CD14^+CD86^+HLA\text{-}DR^+$) and T-cell subsets ($CD4^+CD69^+$ and $CD8^{++}CD69^+$) were identified by flow cytometry. Plasma interleukin (IL)-6 and IL-10 were measured by ELISA.

Results: The exercise intervention did not affect weight or BMI. Average RPE response to a standardised 30 min treadmill walking exercise test after the training period was lower than at baseline in exercisers only ($P<0.05$). After 6-months, CD86 and HLA-DR expression by $CD14^+CD86^+HLA\text{-}DR^+$ monocytes was down-regulated in exercisers (CD86 $P=0.004$, HLA-DR $P=0.037$), but up-regulated in controls ($P=0.018$ and $P=0.043$). Expression of CD69 by both $CD4^+CD69^+$ and $CD8^{++}CD69^+$ T-cells was down-regulated in exercisers (CD4 $P=0.017$, CD8 $P=0.010$), but did not change in controls ($P=0.912$ and $P=0.266$). Plasma IL-6 showed a tendency to reduction and IL-10 to increase in exercisers (IL-6 $P=0.060$, IL-10 $P=0.074$), but was unaltered in controls. Plasma IL-6/IL-10 ratio, reflecting overall inflammatory status, was reduced in exercisers ($P=0.024$), but unchanged in controls (P=0.117).

Conclusions: We found that regular moderate aerobic exercise down-regulated activation markers in stimulated monocytes and both CD4 and CD8 T-cells, and this was associated with an improvement in the ratio of pro-inflammatory IL-6 to anti-inflammatory IL-10 cytokine levels in systemic circulation. These effects indicate that participation in a regular exercise programme can improve systemic inflammatory status and reduce cardiovascular risk in these very vulnerable patients.

O.10. Exercise influence on wound healing and tissue inflammation in obese high-fat diet-fed mice (ECRA)

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Introduction: Impaired wound healing remains a major health concern for obese and diabetic individuals. Studies have demonstrated that prolonged inflammation in the wound tissue can cause delays in wound healing in these population. Therapies which dampen the inflammatory response are therefore of great interest to researchers. Previous studies have shown that exercise can speed healing and reduce wound inflammation in aged populations. We hypothesized that exercise in obese high-fat diet-fed mice would speed wound healing and reduce wound tissue inflammation as measured by inflammatory gene expression.

Methods: Mice were fed a 45% kcal from fat diet for 16 weeks, then treadmill exercised for 3 days prior and 5 days after application of a full-thickness dermal punch biopsy wound. Wounds were photographed daily and assessed for wound area via photoplanimetry. In a separate study, wounds were harvested from mice at 1, 3, or 5 days post-wounding and assessed for inflammation via RT-PCR analysis of inflammation-related gene expression.

Results: Exercise sped wound healing rate in obese mice compared to sedentary controls over the first 5 days post-wounding ($p=.05$). At day 12 post-wounding, a significantly greater percentage of mice from the exercise group were fully healed compared to those of the control group ($p=0.009$). Although wound inflammation was not statistically different ($p>.05$), exercise training resulted in an approximately 2-fold induction in the gene expression of F4/80, IL-10, and TNF-alpha in the wound tissue at day 1 post-wounding, with no differences at later time points.

Conclusion: Exercise sped wound healing rate in obese mice. Contrary to our hypothesis, inflammation in exercise mice tended to be higher at 1 day post-wounding, suggesting that exercise may be speeding the inflammatory process rather than reducing it, thereby shortening the transition time from inflammation to tissue remodeling and increasing healing rate.

Orals: Immunodepression and Intense Exercise: Evidence and Strategies for Prevention

O.11. Imunoglukan P4H® and respiratory tract infections in elite athletes

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Prolonged and exhausting physical activity causes numerous changes in immunity and increases risk of upper respiratory tract infections (URTIs). Nutritional supplements as countermeasures to exercise induced changes have increasingly been studied in the last decade. One of the most promising nutritional supplements is β-glucan, a well-known immunomodulator with positive effects on function of immunocompetent cells.

In this study, we investigated the effect of pleuran, insoluble β-glucan from mushroom *Pleurotus ostreatus*, on selected cellular immune responses and incidence of URTI symptoms in athletes. Fifty elite athletes were randomized to β-glucan or placebo group, taking 200 mg of β-glucan (commercial name Imunoglukan®) or placebo supplements during 3 months. Venous whole blood was collected before and after 3 months of supplementation and after additional 3 months without supplementation. Incidence of URTI symptoms together with characterization of changes in phagocytosis and natural killer (NK) cell count were monitored during the study.

We found that β-glucan significantly reduced the incidence of URTI symptoms and increased the number of NK cells. In addition, β-glucan significantly improved phagocytosis process, whereas phagocytosis remained stable in placebo group.

These findings indicate that β-glucan may serve as an effective nutritional supplement for athletes under heavy physical exertion. Additional research is needed to determine the mechanisms of β-glucan function.

O.12. The effect of acute bovine colostrum supplementation on neutrophil responses to prolonged cycling (ECRA)

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Bovine colostrum (BC) supplementation for periods of 4-12 weeks has been shown to reduce the magnitude of, or speed recovery from, exercise-induced immunodepression [1]. The purpose of this study was to identify whether acute BC supplementation prior to a bout of prolonged exercise has any effect on neutrophil function.

Seven healthy males (age: 23.3 ± 3.9 years; mean ± SD) participated in 2 main trials in a randomised order. Subjects consumed either BC or placebo 1 hour prior to 2.5 hours of cycling at approximately 55% VO_2 max (30 g), immediately prior (5 g) and midway through the exercise (5 g). Venous blood samples were obtained prior to consumption of the supplement (BAS), 1 hour post-drink (immediately pre-exercise: PRE), immediately post-exercise (POST) and 1 hour post-exercise (1-POST). Neutrophil counts were measured using an automated haematology analyser. In-vitro stimulated neutrophil oxidative burst responses (OBA) to PMA and fMLP were measured by chemiluminescence (CL) assay and expressed per neutrophil.

Repeated measures 1 way ANOVA and post hoc paired t-tests (Bonferroni corrected) revealed significant increases at both post-exercise timepoints for blood neutrophil count ($P < 0.01$). For fMLP-stimulated OBA, 2-way repeated measures ANOVA revealed a main effect of time ($P < 0.001$) and a trend for a main effect of trial ($P = 0.068$) but no time × trial interaction ($P > 0.05$). For PMA-stimulated OBA, there was a main effect of time ($P = 0.01$) but no main effect of trial or time × trial interaction ($P > 0.05$). Post hoc comparisons demonstrated significant decreases below BAS at POST ($P < 0.001$) for PMA-stimulated OBA and at POST and 1-POST for fMLP-stimulated OBA ($P < 0.05$). These results suggest that fMLP-stimulated OBA is generally higher with acute BC supplementation but the overall temporal pattern (a post-exercise decrease) is similar between trials.

These preliminary results show trends to support the idea that BC may enhance neutrophil functions by a direct and immediate mechanism, in agreement with findings from in-vitro studies [2]. At present statistical power is low and the intention is to increase the sample size to 12.

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O.13. Comparison of immunologic profile in sera and nasal mucosa in athletes after a marathon (ECRA)

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Introduction: The high prevalence of upper respiratory infections after strenuous efforts is still accepted by many researchers as a systemic condition of immunodepression, explained by theories as "the open window" and "J shaped curve". Recently it was shown that these clinical conditions formerly viewed as infections, in fact encompasses also allergic and inflammatory processes of the upper airways. We studied the immunological responses in sera and upper respiratory mucosa after a marathon to compare the pattern of responses.

Methods: We analyzed the effect of marathon in immunological profile of serum and in the mucosal of upper respiratory airways. The group of marathon runners was composed by 18 male runners from São Paulo, with a range of age of 25 to 60 years (mean 41.4). We evaluated levels ($p<0.05$) of interleukins (IL) 6, 8 and tumor necrotic factor (TNF)-alpha (pg/mL) in serum and protein extract obtained from total lysate of nasal cells, in three moments: at rest, immediately and 72 hours after race.

Results: We observed that levels of IL-6= 1.3(0.48-4.87), IL-8= 2439(476.2-6290) and TNF-α = 2.18(0.41-9.12)] in extract of nasal cells were different of the concentrations of IL-6= 8.56(0.28-46.58), IL-8= 26.68(14.09-48.84) and TNF-α= 3.57(1.95-6.96)] obtained in serum at rest. The results observed immediately after a marathon showed that the levels of IL-6= 7.99(4.76-14.28) in nasal extract were statistically higher than basal levels. However, IL-8 [1020(771.4-2602)] and TNF-α [1.02(0.42-2.72)] levels in extract of nasal cells were reduced didn´t show significant difference. The analysis of the concentrations of IL-6 [95.74(51.22-163.2)], IL-8 [75.49(38.28-129.8] and TNF-α [19.22(4.8-51.09] in serum showed significant increase in comparison to at rest levels. The analysis of results obtained 72 hours after a marathon, shows that the levels of IL-6 [10.93(2.78-17.23)] and IL-8 [5435(3611-7764)] in nasal cells extract were statistically increased compared to at rest levels. Furthermore, the IL-8 and TNF-α [3.33(1.5-11.61] levels obtained from total lysate of nasal cells showed significant increase in relation to concentrations observed immediately after the race. In serum of athletes the IL-6 [9.92(0.28-33.5)], IL-8 [37.26(23.1-56.1)] and TNF-α [2.05(1.95-3.1] levels were statistically reduced in comparison to concentrations immediately after the race, showing a return to basal levels.

Conclusion: Responses in sera and upper respiratory mucosa are quite different and the research of the upper airway disease of the athlete must include the study of upper respiratory mucosa to obtain a complete view of the ongoing immunological process that occurs after a marathon.

Orals: Immunological Impact of Exercise and the Stress Response

O.14. Intensive resistance exercise induces apoptosis in lymphocytes via cortisol and glucocorticoid-receptor dependent pathways (ECRA)

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Introduction: Resistance training is an integral part of athletes training to develop discipline specific exercise performance. In order to stimulate regular adaptation toward specific training goals, progressive and intensive resistance training (RT) protocols are necessary. Intensive endurance exercise is known to induce lymphocyte apoptosis which might affect immune function [1]. Less is known about the effects of intensive resistance exercise on apoptosis and its underlying mechanisms.

Methods: 15 male subjects (age [years] 26.86 ± 1.01, weight [kg] 78.92 ± 2.81, BMI [kg/m²] 24.33 ± 0.79) performed a whole body resistance training program including 8 different exercise sets in 3 series. RT intensity was defined individually as 75% of the 1RM. Exercise protocol was performed with 2 min breaks between each session and 3 min breaks between each series. Total duration of the exercise tests was about 90 min. Blood was taken before, after, 3h and 24h after RT. Analysis of apoptosis-related surface markers, activation status and mitochondrial membrane potential (MMP) were performed by flow cytometry. Serum cytokines were quantified by ELISA.

Results: RT induced a significant increase of lymphocyte apoptosis 3h after exercise which was accompanied by a significant decrease of MMP, a reduction of Bcl-2 and an up-regulation of CD95. Blood lactate, IL-6, CRP and cortisol increased significantly 3h after RT. A significant correlation was observed between the increase of apoptosis and cortisol levels 3h after RT. Incubation of freshly isolated lymphocytes in serum taken 3h after RT (RT serum) indicated an important role of serum correlates for apoptosis induction. In addition, incubation in RT serum slightly increased CD95 expression. Incubation of lymphocytes in concentrations of selected serum parameters corresponding to levels found post in RT serum demonstrated a major role for cortisol in apoptosis induction. This result was confirmed by attenuation of apoptosis after addition of mifepristone before incubation in RT serum.

Conclusion: In summary, resistance exercise induced lymphocyte apoptosis via signalling of serum correlates. In this context, cortisol signalling via glucocorticoid receptors might be an important mechanism for lymphocyte apoptosis after resistance exercise.

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Orals: Technical Advances and Novel Approaches in Immune Assessment

O.15. Development and validation of an oral fluid collection device and its use in the immunoassay of salivary steroids and immunoglobulins in sports persons

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Introduction: Saliva has become acceptable as a diagnostic fluid to monitor health and disease. Its collection is non-invasive and does not require experienced personnel, supply is not limited and analyte levels broadly reflect the levels of free (active) analytes in blood. Saliva can be collected by drooling or spitting into a container which provides a neat sample. However, this is socially unacceptable, may take a long time to collect adequate volume and the collected samples have to be frozen to avoid bacterial growth. Saliva may also be collected using a cotton or polyester based swab. A centrifugation step is required to extract the saliva and there are issues with recovering the analytes from the swab material. Our aim was to develop a saliva collection device suitable for use with real-time (on-site) immunoassay tests for immunoglobulins and hormones. The collection has to be fast, collected volume known and analyte recovery adequate.

Methods: The device consists of a synthetic polymer-based swab material attached to a volume adequacy indicator stem and a dropper bottle with extraction buffer. The indicator stem changes colour upon the collection of about 0.5mL of saliva. The swab is placed in the dropper bottle containing known volume of extraction buffer. The bottle is shaken for 30-60 seconds and the sample is ready for on-site testing or to be sent for laboratory testing. The device was used to collect saliva from 50 volunteers recording both collection time and volume. Enzyme immunoassays were used to determine the recovery of IgG, IgA, cortisol, testosterone and DHEA from saliva collected using the device versus saliva collected by drooling.

Results: The average collection time (as indicated by the colour change) was 27.7 seconds (STDEV: 8.47, range: 19.3-52 seconds) and collection volume was 0.55mL (STDEV: 0.06, range: 0.42-0.64 mL). Analyte recovery following 1 minute shaking was over 85%.

Conclusion: The oral fluid collection device described here can be suitable for the collection and testing of saliva using on-site real time tests.

Orals: Age and Gender Issues in Exercise Immunology

O.16. Ex vivo cytokine production following exhaustive exercise differs in male and female athletes (ECRA)

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Study of the cytokine production from whole-blood cultures incubated with bacterial polysaccharide (LPS) is a method of investigating the capacity of leukocytes to produce cytokines. Sixteen well-trained athletes (8 male and 8 female, six of them in luteal phase of their menstrual cycle) participated in a half marathon (HM). Venous blood samples were collected from each subject before (t0), 30 min after (t1), 3h after (t2) and 24 h after (t3) the run. EDTA-blood of the athletes was incubated with or without LPS (10ng/ml final concentration) for 1hour, and cytokine concentration in supernatants measured by bead-based multiplex immunodetection assay.

Concentrations of IL-10, IL-1ra, IL-6, IL-8, and TGF-β1 rose significantly at t1 in controls as well as in stimulated supernatants for both sexes, and were still above prerace levels at t2. LPS-stimulated production (Δ of LPS values minus controls) of TNF-α was significantly suppressed for both sexes at t1 as compared to t0, while significant increases were observed for IL-8, IL-1ra, and TGF-β1. These findings were even accentuated when the results were corrected for number of monocytes in the blood. LPS-stimulated production of IL-6 showed a significant increase in men, but a mild non-significant decrease in women at t1.

Men showed significantly higher LPS-stimulated IL-1ra, IL-6 and IL-8 and lower TGF-β1 after exercise as compared to women, who were in luteal phase of their menstrual cycle. This was also confirmed when cytokine production was corrected per monocyte numbers. The concentrations of IFN-γ, IL-12p40, and IL-12p70 remained near pre-race levels.

The present study demonstrates that the cytokine response to LPS is dramatically changed in blood samples drawn 30 min or 3h post exercise with massive suppression of TNF-α production, enhancement of IL-1ra in both sexes, and sex-dependent enhancement of TGF-β1 and IL-8. Taken together, these results confirme the anti-inflammatory impact of acute exercise and suggest that this response is different in women in luteal phase as compared with men.

O.17. Cytomegalovirus serostatus increases the proportion of highly differentiated CD8+ T-cells at rest and following acute exercise but the response is influenced by gender and training status (ECRA)

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Highly differentiated CD8+ T-cells (i.e. CD28-/CD57+) are preferentially mobilised in response to acute exercise, before rapidly extravasating the blood compartment after exercise cessation [1,2]. This response is amplified in those carrying a latent CMV infection [3]. We have recently observed greater highly differentiated CD8+ T-cell mobilisation in response to acute exercise in untrained vs. trained, and in males vs. females; however, it is not known if these effects were due to underlying CMV serostatus.

Purpose: To determine the effect of CMV serostatus on highly differentiated CD8+ T-cell mobilisation after acute exercise in trained and untrained males and females.

Methods: Sixteen (8 male, 8 female) trained soccer players aged 18.3 (±1.7) years and sixteen (8 male, 8 female) untrained but healthy active controls aged 19.3 (± 2.0) years performed an incremental treadmill running test to volitional exhaustion. Blood obtained before, immediately after and 1 hour after exercise was analysed for CMV serostatus by ELISA and blood lymphocytes were assessed for cell surface co-expression of CD28 and CD57 on CD3+/CD8+ T-cells by four colour flow cytometry.

Results: The incidence of CMV was significantly ($P<0.05$) greater in trained vs untrained (56% seropositive vs. 25% seropositive) but was similar between genders (44% vs. 38%, females vs. males). The proportion of CD28-/CD57+ CD8+ T-cells was influenced by CMV status ($P<0.01$), training status ($P<0.05$) and gender ($P<0.01$) but there was no interaction effect with response to exercise. There was a greater proportion of CD28-/CD57+CD8+ T-cells in the CMV+ group compared to the CMV- group at all time points, a greater proportion in the untrained CMV+ compared to the trained CMV+, and a higher proportion in CMV+ males compared to CMV+ females.

Conclusion: There is a greater incidence of CMV in trained soccer players compared to age-matched controls indicating that CMV may be more easily transmitted in team sport environments. Despite lower CMV incidence, untrained subjects had a greater proportion of highly differentiated CD8+ T-cells at rest and in response to exercise, indicating that training status affects the frequency of these cells independently of CMV serostatus. Similarly, the lower proportion of highly differentiated CD8+ T-cells in females was not due to differences in CMV serostatus, suggesting a possible hormonal influence that warrants further investigation.

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O.18. The validity of plasma heat shock protein 72 as a biomarker of sarcopenia

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Heat shock protein 72 (Hsp72), an intracellular chaperone, has been identified in the extracellular milieu, where it is clear that eHsps play a role as pro-inflammatory immune effectors [1-3]. There is an age-related decrease in eHsp72, but centenarians are an exception in that they have increased eHsp72, suggesting that higher eHsp72 leads to healthy outcomes later in life. The present study was designed to assess the plasma levels of eHsp72 in elderly people, and to investigate its potential interaction with elderly-specific syndromes.

A total of 665 men and women participated in an official medical health examination, including a physical fitness test. There was a significant difference in the sex ratio (male, n=264; female, n=356, $P<0.001$), but not a significant difference in mean ages between the sexes (male, 73.5 ± 6.0 years; female, 73.4 ± 6.3 years). Enzyme-linked immunosorbent assay (ELISA) kits were used to measure the plasma concentrations of Hsp72. Serum C-reactive protein (CRP), interleukin (IL)-6, tumor necrosis factor (TNF)-α, β_2-microglobulin (β_2-MG), cytomegalovirus antibody titer (CMV-IgG), and herpes simplex virus antibody titer (HSV-IgG) levels were measured.

Significant increases in age, log TNF-α, and β_2-MG, and decreases in height, weight, muscle volume, grip strength, and hemoglobin were observed with the highest tertile of eHsp72. Height, weight, skeletal muscle volume, grip strength, walking speed, HSV-IgG, and hemoglobin were associated with lower eHsp72 (negative correlation). Higher eHsp72 levels in patients with stroke (cerebral infarction), myocardial infarction, and cancer were found compared to those in the healthy control. To correct for the influences of these diseases on eHsp72 levels, we adjusted for sex, age, and the incidence of the diseases, and found that the odds ratios of eHsp72 tertiles on skeletal muscle mass, grip strength, and walking speed tertiles were significantly independent.

Our results revealed that eHsp72 in plasma is linked to sarcopenia and/or is a predictor of frailty. On the other hand, eHsp72 was not associated with biomedical parameters such as cholesterol, HbA1c, and creatinine, or with psychological factors including the geriatric depression scale, self-rated health scale, and mini-mental state examination.

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Abstracts of Poster Communications

Posters: Exercise, metabolism and inflammation

P.1. Effects of vitamin C supplementation on the IL-6 and IL-10 response to a 15 km-run competition

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Exercise induces increases in plasma levels of both IL-6 and IL-10 [1]. It has been reported that IL-10 increase is induced by increased levels of IL-6 [2]. Furthermore, it has been suggested that reactive oxygen species and oxidative stress could be involved in the increased expression on IL-6 (probably activating factor nuclear □□) [3]. The aim of this study was to determine the effects of vitamin C supplementation on plasma and blood mononuclear cell IL-6 and IL-10 levels in response to exercise.

A double-blinded study of supplementation with vitamin C was performed. After 15 days of supplementation with vitamin C (500 mg per day, n=17) or a placebo (n=17), athletes participating in the study completed a 15-Km run competition. Blood samples were taken before competition, immediately after finishing the competition and two hours after finishing the competition. Concentrations of MDA and lipid hydroperoxides were measured in plasma. Vitamin C, IL-6 and IL-10 levels were measured in plasma and in blood mononuclear cells. Furthermore, IL-6 and IL-10 mRNA levels in blood mononuclear cells were measured by Real-time RT-PCR.

Vitamin C supplementation induced higher plasma and mononuclear cell vitamin C levels. Both oxidative markers, MDA and lipid hydroperoxides, increased after the competition, but no differences were found between groups. Similar increases in IL-6 and IL-10 plasma levels were observed after the competition in both the supplemented and the placebo groups. When IL-6 and IL-10 levels were analysed in mononuclear cells, the highest levels were found two hours after finishing the competition, with similar values in both groups. IL-6 mRNA levels in mononuclear cells increased after the competition and returned to basal levels after two hours recovery. However, IL-10 mRNA levels increased after the competition but remained high two hours after finishing the competition. No differences between groups were observed in these mRNA levels. The lack of differences between groups regarding plasma and mononuclear cell IL-6 and IL-10 could be explained by the fact that, in this study, vitamin C supplementation did not prevent oxidative stress.

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P.2. Habitual physical exercise improves fungicidal capacity of macrophages in the obese Zucker rat model of the metabolic syndrome

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There is a causal relationship between obesity-associated diabetes and an increased risk of infection. The obese Zucker rat (fa/fa) is the most commonly used animal model for the study of human type-II diabetes mellitus, and this animal provides an appropriate model for studying the relationship between immune dysfunction and obesity-associated type II diabetes [1-3]. As reported in obese humans, obese Zucker rats are also more susceptible to candidiasis than control lean Zucker rats [1]. Since it is well known that exercise improves the phagocytic and microbicidal capacity against *C. albicans* in healthy animals [4], the purpose of the present investigation was to evaluate the effect of a program of habitual exercise (treadmill running: 5 days/week for 14 weeks reaching 35 cm/s for 35 min in the last month) on the phagocytic and fungicidal activities of peritoneal macrophages from obese rats.

Results indicated that phagocytosis of *C. albicans* was similar between peritoneal macrophages from obese and lean rats. However, a significant depression in the fungicidal capacity of macrophages from obese rats was observed as compared to that measured in macrophages from lean rats. The exercise training program clearly improved the deteriorated fungicidal activity of macrophages from the obese rats, raising values after the habitual exercise in levels even higher than those found in the sedentary lean rats.

In conclusion, habitual physical exercise or training improves the deteriorated fungicidal capacity of peritoneal macrophages associated with metabolic syndrome. Thus, exercise can contribute to ameliorating the risk of infection by candidas that is associated with obesity.

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P.3. The impact of a pre-load bout of exercise on IL-6 response and subsequent time trial performance (ECRA)

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Interleukin-6 (IL-6) is a pluripotent cytokine that has a role in a range of actions including glucose regulation during exercise [1] and also the sensation of fatigue [2]. Whilst it is widely acknowledged that prolonged exercise is associated with increases circulating IL-6 concentrations [3], the influence of this response on exercise performance is unknown. The aim of this study was to determine whether elevations in IL-6 induced by a preload bout of exercise influenced subsequent time trial performance.

Endurance trained males (n=23, age 27 ± 6 yr, VO_{2max} 57.6 ± 4.0 ml·kg·min^{-1}) performed a preloaded time trial where participants ran at 60% vVO_{2max} for 2 h interspersed with 30 s at 90% vVO_{2max} every 10 min, followed by a 5 km time trial. Blood samples were drawn at baseline, following the 2 h preload bout and post time trial and blood plasma was analysed for IL-6, sIL-6R and cortisol using ELISA techniques.

Our results indicate that plasma sIL-6R (r-0.04, p=0.83), cortisol (r-0.08, p=0.73,) and glucose (r 0.13, p=0.55) were not associated with relative time trial performance, however, the plasma IL-6 response to the preload bout of exercise was inversely correlated with subsequent relative time trial performance. Multiple linear regression demonstrated that the plasma IL-6 response had a greater influence on subsequent relative time trial performance ($p<0.05$) compared with habitual training distance and VO_{2max} ($p>0.05$).

We conclude that the circulating levels of IL-6 as a consequence of a preload exercise bout may significantly influence subsequent relative time trial performance independent of training status.

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P.5. Impairment of iron metabolism in young tennis players as a results of low grade systemic inflammation

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Inflammation arising from various etiologies, including infection, autoimmune disorders, chronic diseases, and aging, can promote anemia [1-3]. The purpose of the present study was to verify whether training in young athletes induces low grade systemic inflammation and if the inflammation has influence on ferritin level and hemoglobin synthesis. The problem of iron deficiency has a particular meaning among young athletes training tennis, since their professional path requires participation in many tournaments involving change of climatic and time zones. The constant demand for adaptation makes the preparation training more difficult.

Groups of well-trained young tennis players (15 ± 0.9 years old, average of 9 years training experience) took part in this study. The research was performed in 6-month intervals. The study was approved by the hosting university's Research Ethics Board. Serum inflammatory cytokines (IL-1β, TNFα, IL-6, IL-10) were measured by commercially ELISA kits and hematological measurements by conventional methods.

The data in groups A and B shows that pro-inflammatory cytokines TNFα and IL-1β were elevated in most of the athletes when compared to normal values reported for professionals. Moreover, statistically significant inverse correlation between pro-inflammatory cytokine IL-1β and blood hemoglobin ($r = -0.60$), mean corpuscular hemoglobin ($r = -0.64$) or mean corpuscular hemoglobin concentration ($r = -0.69$), was observed. In both periods of measurements an inverse correlation between IL-1β and blood ferritin has been observed.

We concluded that long lasting tournament season and professional training may cause the low grade systemic inflammation in young tennis players and thus contribute to the lower level of ferritin and disturb the synthesis of hemoglobin. Hence, rest and appropriate diet should be a part of specific training program for young tennis players in order to prevent of low grade systemic inflammation and avoid applying of iron supplementation.

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P.6. Plasma YKL-40 increases with exercise (ECRA)

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Plasma YKL-40 concentration is increased in conditions characterized by inflammation i.e. bacterial infections, inflammatory bowel disease, diabetes, coronary artery disease and several different cancers. YKL-40 has been suggested as a biomarker of disease severity. Very little is known regarding YKL-40 and exercise. Several studies have shown that plasma levels of interleukin-6 (IL-6) increase during an exercise bout, suggesting a link between exercise, IL-6 and immune modulation. We have previously shown that IL-6 infusion stimulate an increase in plasma YKL-40 level, suggesting that IL-6 is a key regulator of YKL-40 during the inflammatory response. Since plasma IL-6 increase during exercise we hypothesized that plasma YKL-40 would also increase during exercise, maybe as a result of increased IL-6 level. Furthermore, since the IL-6 response is attenuated with repeated exercise bouts we hypothesized that a possible acute YKL-40 response would diminish correspondingly following a period of regular training.

Using material from healthy male subjects, we measured plasma YKL-40 during 3 hours bicycling exercise bout obtaining samples well into recovery. Plasma YKL-40 increased continuously throughout the exercise from ~32 ng/ml to ~45 ng/ml. One hour into recovery plasma YKL-40 levels had returned to baseline levels. To assess the effect of repeated bouts of exercise we measured plasma YKL-40 from healthy subjects before and after a 12 week period of 5 exercise bouts per week. The plasma YKL-40 level was increased with ~60 % in the subjects that had been performing regular endurance training to a basal level of ~50 ng/ml. Interestingly, a single 3 hours bicycling exercise bout following the training period increased plasma YKL-40 additionally from ~50 ng /ml to ~66 ng/ml, showing the same kinetics as before the training period.

In conclusion we show that plasma YKL-40 increases continuously during a single exercise bout. Furthermore, regular training increases plasma YKL-40. Though IL-6 increases during exercise, the increase in plasma YKL-40 does not seem to be mediated through IL-6, since the IL-6 response, but not the YKL-40 response, is diminished with repeated regular bouts of exercise.

P.7. Effects of resistance training on serum cytokine and CRP levels in females

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Introduction: Exercise or physical activity is a potent activator of the immune system resulting in changes in the pro-inflammatory marker and cytokine concentrations [1].That is related to exercise intensity, duration, the mass of muscle recruited, and one's endurance capacity [2-5]. Exercise and physical strenuous activity have been demonstrated to increase the serum TNF-α and IL-6 concentrations. The effect of resistance training on serum cytokine and CRP levels in female has been less examined and is unknown. We assessed serumIL-6, TNF-α and CRP concenterations in subjects who performed short-term circuit resistance training.

Methods: Healthy female University students were randomly assigned to four groups; active experimental (AE, n = 8) active control (AC, n= 8) inactive experimental (NE, n= 13) and inactive control (NC, n = 14). Training consisted of, 5 times per week for 2 weeks with free weights and weight training machines including: chest press, leg extention, sit-up, lat. pull down, front row, foot raising, back extention, and leg curl. In the first week the training sessions were done at 40% 1RM, 15 repetitions and 3 sets. The intensity of training was increased to 50% 1RM during the second week, but other properties of training remained constant. Before and 48 hours after the last training session, fasting blood samples were collected at rest to measure serum IL-6, and TNF-α concentrations by ELISA methods and CRP (H-S) by an immunoturbidometric method.

Results: A within group comparison showed: IL-6 concentration in all groups and TNF-α concentration with the exception of the inactive experimental group (which was unchanged) were reduced and CRP increased in the active experimental group and did not change in the inactive group. In between group comparisons revealed that there were significant differences in TNF-α and IL-6 (both $P \leq 0.05$) but not in CRP.

Conclusions: Strenuous exercise induces an increase in the serum levels of pro-inflammatory cytokines such as TNF-α and IL-6. However, it seems that short-term circuit resistance training can reduce inflammatory condition and possibly the risk of infection.

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P.8. Effects of dietary restriction or swimming on metabolism and function of lymphocytes and macrophages from old rats

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Although aging compromises the functionality of macrophages (MØ) and lymphocytes (LY) and dietary restriction (DR) and exercise partially counterbalance immunesenescence [1,2], it is unknown the effects of both strategies on the metabolism of these cells which metabolism is strictly related to their function. Additionally, as a main effect of DR and exercise is body weight reduction and adipose tissue physiology vary in a deposit-specific manner, we investigated if body composition changes are associated with the effects of both strategies on MØ and LY.

Thus, rats were randomically distributed in young controls (C), older submitted to 50% of dietary restriction (ODR) and older submitted to swimming (EX). The function of immune cells (proliferative index, phagocytic capacity and H_2O_2 production), the weight of adipose tissue depots and protein contents of lymphoid organs (thymus and spleen), plasma glutamine concentration, interleukins (IL-1, IL-2, IL-6) and, immunoglobulins (IgA and IgG) were analysed.

DR increased glucose utilization in LY and reduced the plasma levels of anabolic hormones while increasing IL-2. Swimming training stimulated MØ phagocytosis, glutamine metabolism in these cells and increased brown adipose tissue.

These data suggest that DR and exercise affects differentially MØ and LY metabolism and function and that the actions of each strategy seem to be related to its particular effects on body composition, cytokines and hormones.

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P.9. Changes in cytokines and leucocyte activation after low- and moderate- intensity exercise during menstruation (ECRA)

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Previous studies indicate differences in immune function between genders, and between follicular phase and luteal phase [1-3]. We recently demonstrated that exercise-induced changes in cytokines and inflammatory responses are affected by the different phases of the menstrual cycle. Leucocyte activation (measured by plasma calprotectin concentration) was observed following exercise at the intensity of 75% ventilatory threshold (VT) only in the menstrual phase. Therefore, strenuous endurance exercise at the intensity of higher than 75% VT may accentuate stress and inflammatory responses, especially in the menstrual phase. Considering the increasing number of women who exercise to improve their physical fitness, it is important to assess changes in inflammatory markers following exercise during menstruation. The aim of this study was to examine the influence of exercise intensity on cytokines and leucocyte activation during menstruation.

Seven healthy sedentary females completed three separate trials: (1) a 60-min cycling at 75% VT (low-intensity), (2) a 60-min cycling at 100% VT (moderate-intensity) and (3) a resting state without exercise (control). Blood was sampled before, immediately after and 30 min after exercise. We measured the concentrations of plasma cytokines (interleukin (IL)-6 and IL-8) and markers of leucocyte activation (calprotectin and myeloperoxidase) using ELISA.

Aerobic exercise during menstruation increased the circulating concentration of IL-6 and calprotectin. Plasma IL-6 and calprotectin responses following exercise were greater after moderate-intensity than after low-intensity trials. A positive correlation was found between exercise-induced changes in IL-6 and calprotectin.

These findings suggest the possibility that inflammation and leucocyte activation are more closely associated with exercise intensity of the exercise prescription level during menstruation. This knowledge is potentially useful for prescribing safe exercise guidelines that minimize the risk of negative health outcomes during different phases of the menstrual cycle.

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P.10. The comparison of circulating cytokine responses depending on exercise intensity and their biological and pathological significance

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Circulating levels of IL-6, IL-1ra, IL-8 and IL-10 increase remarkably following endurance exercise such as marathon and triathlon, but not so in muscle-damaging eccentric exercise [1-5]. The present study was designed to investigate the effects of exercise intensity on kinetic changes in plasma concentrations of a wide range of cytokines to evaluate the magnitude in an exercise prescription level for health promotion. We also determined stress hormones and metabolic substrates, leucocyte activation and muscle damage markers to estimate the biological and pathological significance and the underlying mechanisms of the cytokine action.

Eight healthy male subjects completed three separate trials in a cross-over design: a 60-min cycling at 50% $VO_{2\ max}$ (moderate intensity), a 60-min cycling at 75% $VO_{2\ max}$ (high intensity) and a resting state without exercise. Blood was analyzed for total and differential leucocyte counts, glucose, FFA, lactate, catecholamines, cortisol, GH, Mb and CK. We measured the plasma concentrations of TNF-α, IL-1β, IL-1ra, IL-2, IL-4, IL-6, IL-8, IL-10, IL-12p70, IL-12p40, IL-17, MPO, calprotectin, IFN-γ, MCP-1, G-CSF and HSP70 using ELISA.

Immediately post-exercise, IL-6 and IL-8 responses were greater after high-intensity than after moderate-intensity trials. Plasma IL-1ra and IL-10 concentrations were greater 2 h after high-intensity trial than immediately after exercise. The leucocyte counts, plasma calprotectin and Mb concentrations increased depending on the exercise intensity. Plasma concentrations of catecholamines, cortisol and GH increased significantly immediately after exercise. FFA levels increased following high-intensity exercise, and lactate response was greatest for the high-intensity trial and significantly larger than that of the moderate intensity trial. As for the associations between measured parameters, IL-6 response was significantly correlated with neutrophil count and calprotectin concentration. Calprotectin was closely correlated with Mb.

These findings suggest that released IL-6 might promote leucocyte mobilization and activation, resulting in systemic acute inflammation and muscle damage after endurance exercise. Compared to the magnitude of changes of cytokines in the exercise prescription level, stress hormones as well as metabolic substances are more closely associated with exercise intensity, and are responsible for metabolic effectiveness of moderate exercise.

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P.11. Bed rest induced insulin resistance is highly reversible by exercise

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Objective: Insulin resistance is a major feature of type 2 diabetes and exercise is known to improve the peripheral insulin sensitivity.

Aim: In the present study we investigate the effect of an acute bout of exercise on bed-rest induced insulin resistance.

Methods: Twelve healthy males participated in the study and were divided in 2 groups. Before and after a 7 days bed rest trial, 6 subjects underwent a hyperinsulinemic euglycemic clamp combined with a glucose tracer [6,6-2H2]glucose and arterial-venous difference across both the leg and arm. Six subjects performed 45 min of one-legged knee extensor exercise before the hyperinsulinemic euglycemic clamp were commenced. 24 h after bed rest, with ample physical activity, another clamp measurement of insulin sensitivity where performed.

Results: All subjects became insulin resistant after 7 days of bed-rest with ~28% reduction of insulin mediated glucose uptake, ($P < 0.05$), which was located in skeletal muscle, whereas the liver was unaffected. The acute exercise bout reversed insulin mediated muscle glucose uptake only in the exercising leg, however the exercise mediated glucose uptake was reduced by bed rest. Surprisingly when the subjects where clamped 24 h after the bed-rest, the insulin sensitivity was increased 1.4 fold ($P < 0.05$) higher than before the bed rest.

Conclusion: These data demonstrate that bed rest induced insulin resistance is easily reversible and disappears after less than 24 h of normal daily activity in young healthy individuals. Whether such a finding will occur in patients with type 2 diabetes needs further investigation.

P.12. Liver SOD , TAC, TG, and CLO responses to endurance training with and without a mastic (Wild Pistachio) extraction in female rats

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Objective: The purpose of this study was to investigate the responses of liver superoxide dismutase (SOD), total antioxidant capacity (TAC), triglyceride (TG), and colesterol (CLO) in female rat liver to endurance training with or without ingestión of a Wild Pistachio (mastic) extract.

Methods: Twenty-eight adult female Wistar rats (6–8 weeks old, 131± 13.33 g) were randomly assigned to saline-control, saline-training, mastic-training and mastic-control groups. The training groups ran for 8 weeks (60 min/d, 5d/wk at 25 m/min and 0% grade). Animals were orally given either Wild Pistachio extract or an equal volume of saline for 4 weeks. Seventy-two hours after the last exercise session and 4 hours before the sacrifice food was removed but not tap water. A portion of liver was collected and frozen in liquid nitrogen for later analysis of liver SOD, TAC, TG, and CLO concentrations. An one-way-ANOVA was employed.

Results: Significantly lower SOD and TAC levels were found in mastic-trained groups when compared with control rats. Liver TG and CLO concentrations were significantly lower in the mastic-trained treated group when compared with the other groups.

Conclusion: Results demonstrated that 8 weeks of treadmill exercise with mastic improved liver SOD and TAC and prevented an exercise-induced elevation in TG and CLO concentrations. It seems that a liver antioxidant capacity improvment following training plus mastic treatment has impact on IL-6. This is in turn might confirm the role of pre-exercise liver antioxidant capacity level on the magnitude of IL-6 responses.

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P.14. Physical exercise and cardiovascular risk reduction in older adults

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Decreases on functional capacities in older people have important implications on the risk to develop cardiovascular disease. A relative risk (RR) of 70% for low fitness, higher than often referred for physical activity, and higher than for smoking (57%), hypertension (34%), high cholesterol (65%), or parental history (18%) was found [1]. The purpose of the study is to analyze the effects of a training programme on triglycerides, total cholesterol, HDL-C, LDL-C, and hs-CRP, BMI, and waist circumference.

Volunteered sixty three sedentary individuals (65-95 years old), randomized into two training groups and one control group. Venous blood samples were collected into EDTA containing tubes, after 12 hours fasting. Differences between evaluations were analyzed with an ANOVA for repeated measures ($p<0.05$ level).

After treatment, the exercising groups attained significant differences on BW, WC, BMI, DBP, TG, TC, HDL-C, LDL-C, TC/HDL-C relationship, and 6-minute walk distance, while the control group only had significant differences on WC. Positive correlations at baseline were attained for BMI with TC, and TG; for WC with TC, TG, and TC/HDL-C; and for BW with TC, TG, and TC/HDL-C.

Diminishing of the TC, LDL-C, TG, and TC/HDL-C with exercise is consistent with previous research [2]. Independent of the mechanism underlying lipid changes, a reduction of 1% on TC has been shown to reduce the risk for coronary artery disease by 2% [3], implying that our exercising participants have reduced about 12% their risk. A 1% reduction in LDL-C reduces the risk of major coronary events by approximately 2% [4], meaning that we have about a 26% gain. Moreover, a decrease of 1% on HDL-C has been associated with a 2-3% increase in the risk for CHD [5], and assuming that the reverse is true, the 5% increase observed in our both programs should decrease CHD by 10-15%.

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P.15. Essential amino acids attenuate the aerobic exercise-induced increase in skeletal muscle MCP1 in older adults (ECRA)

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Many diseases associated with aging are also linked to muscle loss and inflammation. This relationship may be further confounded by inadequate protein intake in older adults. Exercise training is recommended to reduce the risk of chronic disease, and may also have an anti-inflammatory effect on skeletal muscle. Monocytes are a potential source of inflammation in skeletal muscle. In cell culture, monocyte chemoattractant protein (MCP) 1 reduced anabolic signaling molecules in skeletal muscle. The purpose of this preliminary study was to examine the effect of essential amino acid supplementation on MCP1 and mammalian target of rapamycin (mTOR) signaling in human skeletal muscle after an acute bout of aerobic exercise.

Methods: Thus far, seventeen sedentary (4050±1241 steps/day) but otherwise healthy older adults (68-81 years) have participated in this randomized, double-blinded study. A baseline muscle biopsy sample was collected after a postabsorptive rest period. Subjects completed either an acute bout of treadmill walking (EX; 45 min. at ~70% of heart rate reserve) or rested in bed (REST). Immediately following the exercise or rest period subjects ingested either a leucine-enriched essential amino acid (EAA) or placebo (PLA) beverage. One hour and three hours post-beverage additional muscle biopsies were collected for immunoblotting procedures.

Preliminary results: MCP1 increased 179% in Ex+Pla but only 106% in Ex+EAA. Overall, MCP1 was higher (30%) in EX than REST, and lower (31%) in EAA than PLA. Phosphorylated mTOR increased 305% in EX+EAA and 37% in EX+PLA.

Conclusion: These preliminary data suggest that essential amino acid supplementation with aerobic exercise may attenuate skeletal muscle inflammation and prevent sarcopenia.

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P.16. Exercise training with and without body fat reduction: influence on inflammatory monocytes

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Regular exercise elicits anti-inflammatory effects, but whether this anti-inflammatory action can occur in the absence of body fat changes remains controversial. We previously found that resistance exercise training without measurable fat losses significantly lowered inflammatory monocyte percentage [1], but it is not known to what extent fat losses might contribute to the exercise-induced changes in monocyte phenotype.

Purpose: The purpose of this study was to examine the effects of resistance training, with or without energy restriction-induced body mass losses, on monocyte phenotype and monocyte expression of TLR4.

Methods: After baseline testing, physically inactive subjects (PI, N=40; males aged 45-56; females aged 50-75 and post-menopausal) were randomly assigned to 12 weeks of resistance exercise training (EX; n=12) or 12 weeks of exercise training with weight loss (EX-ER; n=14), with 26 PI subjects completing all training and post testing. There were nine physically active (PA) subjects who remained active for the duration of the study and served as a comparison group. Resting blood samples were obtained at baseline and after the 12-week intervention, at least three days after the last exercise session and 24 hours of a standardized diet. Monocyte phenotype (CD14+, "classical" monocytes; CD14+CD16+, inflammatory monocytes) and cell-surface expression of TLR4 were determined using flow cytometry.

Results: Inflammatory monocytes were significantly ($p < 0.05$) higher in EX and EX-ER compared to PA subjects at baseline. Inflammatory monocytes were significantly reduced after 12 weeks of exercise training in EX, but were not changed after the EX-ER intervention or in the PA comparison group. Further analysis of the EX group showed significant post-intervention reductions in inflammatory monocytes in overweight, but not obese subjects. TLR4 expression was lower on CD14+ monocytes compared to CD14+CD16+ monocytes, but TLR4 cell-surface expression was not significantly altered by intervention.

Conclusion: Exercise training reduced inflammatory monocytes without changes in body composition, but the changes were most prominent in overweight subjects. There was evidence of a resistance-to-change in monocyte phenotype for obese subjects in the EX group.

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P.17. Activin A, a potential negative regulator of muscle mass, decreases in response to exercise (ECRA)

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Background: Activin A is a multifunctional protein belonging to the TGF-β superfamily. In cell cultures, it has the ability to inhibit proliferation and differentiation of skeletal muscle during development [1,2]. Together with another TGF-β superfamily member, myostatin, it is suggested to play a key role as a negative regulator of skeletal muscle growth *in vivo* [3]. And as for myostatin, circulating activin A is regulated by follistatin [2].

Aim: 1) To investigate if activin A is regulated by an acute bout of exercise and 2) to determine, in which tissues activin A is regulated during exercise.

Methods: 1) Five healthy young men were recruited to do three hours of bicycling at 50% of $\dot{V}O_2\,\max$ followed by six hours of rest. Blood samples were drawn from the antebrachial vein during exercise and rest.
2) Twenty-four mice performed 1h of swimming and 8 mice served as controls. The expression of activin A was examined in muscle, fat, spleen, liver, heart and kidneys using qPCR.

Results: 1) Activin A increases slightly during exercise (insignificant). In contrast, during recovery after exercise, activin A decreases markedly at time point 3h (91.9 vs. 45.9 pgl/ml, $p< 0.01$) and 6h (91.9 vs. 44.7 pg/ml, $p < 0.01$) compared to baseline concentration. At 24h, the concentration is back to baseline.
2) In line with the decrease in circulating acticin A, the hepatic expression of the protein also decreases during recovery and reaches the lowest expression at time point 3h (16.9 vs. 8.0, $p > 0.01$) compared to baseline.

Conclusion: Circulating activin A decreases during recovery from an acute bout of exercise. The decrease in the circulating activin A corresponds to a decrease in the hepatic expression of the protein. Furthermore, it corresponds to the increase in circulating follistatin, a known regulator of activin A [4]. Thus, this might represent a functional mechanism in the regulation of muscle mass in response to exercise.

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P.18. Fitness, fatness and low grade inflammation in adolescents. The AFINOS study

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In order to achieve a better understanding of the development of chronic diseases in youth, it is necessary to know objectively measured levels of physical activity (PA). The objective of the AFINOS (*Physical Activity as a Preventative Agent of the Development of Overweight, Obesity, Allergies, Infections, and Cardiovascular Risk Factors in Adolescents*) Study has been to find out possible associations between PA, cardiorespiratory fitness (CRF) and fatness with low-grade inflammatory biomarkers. To this end, a cross-sectional sub-sample of 202 adolescents (99 girls), aged 13-17 years was selected. The results showed: (a) The relevance of PA, especially vigorous PA, to provide cardiorespiratory fitness (CRF) in adolescents and the key role of CRF on the metabolic syndrome (MetS) measured by triacylglycerol (TG), total cholesterol (TC), high-density lipoprotein cholesterol (HDL-C), insulin and glucose. Furthermore, a unified pediatric definition of MetS might minimize the discrepancies among studies [1]. (b) The key role of CRF and fatness on low-grade inflammation measured by C-reactive protein (CRP), interleukin-6 (IL-6) and complement factors C3 and C4, as well as the possible indirect role of habitual PA through CRF and body fat in adolescents [2]. (c) An association between sedentary behaviour and CRF score [systolic (SBP) and diastolic blood pressure (DBP), glucose, total cholesterol (TC), triacylglycerol (TG), HDL-C, low density lipoprotein cholesterol (LDL-C), and apolipoproteins A-1 and B-100] in adolescents, especially in obese adolescents. Abdominal adiposity seemed to play a more significant role in the development of CRF than overall adiposity [3]. (d) PA, CRF and muscular fitness (MF) are inversely and jointly associated with adiponectin and leptin concentrations in adolescents [4]. Conclusion: An active lifestyle and a desirable CRF may attenuate the development of chronic diseases in youth. This outcome may have important implications for public health. Therefore, specific strategies should be developed for adolescents due to the well-known decline in PA levels during this crucial life period. Likewise, other types of PA related to MF (that is, resistance training) might be taken into consideration during adolescence because high levels of muscular fitness have shown negative associations with inflammatory proteins.

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Posters: Exercise and the intestinal microbiota: Function and immunity

P.19. Health promoting and immune enhancing effects of *Bifidobacterium lactis BI-04:* a randomised controlled trial

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Healthy active individuals are consumers of probiotic supplements and probiotic-enriched foods to enhance immunity and reduce susceptibility to illness. There is a paucity of data on the efficacy of probiotics in this sub-group of the population. This study investigated whether probiotics reduce upper respiratory tract (URTI) and gastrointestinal (GI) illness symptoms in a double-blind placebo-controlled trial over 150 d. Physically-active healthy adults (117 males, 109 females, age 36 ± 10 y; mean ± SD) were randomly assigned to either probiotic ($2x10^9$ *Bifidobacterim lactis* Bl-04™) or placebo. Probiotic and placebo were ingested as a powder dissolved in a drink taken daily. All subjects kept an illness and training diary for self-reporting of the frequency, type, duration, severity, and load (duration x severity) of illness symptoms on a daily basis, as well as daily patterns of exercise and athletic training. A cohort of 87 individuals (47 males, 40 females, age 35 ± 10 y) provided a blood sample for analysis of innate immune parameters. A reduction of 10% in illness symptoms was established as the threshold value for a substantial difference between treatments - ratio 1.0 ± 0.2 (or ratio interval 0.83-1.2). Males and females taking *B. lactis* Bl-04 had a substantial ~26% (-17 to 55%; 99% confidence interval) lower upper respiratory tract load and symptom duration than the placebo group. The clinically beneficial effect of *B. lactis* Bl-04 on URTI symptom load and duration was evident in females (45% lower; -14 to 73%) but not males. The severity of chest illness symptoms for males and females was ~17% (-23 to 45%) lower in the *B.lactis* Bl-04 group than the placebo group. Males and females on *B.lactis* Bl-04 had 32% (-9% to 57%) fewer total days of medications compared with those on the placebo. *B. lactis* Bl-04 enhanced neutrophil phagocytic activity by 25% (-5 to 65%) and monocyte phagocytic activity by 27% (-6 to 73%). This study provides evidence of clinical benefit using probiotics to reduce respiratory illness in healthy active individuals. Maintenance of phagocytic activity may be one mechanism underpinning the beneficial clinical outcomes with *B. lactis* Bl-04™ supplementation.

P.20. Investigation of the clinical and immunological effects of a multi-strain probiotic: a randomised controlled trial

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Administration of a probiotic supplement combining two strains has shown efficacy in reducing clinical symptoms of cold symptoms in healthy (3-5 y age) children (1). Less is known regarding the efficacy of multi-strain probiotics in healthy active individuals. This study investigated whether probiotics reduce upper respiratory tract (URTI) and gastrointestinal (GI) illness symptoms in a double-blind placebo-controlled trial over 150 d. A secondary aim was to investigate the effects on innate immunity. Healthy physically-active adults (111 males aged 37 ± 11 y and 109 females aged 38 ± 11 y; mean ± SD) were randomly assigned to either probiotic (*Lactobacillus acidophilus* NCFM™ and *Bifidobacterium lactis* Bi-07™ at $1x10^{10}$ (5 billion each)), or placebo, taken daily as a powder dissolved in a drink over 150 d. A sub-group of 43 males (aged 37 ± 9 y) and 38 females (aged 36 ± 10 y) provided pre- and post-supplementation blood samples. Clinical benefit was established where the likelihood of 10% reduction in symptoms was >25% provided the likelihood of harm (a 10% increase in symptoms) was <5%. Precision of estimation for clinical benefit and harm was reported with a 99% confidence interval. Compared to the probiotic group, the placebo group had shorter duration of mild GI symptoms (-26%;-50 to 9%; 99% confidence intervals) and symptom load (-31%; -55 to 5%). In comparison to males in the probiotic group males in the placebo group had a 45% (-9 to 130%) higher number of total days of medication use. There was a relative 11% (-17 to 32%) greater increase in monocyte phagocytic activity in the combined probiotic group compared to placebo. No substantial effects of supplementation were evident in neutrophil phagocytic activity or NK cell activity. Decrements in clinical indices of URTI conflict with previous reports while increased mild GI symptoms may be associated with adaptive responses to the probiotics. Further work is required to clarify whether combined NCFM™ and Bi-07™ probiotic-induced enhancement of phagocytic activity elicits substantial clinical benefits in adult population.

P.21. Ingestion of *lactobacilus casei* contribute regeneration of skeletal muscle in older adult mouse

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Background: Regeneration of damaged skeletal muscle requires sufficient supply of nutrients. Fully functional intestine and colon assure sufficient supply of nutrients. Since gut commensal bacteria support intestinal function. It was reported that lactobacillus treatment was effective for athletes presenting fatigue and impaired performance. We hypothesized that *Lactobacillus casei* (*L.casei*) administration may facilitate recovery of skeletal muscle tissue from damage in older animals in which muscle regeneration is compromised.

Objective: To investigate contribution of *L.casei* to the recovery of damaged skeletal muscle in young and older adult mice.

Methods: *L.casei* was given orally at a dose of 10^{-8} /day for 7days to 10 weeks old (young) and 45-55 weeks old (older adult) male C57BL/6 mice. Vehicle control mice received an equivalent volume of water for 7 days. On the eighth day, cardiotoxin (CTX) was injected to gatrocnemius muscle to induce muscle damage. On days 3, 5, 7, 10, 14 and 20 after CTX injection, mice were sacrificed. Excised gastrocnemius muscle was subjected to weight measurement and immunohistochemical analyses.

Results: There was no difference in both the recovery of muscle weight and the regeneration process of gastrocnemius muscle examined immunohistochemically between control and *L.casei* treated young mice. The expression of dMHC, a marker of premature regeneration, was positive up to 5 days in both groups. The delay in the recovery of muscle weight was obvious in old mice regardless of the treatment. However, while the expression of developmental MHC (dMHC) was prolonged up to day 7 in the vehicle control, dMHC expression was notable up to day 3 or day 5 in the *L.Casei* treated. Therefore, in older mice *L.casei* treatment may have facilitated the maturation process of regenerating skeletal muscle.

Conclusion: Our result suggests that *L.casei* favor the recovery of skeletal muscle from muscle damage in older adult mouse.

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Posters: Exercise and neural inflammatory diseases

P.22. Freewheel training alters mouse hippocampal cytokine expression (ECRA)

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Introduction: Alzheimer's disease (AD) and dementias represent a significant public health burden. It is estimated that one in 85 people may be living with AD by 2050 [1]. Dementias are a spectrum of diseases with common traits including inflammation in the central nervous system (CNS) [2]. Epidemiological studies indicate possible risk reduction through modifying lifestyle factors such as exercise [3]. It has also been suggested that training reduces the negative impact of central inflammatory cytokines [4].

Purpose: We examined the effects of long-term voluntary exercise in healthy mice on cytokine expression (TNF-α, IL-1β, IL-12, IL-6, IL-10, and IL-1ra), differences in cell phenotypes (CD45+ and CD11b+), and cellular apoptotic status (Annexin+, Annexin/PI+, PI+) in hippocampal cells. Measures of training (running volume, body weight, run-to-exhaustion time, and skeletal muscle cytochrome c oxidase activity) were collected.

Methods: Female C57BL/6 mice were assigned to wheel running (WR, n= 20) or a control (No WR, n= 22) condition and sacrificed after 16 weeks. Hippocampi were removed and prepared as single cell suspensions for immunophenotyping and analysis of apoptosis (flow cytometry) and determination of cytokine protein expression (Western blotting).

Results: Long-term freewheel running induced significant training effects in WR compared to No WR mice. WR mice had lower TNF-α ($p<0.05$), and higher IL-6 ($p<0.05$), IL-1ra ($p<0.05$), and IL-12 ($p<0.05$) expression in the hippocampus compared to No WR animals. No group differences in hippocampal expression of IL-1β and IL-10, cell phenotypes, and % apoptotic and dead cells were observed.

Conclusions: The results indicate that long term voluntary freewheel running in healthy young mice alters central cytokine balance and may reduce the damaging effects of inflammation due to TNF-α. Moreover, IL-6, IL-1ra, and IL-12 were all elevated suggesting that exercise training does not result in a simple dichotomous pro- vs. anti-inflammatory cytokine response in the hippocampus. Further investigations will be required to elaborate on the specific pathways through which these complex cytokine interactions in the hippocampus may influence cognitive function and dementia-like conditions in older animals given long-term training.

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Posters: Exercise and health: Disease prevention and treatment

P.23. IL-18 released by monocytes is deregulated in fibromyalgia patients: Differential effects of exercise in healthy and fibromyalgia women (ECRA)

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Fibromyalgia (FM) is a form of non-articular rheumatism characterized by chronic (of more than 3 months duration) widespread pain and allodynia to pressure. The current hypothesis of the aetiology of FM includes inflammatory disorders involving high levels of circulating and monocyte-derived inflammatory cytokines. Exercise is recommended as a non-pharmacological intervention in these patients [1,2]. Interleukin-18 (IL-18), initially described as interferon-□ inducing factor, is ranked as a pro-inflammatory cytokine and contributes to systemic and local inflammation and host defense. Thus, IL-18 is seen as a major pro-inflammatory cytokine implicated in inflammatory diseases [3,4].

The aim of the present work was to investigate a hypothesized deregulation in the plasma concentration of IL-18 and IL-18 released by cultured monocytes in FM patients (determined by Luminex, Bio-Plex system), as well as to know the effect of a single session of moderate exercise (cycling for 45 min at 55% VO_2 max) in FM patients compared with healthy women. No significant changes were found between healthy controls and FM patients in the circulating concentration of IL-18. However, monocytes from FM patients released, both constitutively and in response to LPS, more IL-18 than those from age-matched healthy women. Exercise did not modify significantly the circulating concentration of IL-18 in either sedentary healthy women or sedentary FM patients. In contrast, exercise stimulated both the constitutive and LPS-induced release of IL-18 by monocytes from sedentary healthy women but decreased the release of IL-18 by monocytes from sedentary FM patients.

It is concluded that the release of IL-18 by monocytes is deregulated in the fibromyalgia syndrome. In addition, acute moderate exercise may be anti-inflammatory (at least for IL-18) only in fibromyalgia patients (with a high “inflammatory status”), but not in sedentary healthy women.

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P.25. Exercise effects on colon carcinogenesis in rats are subordinate of normal circadian rhythm (ECRA)

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It has been shown that appropriate levels of exercise can reduce colon cancer (CC) risk [1]. Constant light exposure conveys endocrine changes on melatonin rhythm secreted by pineal gland, a hormone that is involved in immunomodulation and has oncostatic properties [2]. Exercise is known to affect circadian rhythmicity in a variety of ways, however, the interaction of exercise and constant light regime on colon carcinogenesis is unknown.

Purpose: To verify the effects of physical exercise and circadian disruption on colon carcinogenesis.

Methods: 32 male Wistar rats (±200 g) were randomized into 4 groups: C (control), L (constant light), E (exercise) and LE (constant light + exercise). The circadian clock were synchronized with 12 hours of light and 12 hours of darkness (12:12h) to normal groups and 24 hours of light (24:0h) to constant light groups. Exercise was performed by progressive protocol of swimming training, 5 d-wk^{1} for 10 wk [3]. Other 32 rats were randomized into four groups (D, LD, ED and LED) treated with the four doses of carcinogen 1,2 dimethyl-hydrazine (DMH, 40mg/kg) in the first two weeks [4]. At the end of the protocol, the animals were euthanized and the distal colon was processed to immunohistochemistry analysis and histopathological study of Aberrant Crypt Focus (ACFs). Statistical analysis was performed using one-way ANOVA and two-way ANOVA.

Results: In relation to non DMH-treated groups, exercise performed under normal circadian or under constant light did not show any statistical difference in PCNA-labeling index, cyclooxygenase 2 (COX-2) and Caspase-3. However, in DMH-treated groups, exercise performed under constant light (LED group) in comparison to normal circadian exercise (ED group) showed a significant difference in the carcinogenesis parameters PCNA-labeling index (0.271±0.06 vs. 0.176±0.03, $p<0.001$), COX-2 (5.75±1.53 vs. 2.81±0.42, $p<0.001$), Caspase-3 (3.68±0.16 vs. 4.88±0.43, $p<0.01$) and ACFs (34.53±3.8 vs. 8.07±0.9, $p<0.001$), respectively.

Conclusion: These data indicated that normal circadian rhythm may be crucial to the effects of exercise against CC in rats treated with the chemical carcinogen.

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P.26. Practice of soccer four hours per week gives distinctive results in some fitness components: the sports popularity among faculty of medicine universitas indonesia's fitness challenge 2011 champions

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In celebrating the 4th birthday of Exercise Clinic, Sports Medicine Program held a series of fitness tests in competition setting titled "Fitness Challenge 2011". It was held on last 23rd February 2011. There were threes age group champions (college student, under 40, and over 40 years old) and four fitness component champions category (most ideal body composition, best flexibility, the fittest abdomen, and best aerobic fitness).

The fitness components being judged were body composition, back and hip flexibility, muscle strength, and muscle endurance. All components had been given score, and total scores were used to determine the champions. Before each test, all participants had been explained orally and visually using videos and signed an informed consent.

There were 13 champions, all had most common practice of sports: soccer, as well as futsal (indoor soccer), twice a week, for 2 hours per session. The data was taken by anamnesis before tests. All data had not yet been statistically analysed, however, some concerns were taken.

Soccer and futsal were very popular among the participants, just as they were for all Indonesians. Although this relate that more active people had better fitness components than those who didn't practice or those who practive sports rarely and irregularly, this type of sports were dominantly anaerobic. With nearly all range of age participating in this kind of sports, there was a warning that, some of the older age participants might have general condition that will be put in more danger by engaging in this sports. Recently there had been some sudden death cases in futsal in Indonesia, that gave more attention to health practitioner especially sports medicine. Sports doctors had to encourage people assessing their fitness in pre-participation examinations, like some kind of the tests being held in "Fitness Challenge 2011", before they were involved intensely in sports with anaerobic dominancy, thus needed more power than endurance.

Unfortunately the consciousness to have "active" medical check-up such as doing some fitness tests had yet to embrace in most of Indonesian people. This was a huge homework for medical practitioners, particularly those who evolved in sports medicine.

P.28. The effects of childhood obesity status on monocyte concentration and plasma chemokine concentration (ECRA)

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Background: Overweight/obesity is an independent risk factor for chronic diseases, such as type 2 Diabetes Mellitus and cardiovascular disease. In recent years, the prevalence of overweight in children has nearly tripled, especially among Mexican-American children. Childhood overweight greatly increases the risk for obesity and the associated chronic diseases in adulthood. Peripheral blood monocytes are altered with obesity and are purported to contribute to the systemic inflammation that appears to mediate the relationship between obesity and chronic disease. In addition, adult obesity alters the circulating levels of chemokines that influence monocyte behavior. Few published studies have examined monocyte-chemokine relationships in children.

Purpose: The purpose of this study was to investigate alterations in total blood monocyte and subset concentrations and plasma chemokine levels among normal weight (N=66) and overweight/at risk for overweight (N=56) Mexican-American children.

Methods: Blood samples were analyzed for total monocyte concentration, pro-inflammatory monocyte concentration, and classic monocyte concentration via flow cytometry. Plasma chemokines MCP-1, Fractalkine, IL-8, MIP-1α, and MIP-1β were measured using a Luminex MagPix assay.

Results: Total, pro-inflammatory, and classic monocyte concentration were significantly elevated in overweight/at risk for overweight children ($P<0.05$). In addition, circulating levels of MCP-1 ($P=0.009$) and Fractalkine ($P=0.027$) were significantly greater in overweight/at risk for overweight children.

Conclusion: Elevations in circulating monocytes, MCP-1, and Fractalkine have been implicated in the development of obesity-related chronic disease in adults. Childhood overweight alters monocytes and circulating chemokines in a similar manner as adult obesity, putting children at a greater risk of developing obesity-related chronic diseases in adulthood. More research is needed to determine if the observed changes can be altered following a period of exercise and/or weight loss.

P.29. The effect of an acute aerobic exercise intervention on antibody response to pneumococcal and influenza vaccination (ECRA)

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Introduction: Recent literature has shown that people who complete an intense bout of acute aerobic or resistance exercise prior to receiving a vaccination have enhanced antibody responses [1]. In resistance exercise, this effect has been shown to be independent of the intensity of the exercise; a less intense bout of exercise was just as effective as the more intense exercise [2]. The current study investigated the effects of a moderate intensity bout of aerobic exercise prior to vaccination. In addition, as most previous research has focussed on young, healthy adults, this study compared the effects of this intervention in younger (18-30 years) and older (50-64years) adults.

Methods: Sixty young (age (SD) =22.0 (6.1) yrs) and 60 older (age (SD) = 57.5 (6.5) yrs) adults attended the laboratory on two separate occasions. At the first session, a blood sample was drawn to determine baseline antibody titres, before participants completed either a brisk walk around campus at >55% of their age-predicted heart rate maximum (30 older, 30 younger), or a resting control condition, for 45 minutes. After the intervention, all participants received a full-dose pneumococcal vaccination and a half-dose influenza vaccination. They returned four weeks later for a follow up blood sample.

Results: Participants in the intervention group achieved an average heart rate of 118.7(14.2) beats per minute (bpm) during the walk, exceeding 55% of the predicted heart rate maximum (mean = 99.2(10.27) bpm). Multivariate ANOVA revealed pneumococcal antibody titres increased in response to the vaccination ($F_{(12,106)} = 27.66$, $p < .01$, $\eta^2 = .76$), but there were no significant time × group ($F_{(12,106)} = 0.53$, $p = .89$, $\eta^2 = .06$)) or time × age × group ($F_{(12,106)} = 0.84$, $p = .61$, $\eta^2 = .09$) interactions. Similarly, there was a significant increase in antibody titres against influenza strains A/California ($F_{(1, 116)} = 187.56$, $p < .01$, $\eta^2 = .62$), B/Brisbane ($F_{(1, 116)} = 163.73$, $p < .01$, $\eta^2 = .59$) and A/Victoria ($F_{(1, 116)} = 166.09$, $p < .01$, $\eta^2 = .59$), but again there were no significant effects of the intervention (p's all >.05). Younger participants had significantly higher antibody titres against five of the twelve pneumococcal strains and two of the three influenza strains, compared to the older adults.

Conclusion: A 45 minute brisk walk prior to influenza and pneumonia vaccination did not affect antibody response to vaccination. It is possible that higher intensity exercise is necessary to augment antibody response to vaccination.

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P.30. The effect of a lifestyle physical activity intervention on antibody response to vaccination (ECRA)

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Introduction: Recent literature suggests that long-term moderate physical activity is associated with improved immune function, such as antibody response to vaccination [1,2]. However, these studies used highly structured and supervised exercise interventions. This study investigates whether a lifestyle physical activity intervention can have similar benefits for immune function in sedentary middle-aged women.

Methods: Eighty-nine sedentary women (Mean age (SD) = 47.35 (6.9) years) were recruited and randomised into either an intervention (N = 44) or control (N = 45) group. Participants attended an initial familiarisation session, where they were given a sealed pedometer to wear for one week to assess baseline walking activity. At the second session, body composition (BMI, body fat percentage, waist-hip ratio, and blood pressure) and aerobic fitness (sub-maximal VO^2 treadmill test) assessments were performed and psychosocial questionnaires were completed. The intervention group then received a physical activity consultation (Kirk et al., 2007), use of an unsealed pedometer and weekly telephone/email prompting, whereas the educational control group received an advisory leaflet on increasing walking behaviours. Twelve weeks later, participants returned to the laboratory for a baseline blood sample before receiving a pneumococcal vaccination. Four weeks later, participants attended the laboratory for the final session to assess antibody status, complete further questionnaires and to re-measure body composition and aerobic fitness. During the final week of the intervention, objective walking behaviour was again measured using sealed pedometers.

Results: Seventy-four participants (N = 39 intervention group) completed the full study. There was a significant time × group interaction for step counts ($F_{(1, 67)} = 9.63$, $p < .01$, $\eta^2 = .13$) and self-reported walking behaviour ($F_{(1, 68)} = 11.25$, $p < .01$, $\eta^2 = .14$); the consultation group increased both objective and subjective measures of walking, whereas the educational control did not change. Multivariate ANOVA revealed a significant increase over time for pneumococcal antibody response ($F_{(11, 61)} = 20.43$, $p < .01$, $\eta^2 = .79$). However, there was no significant time × group interaction ($F_{(11, 61)} = 0.90$, $p = 0.55$, $\eta^2 = .14$) indicating that the intervention did not change antibody responses. Similarly, univariate analyses of individual strains found no significant interactions.

Conclusion: The lifestyle physical activity intervention did not affect vaccination response in women. It is possible that the intensity of exercise was not sufficient in changing antibody response, or that the study population were too young to gain the benefits from physical activity.

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P.31. Chronic systemic low-grade inflammation, cardiovascular risk factors, physical activity, fitness and adiposity in children and adolescents aged 10 – 19 years

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Introduction: Regular physical activity exerts well documented beneficial effects on traditional markers of the metabolic syndrome, on markers of chronic low-grade inflammation, on so-called adipokines as well as on cardiovascular risk factors in both, children and adults [1,2]. The aim of this cross-sectional study was to investigate whether additional physical activity through sports activities on a competition level is able to influence the above mentioned parameters in school-aged children.

Methods: During the school year 2008/09 fasting blood samples were withdrawn from 201 pupils (aged 10-19y) attending one particular high school in Vienna with two distinct school-types: Half of the participants (40 girls, 61 boys) visited the regular school whereas the other half (40 girls, 60 boys) attended the elite sports school with individual extra-curricular sports activities. We measured metabolic parameters (blood glucose, insulin, cholesterol, and triglycerides), inflammatory parameters (hs-CRP, IL-6, TNF-α), adipokines (leptin, leptin sR, adiponectin, ghrelin) and cardiovascular risk factors (homocysteine, NT-proBNP). Additionally, anthropometric data, physical activity and physical fitness were assessed. Statistical analyses were performed using SPSS 17.0.

Results: As expected, pupils from the elite sports classes exercised on more days per week than those from the regular school and showed a higher endurance capacity and strength of upper and lower extremities than their counterparts. Adiponectin, leptin and leptin sR were associated with BMI, but only leptin levels differed due to the amount of sports activities. Inflammatory parameters were significantly associated with age and BMI but were not different between school types.

Discussion: It has been shown that illnesses such as diabetes, cardiovascular diseases, cancer and others are associated with physical inactivity – a risk factor for the accumulation of visceral fat and consequently of chronic systemic inflammation [3]. The results of this study contribute to the growing evidence that extra-curricular physical activity is able to modify these risk factors in a positive way as early as in childhood.

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P.32. Endurance training and erythropoietin reduce the inflammatory status in cachectic mice without preventing muscle wasting

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Cancer cachexia is a syndrome characterized by loss of skeletal muscle protein, depletion of lipid stores, inflammation, anorexia, weakness, and perturbations of the hormonal homeostasis [1]. Despite several therapeutic approaches were described in the past, effective interventions countering cancer cachexia are still lacking [2]. In addition to nutritional interventions, exercise training was proposed as a suitable tool, in view of recent observations suggesting that decreased physical activity plays a role in the onset of muscle atrophy in cancer patients [3].

Aim of the present work was to verify if endurance training coupled to erythropoietin (EPO) administration could prevent the wasting process in Colon26 carcinoma(C26) -bearing mice.

C26 mice were got used to a treadmill for 5 days before tumor injection and then exercised 5 days/week (14m/min for 45 min) during the two weeks of tumor growth. At the end of the protocol, the results showed that exercise alone not only did not prevent, but even worsened muscle wasting. The combination of exercise with EPO (100U/mouse, i.p., weekly) prevented tumor-induced haematocrit reduction and partially attenuated adipose tissue depletion, while the loss of muscle mass was similar to the sedentary tumor-bearing mice. Moreover, the combined approach partially counteracted spleen hypertrophy and decreased interleukin(IL)-6 serum levels. In the attempt to reduce the negative effects exerted by the 2 weeks exercise protocol, a prolonged training (8 weeks, starting 6 weeks before tumor injection) was tested. In this case, C26 mice well tolerated exercise, muscle mass being unaffected and the drop of both muscle strength and food intake being prevented. The concomitant administration of EPO confirmed the positive effects on haematocrit and adipose tissue, while the reduction of both spleen hypertrophy and IL-6 circulating levels was less evident.

Overall, the present data suggest that endurance exercise can be an effective tool to be added to combined therapeutic approaches against cancer cachexia. Further studies are needed to unravel the molecular mechanisms underlying the reported effects.

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Posters: Immunodepression and intense exercise: Evidence and strategies for prevention

P.33. Influence of sports massage on immunological parameters after high-intensity interval training (ECRA)

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Introduction: Sports massage is frequently used by athletes to improve recovery from strenuous exercise. Data from randomized controlled trials provide moderate evidence for the efficacy of massage therapy to facilitate recovery from repetitive muscular contractions [1]. The aim of the study was to investigate whether sports massage is able to affect the immunological response after high-intensity interval training (HIIT).

Methods: Twelve male, active subjects (age: 25±2 years, BMI: 22.6±2.7kg/m²) were included in this randomized, single-blind, controlled crossover intervention study with a 4-wk wash-out period. The exercise protocol consisted of HIIT (7min warm-up; 4x4min at 90-95% and 50% iHRmax, respectively; 7min cool-down). After exercise the participants received a standardised massage on legs and back or rested for 35min (control). Blood samples were taken before exercise (t0), directly after exercise (t1), after intervention (t2), 1h (t3) and 22h (t4) after intervention. Leukocyte counts as well as plasma IL-1ra, IL-6, sIL-6R, IL10, TNF-α, sTNF-αRI, CK and myoglobin concentrations were measured. Statistical differences were determined using repeated measured ANOVA.

Results: Leukocyte counts (+120% for granulocytes, $p<0.001$), muscle damage markers (+120% for myoglobin, $p<0.001$), and cytokines (+200% for IL-10, $p<0.001$) increased significantly after exercise. Almost all parameters showed their maximum increase directly after exercise (t1) with the exemption of IL-1ra and CK concentrations and granulocyte counts which peaked at later time points (t3 and t4). Sports massage did not affect any of the measured parameters.

Discussion: In our study we could confirm previous results showing a challenge of the immune system by strenuous exercise [2,3]. However, sports massage as recovery measure did not affect these parameters. This is slightly in contrast to a study [4] which demonstrated higher secretion rates of salivary IgA after massage recovery intervention. In conclusion sports massage might improve subjective parameters and delayed onset muscle soreness but not the recovery of immunological parameters after HIIT.

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P.34. Immunologic profile of athletes presenting upper respiratory airways symptoms after a marathon

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Introduction: The concept that the high prevalence of respiratory tract infections in athletes after a marathon is caused by a systemic process of immunodepression, has been challenged recently by authors that shown that this clinical condition is associated not only to infection but also to allergic and inflammatory processes. We studied the effect of a marathon in the mucosal immunologic profile from upper respiratory airways, comparing athletes that present or not symptoms after a marathon race.

Methods: The group of marathon runners was composed by 18 male runners from São Paulo, with a range of age of 25 to 60 years (mean 41.4). The athletes were separated by clinical examination and questionnaire analysis in two groups, one that shows upper respiratory tract symptoms (SY, n=8) and another without symptoms (WSY n=10). We evaluated the levels ($p<0.05$) of secretory immunoglobulin A (sIgA, μg/mL) in saliva and cytokines (pg/mL), interleukins (IL) 6, 8 and the tumor necrotic factor (TNF) alpha in protein extract obtained from total lysate of nasal cells, in three moments: at rest, immediately and 72 hours after race.

Results: We observed that levels of sIgA [133.7(102.1-164.1)] and cytokines [IL-6= 1.38(0.54-12.71), IL-8= 1985(469.5-4958) and TNF-α= 2.18(1.15-14.37)] in SY group at rest did not differ in relation to WSY group [sIgA = 104.2(43.07-242.4), IL-6= 1.31(0.49-2.82), IL-8= 2659(583.1-7786) and TNF-α= 1.72(0.41-5.77)]. The results observed immediately after the marathon showed that levels of sIgA in SY [76.56(62.03-103)] group, but not in WSY [92.16(55.27-173.4)] group, were significantly reduced compared to basal levels. The analysis of cytokines immediately after the marathon didn't showed statistical difference between groups [SY - IL-6= 6.44(5.33-14.28), IL-8= 1342(901.7-4759) and TNF-α= 1.95(0.79-3.66); WSY - IL-6= 9.55(4.35-13.41), IL-8= 980.9(727.4-1049) and TNF-α= 0.71(0.41-2.13)]. After 72 hours levels of sIgA in both groups [SY= 107.6(84.71-178.1) and WSY= 96.74(85.06-172.5)] return to basal levels, and analysis of concentration of cytokines showed levels of IL-8 [7367(6213-7786)] and TNF-α [6.95(4.2-21.27)] in SY group significantly higher than the values observed in WSY group [IL-8= 4015(1912-6201) and TNF-α= 2.03(1.51-14.37)]. IL-6 levels [SY= 10.42(1.86-15.86); WSY= 10.93(3.24-20.95)] didn't showed difference between groups.

Conclusion: The demonstration of decreased sIgA levels in saliva after the marathon in the symptomatic group, according to the literature, can be associated with infection. Otherwise, the maintenance of increased levels of pro-inflammatory cytokines (TNF-α and IL-8) in nasal cells extract after 72 hours could explain the allergic/ inflammatory symptoms in these athletes.

P.35. Assessment of oxidative stress in lymphocytes with exercise (ECRA)

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This study investigated whether changes in the cellular composition of blood during exercise, could partly account for observations of exercise-induced changes in lymphocyte oxidative stress markers.

Markers of oxidative stress and antioxidant capacity were assessed before and after 60 minutes of treadmill running at 80% $\dot{V}O_2$ max. Samples were collected from sixteen men (mean ± SD; age 33 ± 13 y; body mass index 23.8 ± 2.5 $kg \cdot m^{-2}$, maximal-oxygen uptake 59.7 ± 5.2 $ml \cdot kg^{-1} \cdot min^{-1}$). Peripheral blood lymphocytes were assayed for protein carbonylation and plasma was assessed for lipid peroxidation and antioxidant capacity. In a separate study, intracellular thiol concentration was determined in lymphocyte sub-sets from 8 characteristically similar men by flow cytometry, of which T cell memory populations were further identified on the basis of CD27, CD28, and CD45RA expression.

The results showed that total lymphocyte protein carbonylation was transiently increased with exercise and returned to baseline within 15 minutes ($p < .001$). This change was accompanied by increased plasma lipid peroxidation ($p < .05$) and total antioxidant capacity ($p < .001$). Correlation analyses showed that lymphocyte protein carbonylation was not related to changes in the cellular composition of peripheral blood during exercise. Natural killer cells ($CD3^-CD56^+$) and late-differentiated/effector memory cells ($CD4^+/CD8^+CD27^-CD28^-/CD45RA^+$) which mobilised most with exercise, showed high intracellular thiol content ($p < .001$).

High thiol content suggests a lower 'oxidative load' carried by these lymphocytes. Thus, vigorous exercise resulted in a transient increase in lymphocyte oxidative stress. Results suggest this was un-related to the alterations in the cellular composition of peripheral blood.

P.36. Increase in T regulatory lymphocytes after a winter training season in swimmers

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Regulatory T cells (Tregs) have been identified as $CD4^+$ expressing high levels of the IL-2 receptor (CD25). Another characteristic is the expression of the FoxP3 protein, a key transcription factor that governs maturation of these cells. They play an important role in controlling or suppressing immune responses mediated by other T lymphocytes. It is believed that the immunosupressor mechanism used is mediated by inhibitory cytokines like IL-10 and TGF-b. The number of Tregs has been shown to increase with age and a recent study has also found increases after exercise in a group of adolescent elite swimmers [1]. The aim of our study was to look at the effect of training load during a winter training season on the Treg subpopulation of a group of elite swimmers and how it could affect susceptibility to disease. A group of high-level swimmers (13 males and 6 females), and 11 non-athletes, were recruited to this study. Upper respiratory symptoms (URS) episodes were monitored using daily logs. Blood samples were taken at rest at 4 time points during the swimming season: before the start of the season (t1 – middle September), after 7 weeks of an initial period of gradually increasing training load (t2 – early November), after 6 weeks of an intense training cycle (t3- late February) and 48 hours after a main competition (t4 - early April). Blood samples were taken from the non-athlete group at 3 similar time points (t1 – early November; t2- late February; t3- early April). In the swimmers a trend for the occurrence of URS around the periods of elevated training load was observed while no URS were observed for the non-athlete group. The athletes showed an increase in the % of the Tregs subpopulation at the end of the training season while there was no variation in the non-athlete group. The levels of Treg cells were lower in the non-athletes when compared to the athletes. The number of Tregs peaked at t3 for the athletes coinciding with an increase in URS episodes.

Periods of highly demanding training seem to increase the number of Tregs probably leading to an increased suppression of immune responses, which could partially explain, the higher frequency of URS observed in the athletes.

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P.38. The effects of a simulated altitude device on the mucosal immune system of triathletes (ECRA)

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"Live High-Train Low" (LHTL) altitude acclimatization has been shown to increase athletic endurance performance; however, this type of training has been associated with decreases in mucosal immune system function as measured by salivary IgA antibodies [1]. The physiological effects of altitude, strenuous training, and isolation from family could be additive stresses that depress the immune system [2]. Babcock [3] has shown that a simulated intermittent altitude exposure (SIA) device utilized for short durations has the same performance enhancing effects as LHTL; however, the effect of SIA via re-breathing on running performance and maintenance of immune system functionality has not been established.

Purpose: To determine the effects of SIA on running performance and the mucosal immune system.

Methods: Twenty well-trained male triathletes were exposed to the hypoxic stimulus for fifteen days. SIA was administered using a simple device (Alto2Lab, Pharma Pacific, Inc.) consisting of a breathing tube attached to an open-ended silo containing carbon-dioxide absorptive sodalime. Oxygen saturation was monitored with a pulse oximeter, and progressively reduced (90% on Day 1 to 77% on Day 6-15; equivalent altitudes equal 3600-6200 m) for treatment group 1 (LON) or held constant for treatment group 2 (SHO, 90%; Day 1-15); the control group (CON) was not exposed to the SIA. Time to complete a 10K race was tested pre- and post-treatment. Salivary IgA was tested pre- and post-treatment using ELISA. Comparisons between groups were made using the non-parametric unpaired Mann-Whitney test.

Results: Time to complete the 10K race significantly decreased for the SHO and LON treatment groups compared to CON ($p<0.05$). There was no significant difference in run performance between the SHO and LON groups. No significant difference in salivary IgA antibodies was detected between the groups ($p<0.05$).

Conclusion: A simple, cost-effective, device can elicit the same performance enhancing effects as LHTL in a much shorter time, without compromising the mucosal immune system. Such a method to simulate altitude exposure while remaining at sea-level is considered highly desirable among competitive endurance athletes.
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P.39. Acute effects of green tea consumption following intensive taekwondo training on salivary defense factors and antibacterial capacity

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Many factors presented in mucosal secretions serve as a first line of defense against microbial infection, including immunoglobulins (Igs), α-amylase and anti-microbial peptides (AMPs) [1]. Salivary immunoglobulin A (sIgA) contributes to mucosal immunity by preventing adherence of microbes to the mucosal surface [2]. Amylase was shown to function as an antibacterial protein inhibiting bacterial growth and colonization in the oral cavity. Lactoferrin, one of the most abundant salivary AMPs, exerts an antibacterial effect by sequestering iron, an essential nutrient for bacterial growth, as well as directly interacting and damaging bacterial membrane [3]. This study was aimed to investigate the acute effects of green tea consumption on selected salivary defense proteins, antibacterial capacity and anti-oxidation activity in taekwondo (TKD) athletes following intensive training.

Twenty-two taekwondo athletes performed a 2-hr TKD training. After exercise, participants ingested green tea or equal volume of water. Saliva samples were collected before training, immediately after training but before drinking, and 30 min after drinking green tea or water. Levels of salivary total protein, IgA, lactoferrin, free radical scavenger activity (FRSA), α-amylase activity and salivary antibacterial capacity were measured.

Results show that immediately after intensive TKD training, concentrations of lactoferrin, sIgA and α-amylase activity were significantly increased; while salivary antibacterial capacity was not affected by intense training. Levels of sIgA and lactoferrin returned to pre-exercise values after 30 min of rest. However, consumption of green tea after training further stimulated α-amylase activity and enhanced salivary antibacterial capacity. Additionally, we observed that salivary FRSA was markedly suppressed immediately after training and quickly returned to pre-exercise values regardless of which fluid was consumed.

Our results demonstrated that consumption of green tea exerts acute effect on the concentration of salivary oral defense-related proteins and significantly enhances salivary antibacterial capacity. However, detailed mechanisms underlying effects of green tea ingestion are still unclear and require further investigation.

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P.41. Inflammation-modulating effects of proanthocyanidolic oligomer supplementation in a model of contusion injury

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The inflammatory response to experimental contusion injury after 14 days oral supplementation with grapeseed-derived proanthocyanidolic oligomers (PCO, 20 mg/kg/day) was previously investigated in an *in vivo* placebo-controlled rodent model. PCO supplemented rats had relatively earlier satellite cell response (Pax7+ cells, ANOVA treatment effect $P<0.001$) and muscle fiber regeneration (MHC_f expression, ANOVA treatment effect $P<0.001$). Muscle and circulating cytokine (TNF-α, IL-6 and IL-10) levels, as well as magnitude and time course for neutrophil and macrophage infiltration into injured muscle and surrounding tissue, suggested an earlier switch from pro- to anti-inflammatory processes (Myburgh *et al*., paper submitted). These effects of PCO on the response to contusion injury were further investigated in the current study.

Wistar rats were supplemented with placebo (PLA) or PCO for 2 weeks prior to experimental contusion injury to the gastrocnemius muscle, and blood collected at different post-injury time points. Circulating neutrophil numbers were increased significantly from baseline to 12hr post-injury in both groups ($P<0.05$), but were significantly lower in PLA than PCO at day1 ($P<0.05$), suggesting increased migration into tissue. An influence on motility was confirmed using an *ex vivo* migration assay. Primary cultures of neutrophils from control animals, cultured in the presence of the neutrophil activator, G-CSF, were exposed to PLA or PCO conditioned plasma (obtained at different time points post-injury). Using co-culture, these neutrophils were then allowed to migrate from the insert into the bottom well, which contained RPMI media enriched with the chemoattractant, N-formylmethionyl-leucylphenylalanine. Neutrophils exposed to plasma obtained from PCO supplemented rats 1 day post-injury, exhibited significantly reduced migration capacity compared to PLA ($P<0.05$). Also, circulating monocytes/macrophages were subtyped using flow cytometry. M1 macrophage numbers peaked on day 3 post-injury in PCO rats, compared to day 5 in PLA. While M2c macrophage count showed no significant peak in PLA, a significant peak was seen in PCO on day 5 post-injury ($P<0.001$ versus day 5 PLA; $P<0.01$ versus all other time points for PCO).

We conclude that chronic oral supplementation with PCO limited the extent of neutrophil migration into injured tissue, potentially limiting secondary damage and facilitating an earlier pro- to anti-inflammatory macrophage phenotype switch.

P.42. Acute bovine colostrum supplementation enhances neutrophil oxidative burst at rest and following immunodepressive exercise: a pilot study

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Daily supplementation with bovine colostrum (COL), for between 2 and 12 weeks, has been shown to enhance neutrophil function but the underlying mechanism remains unclear [1]. Evidence from *in vitro* studies has suggested that low molecular weight (≤ 10 kDa) components exert a direct effect [2,3]. If this is the case, then it is possible that benefits would be apparent after acute supplementation. The aims of this preliminary study were to determine whether acute COL supplementation has an acute effect on the functional capacity of blood neutrophils at rest and following '*immunodepressive*' exercise.

Three males aged 21-25 took part in 2 resting trials and 1 subject (age 25) also participated in a case study with prolonged exercise. In the resting study, venous blood samples were obtained following an overnight fast for the determination of neutrophil PMA-stimulated oxidative burst (OBA) before (Pre) and 1 h after (1h-Post) the consumption of 30 g of COL powder or an isocaloric and macronutrient placebo (PLA). Trials order was randomised and separated by 1 week. For the case study, the subject undertook exercise a further week later (2.5 h cycle at 60% maximal oxygen uptake). Blood samples were obtained before and after the exercise. PMA-stimulated OBA was measured in pre- and post-exercise samples that were incubated with the subjects own plasma from the resting study (i.e. post-ingestion of COL or PLA).

Results. Neutrophil OBA

	Pre	1h Post
Resting Study		Mean (SD)
PLA	100 (-)	99.7 (30.1)
Colostrum	100 (-)	132.6 (30.4)
Exercise Study	Pre-exercise	Post-exercise
PLA	100	52.5
Colostrum	100	57.0

OBA results are expressed on a per cell basis as % of Pre

The ~50% post-exercise decrease in OBA in the PLA condition is consistent with previous research. These findings provide supporting evidence that bovine colostrum has acute and direct effects on neutrophil function. The idea that bioactive components within bovine colostrum (or biologically available metabolites), which can appear in the systemic circulation after oral ingestion, contribute to the previously observed enhancement, both *in vivo* and *in vitro* [1-3], is also supported. Further work is required to identify the specific components responsible for this enhancement.

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P.43. Effect of moderate exercise training on LPS-induced TNF-α production in rats (ECRA)

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Objectives: Moderate exercise training has many beneficial effects in physical and immunological functions. However, little is known that the influence of exercise training on pro-inflammatory cytokine in response to pathogen, although acute strenuous exercise attenuates lipopolysaccharide (LPS)-induced cytokine production. We examined whether exercise training affects systemic tumor necrosis factor (TNF)-α production in response to LPS after strenuous exercise.

Methods: Female F344 rats were divided into two groups, exercise trained and sedentary. The exercise trained rats completed 4 weeks of treadmill running 5 days/week for 15-30 min at 15-21 m/min on 15% grade, while the sedentary rats remained in their cages. The rats in both groups were treated an exhaustive exercise or a rest for 2 h. The exhaustive exercise was a treadmill running exercise until exhaustion. To measure plasma catecholamine and TNF-α concentrations, and TNF-α mRNA expression in tissue in response to LPS, the rats received an injection of LPS and were killed 1 h after the LPS injection.

Results: Running time until exhaustion in exercise trained rats was significantly longer than that in sedentary rats. In sedentary group, plasma catecholamine after exhaustive exercise was significantly higher than that after rested condition. In trained group, plasma dopamine was significantly increased by the exhaustive exercise. In both trained and sedentary groups, plasma TNF-α concentration was greatly inhibited by the exhaustive exercise. However, there was no difference between LPS-induced TNF-α in each sedentary and trained groups. In sedentary group, the exhaustive exercise slightly inhibited the expression of TNF-α mRNA in liver, but not in trained group. Also, there was no difference between each groups in the expression in lung.

Conclusion: These results suggest that exercise training might not affect strenuous exercise-induced TNF-α suppression in response to LPS.

P.44. Effect of prolonged exercise on oxidative burst function of peripheral blood and oral neutrophils: A pilot study

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The response of blood neutrophils to prolonged and/or strenuous exercise has been extensively studied but there is very little research on subpopulations other than peripheral blood neutrophils. In one previous study [1] neutrophils were obtained from nasal lavage following a 20 km run, in which a significant post-exercise increase in neutrophil number and a significant post-exercise decrease in phagocytic activity was observed. Neutrophils obtained in this way may represent a more appropriate subpopulation to study in relation to upper respiratory illness in athletes but there is limited research in this area. Such studies are widespread in dentistry research, in which strong associations have been observed between oral neutrophil functions and oral infection [2]. The aim of this pilot study was to determine the effects of prolonged exercise on the number and function of neutrophils obtained from the oral cavity.

Three healthy men participated in a prolonged exercise bout (2.5 h at ~55% maximal oxygen uptake). Oral neutrophils were obtained with methods modified from Lukac et al. (2). Briefly, 20 ml saline was swilled in the mouth for 2 min before expectoration. This was then concentrated 15 times by gentle centrifugation (450 × g) and replacing the supernatant with a smaller volume of buffer (HBSS). Blood samples were obtained pre- and post-exercise and both samples (blood and concentrated oral rinse) were used for determination of PMA-stimulated oxidative burst (OBA) by chemiluminescence assay.

Blood neutrophil count was increased ~2.5-fold post-exercise but there was no change in the oral samples. Overall, there was a mean post-exercise decrease in OBA per neutrophil in both sample types (Oral: 15 ± 28% decrease; Blood: 40 ± 14% decrease). However, an actual post-exercise decrease in oral neutrophil OBA was only evident in 2 of the 3 participants compared to the clear decrease of blood neutrophil OBA evident for all 3 participants.

The main findings of this pilot study suggest that prolonged exercise may induce immunodepression of oral neutrophils but there is poor agreement between the oral and blood neutrophil responses, although further research is required.

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P.45. The impact of an ironman triathlon race on Epstein - Barr virus and Varicella - Zoster virus antibody titres and the frequency of highly differentiated and senescent blood T-cells (ECRA)

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Epstein - Barr Virus (EBV) and Varicella - Zoster Virus (VZV) are persistent herpesviruses that are maintained in a latent state after primary infection, but may reactivate in response to acute or chronic stress. The reactivation of latent viruses is indicative of a compromised immune system, which may also increase the frequency of highly differentiated and senescent T-cells. Such changes in response to a prolonged endurance race could have important implications for athlete infection risk.

Purpose: To examine the impact of an ironman triathlon race on EBV and VZV antibody titres and the frequency of low and high differentiated and senescent blood T-cells in trained endurance athletes.

Methods: Eight trained male triathletes (age 44 $\pm$ 5 yrs) provided a fasted resting blood sample in the morning 3-wks before an ironman triathlon race and 1, 2, 4 and 6-wks after the race. EBV and VZV IgG antibody titres were measured in plasma by ELISA. Isolated blood lymphocytes were labelled with monoclonal antibodies to assess cell surface co-expression of CD57 and CD28 to identify the proportions of low differentiated (CD28+/CD57-), high differentiated (CD28+/CD57+) and senescent (CD28-/CD57+) cells within both CD4+ and CD8+ T-cell subsets using four-colour flow cytometry.

Results: 75% of triathletes were seropositive for EBV and 75% were seropositive for VZV. A main effect for the EBV-specific IgG titres over time ($P<0.05$) revealed higher titres 3-wks pre-race than 2, 4 and 6-wks post race and higher titres 1-wk post race than 6-wks post race. VZV-specific IgG titres did not change ($P>0.05$). Similarly, the proportion of low or high differentiated or senescent T-cells within CD8+ or CD4+ T-cells did not change.

Conclusion: The triathlon race did not appear to elicit EBV or VZV reactivation or alter the frequency of low or highly differentiated or senescent T-cells in blood. The decrease in EBV-specific IgG titres indicates enhanced viral clearance during recovery from the race, which may have been facilitated by a substantially reduced training load.

P.47. Effect of OLL 1073R-1 yogurt intake on NK cell cytolytic activities during intense training

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Purpose: Natural killer (NK) cell cytolytic activities are influenced by acute and chronic exercise. We previously reported that NK cytotoxicity decreased after intensive training [1]. However, oral intake of some lactic acid bacteria can have beneficial effects on the immune responses [2]. *Lactobacillus delbrueckii* ssp. *bulgaricus* OLL 1073R-1 (R-1) produces extracellular polysaccharides (EPS), and these in turn induce IFN-g production. In this study, we investigated the effect of R-1 yogurt intake on the cytotoxicities and expression levels of activating/inhibitory receptors on NK cells during intense training.

Methods: Trained male cyclists were randomized to R-1 (N=6) or Control (N=5) groups. Subjects practiced cycling for 6 hours/day. Subjects in the R-1 group received skimmed milk fermented with R-1 (224ml/day), while the Control group received the same amount of milk without R-1. Three morning resting blood samples were taken; before training (PRE), at the end of training (END) and 5 days after training (POST). Lymphocyte phenotypes and receptor density of NK cells were analyzed by flow cytometry. Cytotoxicity was measured by a standard ^{51}Cr release assay. The lytic unit was calculated as an index of per cell cytotoxicity.

Results: Proportions of $CD4^+$ T, $CD8^+$ T and $CD56^{dim}$ and $CD56^{bright}$ NK cells did not change throughout of the experiment. Total NK cell cytotoxic activity decreased at END in Control group. However, per cell cytotoxicity did not change in either group. In regard to the expressions of receptors on $CD56^{dim}$ NK cells, IL-12R expression increased significantly ($p=0.037$) at END in the R-1 group. DNAM-1 expression tended to decrease in both groups, although a significant decrease was seen only at POST ($p=0.005$) in the R-1 group. Expressions of NKG2D, NKp46 and NKG2A did not change in both groups.

Conclusion: These results suggested the possibility that R-1 intake had beneficial effects on NK cell responses during intensive training.

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P.48. Liver interleukin-6 , interleukin-10, glucose, and glycogen responses to endurance training with and without an alcoholic magnolia crude extraction in male rats (ECRA)

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Objective: The purpose of this study was to investigate the responses of interleukin(IL)-6, IL-10, glucose, and glycogen in male rat liver to exercise training with and without supplementation with a Magnolia crude extract.

Methods: Twenty-one adult male Wistar rats (6–8 weeks old, 142 ± 13 g) were randomly assigned to saline-control, saline-training, and magnolia-training. The training groups ran for 8 weeks (60 min/d, 5d/wk at 25 m/min and 0% grade). Animals were orally given either Magnolia crude extraction or an equal volume of saline for 4 weeks. Seventy-two hours after the last exercise session and 4 hours before the sacrifice food was removed but not tap water. A portion of liver was collected and frozen in liquid nitrogen for later analysis of liver interleukin(IL)-6, IL-10, glucose and glycogen concentrations. An one-way-ANOVA was employed.

Results: Signficantly lower IL-10 levels were found in trained groups when compared with control rats. Liver IL-6 and glucose but not glycogen concentrations were significantly lower in trained-saline treated group when compared with other groups. .

Conclusion: Results demonstrated that 8 weeks of treadmill exercise with magnolia improved liver glycogen and prevented an exercise-induced elevation in IL-6 concentrations. Data also indicate that training but not magnolia lowered IL-10 concentrations in trained rats. It seems that a liver glycogen improvement following training plus magnolia treatment has impact on IL-6 but not IL-10. This is in turn might confirm the role of pre-exercise liver glycogen level on the magnitude of IL-6 and IL-10 responses.

P.49. Profile of white blood cells differential counts of Satria Muda Britama Players in ASEAN basketball league and Indonesian national basketball league year 2011

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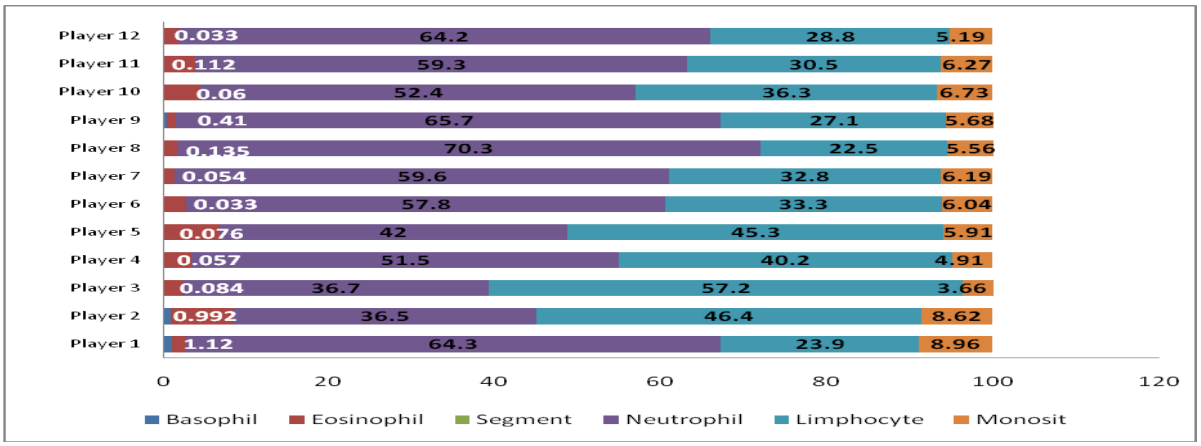
Background: In year 2011, Satria Muda Britama, the Indonesian runner up of ASEAN Basketball League 2009 also (Indonesian) National Basketball League 2011 champion joined two simultaneous competitions. They are ASEAN Basketball League 2011 and (Indonesian) National Basketball League 2010/2011.The team physician did notice some habitual common colds and flus experienced by the players, during these two seasons and suspected that some players might have problems in their immunity. However, there has not been any specific immune markers in relation to immune system function being periodically examined such as immunoglobulin level, cytokines level, or other markers of the players..

Methods: Before examining the players immune status more thoroughly by laboratory investigation, we tried to pay attention the leucocyte profile, represented by differential count. The blood samples taken on 1st February were part of twice a year routine health examination held from the team physician. results were taken 24 hours after the last practice, two sessions, morning strength training at the gym (1 hour) and 3 hours strategy/tactical training in the field.

Results: From all 12 players, there were 5 players with lymphocyte abnormal values. Two were lymphopenia and the rest were lymphocytosis. Two players got common cold and needed symptomatic medications. One player had prolonged symptoms and took antibiotics to relieve the symptoms. And one player who had lymphocytosis had knee injury later.

Analysis: During this season of competitions the training volume were huge, with every day practice, two sessions per day and only one day off. This suggested that the training had put stress that it affected not only the leucocyte profile but also general condition of the players, suggesting that there might be effect on the immunity as well.

Discussion: There had been indication that the training volume couldn't be adapted to some players. Further investigation for athlete immune status especially in basketball players with many simultaneous season of competitions is needed. And this investigation might have to consider many factors, such as recovery time after competition and is supplementation or certain vaccination needed, to keep the players in optimum immune status.



P.50. The effects of yeast β-glucan supplementation on monocyte and cytokine response to exercise (ECRA)

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Introduction: Strenuous exercise in hot, humid environments is known to suppress the immune system, which can increase the chances of getting sick in the hours after exercise. Supplementation with yeast β-glucan has been shown to reduce perceived sickness symptoms. The purpose of this study was to determine if 10-d of supplementation with yeast β-glucan alters monocyte concentration, LPS-stimulated cytokine production, and plasma cytokine concentration in recreationally active subjects.

Methods: 27 subjects (15 men, 12 women, 22±4 y) completed 49±6 min of cycling (37±2°C, 45±5% relative humidity) after consuming either yeast β-glucan (250 mg/d) or a placebo (sugar pill) for 10-days prior to each exercise session. The investigators were blinded to the supplement conditions (706 vs. 223). Blood was collected at baseline (prior to supplement), pre-, post-, and 2H-post exercise. Each subject completed both trial conditions in a random order, separated by a 7-day washout period. Total and subset monocyte concentration was measured by flow cytometry (Guava EasyCyte 6HT-2L). LPS-stimulated production of 12 cytokines was measured using a whole blood assay. Plasma concentration of 13 cytokines was measured using a high-sensitivity Luminex MagPix assay.

Results: There were observed differences between the two coded supplement conditions. Specifically, total monocyte (CD14+) concentration was significantly greater at 2H-post exercise (P=0.047) with supplement 706. The 706 supplement also boosted LPS-stimulated cytokine production at 2H-post exercise and blunted an increase in plasma cytokines following exercise.

Conclusions: The 223 response is typical for a stressful exercise session, while 706 showed a less pronounced activation of monocytes at post, but improved function at 2H compared to 223. Supplementation with 706 prevented stress-induced monocyte activation immediately and 2-h after exercise. Combined with previous sickness surveillance studies yeast β-glucan supplementation may be a useful countermeasure against exercise-induced immunosupression.

Acknowledgements: This study was funded by Biothera, The Immune Health Company

P.51. The effect of fish oil supplementation on immune function after exercise

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An increase in the consumption of the n-3 polyunsaturated fatty acids (n-3 PUFAs) eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA) has been shown to have immunomodulatory effects, including an increase in Th1 cytokine production [3] and decrease in neutrophil oxidative burst [2]. It has been recommended that athletes consume 1-2 g/day of EPA/DHA [1]. The aim of the current study is to investigate the effect of an increase in EPA/DHA consumption on Th1/Th2 cytokine production and neutrophil function after a intense exercise.

Following local ethics committee approval twelve healthy males (age 25 ± 3 yrs, height 1.85 ± 0.06 m, body mass 87 ± 16 kg; mean ± SD) volunteered to participate in the study. Groups were randomised to a consume 1.3g/day EPA plus 0.3g/day DHA (FO) or placebo (CON) for six weeks. Participants then performed 1 h of cycling at 70%VO_{2max}. Blood samples were collected pre-supplementation, baseline, immediately, 1 h and 3 h post-exercise. Whole blood was used for the analysis of neutrophil phagocytosis and oxidative burst. Peripheral blood mononuclear cells (PBMC) were also extracted and stimulated with concanavalin A for 24 h, after which IL-2, IL-4 and IFNγ were measured using multi-analyte profiling bead Luminex assays. Plasma content of EPA/DHA was also determined. Data were analysed using two-way repeated measures ANOVA, post-hoc t-tests and are presented as mean ± SD.

After six weeks of supplementation with fish oil there were no differences in resting neutrophil function or PBMC IL-2, IL-4 or IFNγ production. Plasma EPA content increased ($P<0.05$) 3-fold, with no significant increase in DHA. Exercise resulted in a decrease ($P<0.05$) in neutrophil oxidative burst at 3 h post-exercise in CON, with a decrease ($P<0.05$) immediately, 1 h and 3 h post-exercise in FO. Exercise resulted in a decrease in PBMC IL-2 and IFNγ production at 1 h post-exercise. At 3 h after exercise IL-2 production was greater ($P<0.05$) in the FO compared to the CON group.

Six weeks supplementation with fish oil potentially results in a greater decrease in neutrophil oxidative burst after exercise, with a greater PBMC IL-2 production.

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Posters: Immunological impact of exercise and the stress response

P.52. Influence of training load on upper respiratory tract infection incidence and antigen-stimulated cytokine production in whole blood culture (ECRA)

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Regular moderate exercise is associated with improved health, while high volumes of strenuous exercise are believed to depress immune function and increase susceptibility to infection [1-3]. The purpose of this study was to examine the effect of training load on upper respiratory tract infection (URTI) incidence in 18-35 year-old men and women engaged in endurance-based physical activity during the winter months and to establish if there are training associated differences in immune function that might help to explain these patterns of illness. Seventy five individuals (44 males, 31 females) provided resting blood and saliva samples for determination of markers of systemic immunity. The latter included the measurement of whole blood culture cytokine production in response to a multi-vaccine stimulant in order to simulate exposure to a pathogen challenge. We hypothesised that an impaired pro-inflammatory cytokine response or an elevated anti-inflammatory cytokine response to the multi-antigen challenge might be associated with a higher risk of URTI infection.

Weekly training and illness logs were kept for the following 4 months. Comparisons were made between subjects who reported that they exercised 3-6 h/week (LOW, n=25), 7-10 h/week (MED, n=25) and those who exercised ≥11 h/week (HIGH, n=25). Our study population was a group of university athletes on a single campus site so that environment and pathogen exposure were likely to be similar for all subjects.

The HIGH and MED groups had more URTI episodes than the LOW group (2.4 ± 2.8 and 2.6 ± 2.2 *vs* 1.0 ± 1.6, respectively: $P < 0.05$). The HIGH group had ~3-fold higher IL-2, IL-4 and IL-10 production (all $P < 0.05$) by antigen-stimulated whole blood culture than the LOW group and the MED group had 2-fold higher IL-10 production than the LOW group ($P < 0.05$). There were no significant differences between the groups for other measured immune variables including the concentrations of plasma and salivary immunoglobulins, numbers of circulating leukocytes, monocytes, neutrophils, lymphocytes, T cells, B cells and NK cells. It is concluded that high levels of physical activity are associated with increased risk of URTI and this may be related to an elevated anti-inflammatory cytokine response to antigen challenge.

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P.53. Exercise differentially affects antigen stimulated cytokine production in whole blood cultures (ECRA)

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The impact of exercise on the immune system and leukocyte functions has been intensively discussed. Many researchers have especially focused on changes of immune cell numbers, functions and a wide range of inflammation regulating cytokines. Several direct measurements or in vitro methods have been used either to determine exercise induced changes of immune cell activities or cytokine patterns [1]. Recently it has been demonstrated that whole blood cultures are one of the most effective ways of measuring immune function as it preserves crosstalk of all blood components [2,3]. Therefore we utilised this model to investigate the capacity of leukocytes to produce cytokines after antigen stimulation before and at different time points after exercise.

Methods: 20 male untrained subjects performed a 60 minute bout of cycling exercise at 80% VO_2max. Blood was drawn with multi-functional tubes (containing 0,1 µg/ml f.c. LPS + SEB) serving as culture vessels before, post, 1, 3 and 24 hours after exercise. Immediately after drawing blood the tubes were incubated at 37°C for a period of 24 hours. Thereafter supernatants were separated from leukocytes by inserting a valve septum. Cytokine concentrations were measured out of the supernatants by a multiplex immunoassay.

Results: Concentrations of CCL-3 and CCL-4 peaked directly after exercise, while we observed a delayed increase of IL-6 and IL-8 1 hour and for IL-1ra and IL-1beta 3 hours after exercise. In contrast IL-2 and IL-4 production was diminished immediately and for IFN-gamma 1 hour after exercise.

Conclusion: Intensive endurance exercise differentially regulates immune cell capacity to produce cytokines after antigen challenge. Leukocyte capacity to produce monocyte chemoattractant proteins was increased early. In contrast production of some lymphocyte stimulating cytokines was temporally suppressed by acute exercise.

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P.54. High-altitude impairs the induction phase of an *in-vivo* T-cell-mediated immune response in humans

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Increased upper respiratory and gastrointestinal symptoms reported at high-altitude may be attributable to alterations in the immune system. A small body of evidence indicates that altitude suppresses *in-vitro* cell-mediated immunity (i.e. decreases CD4+:CD8+ T-lymphocyte ratio and T-lymphocyte proliferation). Nonetheless, the effect of altitude exposure on clinically relevant *in-vivo* immune responses remains unknown. Therefore the objective of this study was to investigate the effect of high-altitude on the induction phase of the T-cell-mediated immune response to a novel antigen, diphenylcyclopropenone (DPCP)[1].

We hypothesised that exposure to high-altitude would impair the normal *in-vivo* T-cell-mediated immune response to a novel antigen. To examine this hypothesis, ten mountaineers on a European Alpine expedition (30% females, Mean ± SD, age 22 ± 3 years, BMI 22 ± 2 $m \cdot kg^{-1}$) were recruited and compared with twenty-seven healthy controls (30% females, age 26 ± 6 years, BMI 24 ± 3 $m \cdot kg^{-1}$). Prior to this study, subjects had not been exposed to altitude for at least 1 year. Having avoided exercise for 24 hours, DPCP was applied (sensitisation) to the lower back of controls in a sea-level laboratory and to mountaineers after they had resided in a cable car station at 3777 m for 24 hours (sensitisation patch = 22.8µl of 0.125% DPCP in acetone[1]). The cable car ensured ascent was rapid but passive. Following sensitization, mountaineers completed typical alpine activities for a range of 11-18 days reaching a maximum height of 4206 m. Exactly four weeks after sensitisation, the strength of immune memory induction was quantified, by measuring the response to a low, dose-series challenge of DPCP, read at 24 and 48 hours as clinical scores of oedema (skinfold thickness) and redness (erythema).

Compared with control responses, oedema and redness were reduced in the mountaineers at 24 and 48 hours (Oedema, -50 and -52%, respectively, $P = 0.03$, $\eta^2 = 0.12$ large effect size; redness, -42 and -36%, respectively, $P = 0.05$, $\eta^2 = 0.10$ medium-large effect size).

These findings indicate that prior high-altitude exposure impairs the induction phase of *in-vivo* T-cell-mediated immunity.

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P.55. Latent cytomegalovirus infection robustly amplifies CD8+ T lymphocyte mobilisation in response to exercise, stress, and isoproterenol infusion

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Introduction: Exercise and psychological stress are known to elicit mobilisation of CD8+ T lymphocytes (CD8TL) into the peripheral blood, partly via β-adrenergic receptor stimulation. Humans show large individual variation in such mobilization responses, which has traditionally been attributed to genetic, physiological, and psychological differences. The data presented here identifies latent Cytomegalovirus (CMV) infection as a novel and major determinant of such immune system responses.

Methods and Results: Employing standard models of acute exercise (treadmill running for 60 min at 80% VO_{2max}; N=14) and mental stress (Trier Social Stress Test; N=60) we found that the CD8TL increase in peripheral blood was fully explained by a selective mobilization CD8TL with an “effector-memory” (EM; CD27−CD45RA−) and “differentiated effector memory” (EMRA; CD27−CD45RA+) phenotype. The EMRA subset numbers increased by 200% and 500% during stress and exercise, respectively ($p<.001$). Subset mobilization was abrogated after administration of the β-adrenergic receptor blocker propanolol (80mg), and replicated by the β-adrenergic agonist isoproterenol (2ng/min/kg) (all $p<.001$). EMRA CD8TL showed a selective 7-fold upregulation of the β2-adrenergic receptor gene (as compared to phenotypically naive cells).

As CMV infection causes dramatic expansion of these responsive EMRA CD8TL cells, we also compared responses between CMV+ and CMV− individuals. CMV+ individuals mobilized 200% (exercise), 400% (stress) and 1800% (ISO) higher numbers of CD8TLs than CMV− individuals (all $p<.001$). Tetramer studies confirmed that CMV-specific CD8TLs were mobilized to a 3 to 4-fold larger extent than EBV-specific, naive, or total CD8TLs ($p<.01$). Moreover, CMV-specific CD8TL showed an 8-fold up-regulation of β2-adrenergic receptor gene.

Conclusion: This is the first study to identify infection history as a major determinant of immune system responses to exercise, stress and adrenergic stimulation.

P.56. Cytomegalovirus infection and acute exercise alters the type 1/type 2 cytokine balance in both low (CD27+) and highly (CD27-) differentiated subsets of CD4+ and CD8+ T-cells (ECRA)

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Acute exercise alters the type1/type2 cytokine balance in total blood T-cells, but it is unknown if this is due to the increased number of highly differentiated T-cells that appear in the blood compartment after exercise. Moreover, because those carrying a latent cytomegalovirus (CMV) infection have a greater frequency of highly differentiated T-cells, the cytokine response to acute exercise could differ with CMV**.**

Purpose: To examine the effects of an acute bout of cycling exercise on the expression of type 1 (IL-2, IL-6, IFN-γ, TNF-α) and type 2 (IL-4, IL-10) cytokines in low (CD27+) and high (CD27-) differentiated T-cell subsets in CMV-infected and non-infected participants.

Methods: Trained male cyclists (Age: 31±6.1 yrs) cycled for 60-min at 95% maximal steady state. Blood samples collected before, immediately after, and 1h after exercise were prepared for cell culture and flow cytometry analysis. CMV serostatus was determined by ELISA.

Resulst: Following mitogen (PHA) stimulation, low differentiated (CD27+) T-cells had a greater expression of IL-2, IL-4, IL-6, and IL-10 than highly differentiated (CD27-) T-cells, whereas a greater proportion of CD27-T-cells expressed TNF-α. Likewise, CMV-infected subjects, who had more CD27- T-cells, had a greater proportion of T-cells expressing TNF-α and IFN-γ. Non-infected subjects had greater proportions of CD27+ T-cells and more cells that expressed IL-6 and IL-10. There was a decrease in the proportion of total CD4+ and CD8+ T-cells expressing IL-2, IL-4, Il-6, IFN-γ, and TNF-α immediately after exercise, whereas the proportion of all T-cells expressing IL-10 increased after exercise. Similar changes in cytokine profiles with exercise were documented for both CD27+ and CD27- cells.

Conclusion: Acute exercise is associated with a shift from a type1 to a type2 cytokine response in both CD4+ and CD8+ T-cells, affecting both low and highly differentiated T-cell subsets. Moreover, individuals with latent CMV infection appear to have a stronger type 1 cytokine response following acute exercise, which could have important implications for CMV-infected athletes.

P.57. The impact of latent CMV and EBV infections on the mobilization of recent thymic emigrants and extrathymic T-cells in response to acute exercise in man

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Background: The thymus gland atrophies with age resulting in a progressively lower output of naïve T-cells during the natural course of aging. As such, there is an increased reliance on the extrathymic maturation of T-cells to maintain a diverse peripheral T-cell repertoire with advancing age. Acute exercise mobilizes T-cells into the blood compartment; however, it is not known if recent thymic emigrants (RTE) or T-cells of extrathymic origin contribute to the lymphocytosis associated with exercise. Moreover, the impact of latent herpesviruses on the frequency or exercise response of these cell types is unknown.

Purpose: To examine the impact of latent cytomegalovirus (CMV) and Epstein-Barr virus (EBV) infections on the mobilization of RTE and extrathymic T-cells in response to acute exercise.

Methods: 22 healthy males (age 21-35yrs) cycled for 30-min at 85% of maximum power. Blood lymphocytes isolated before, immediately after, and 1h after exercise were surface stained with monoclonal antibodies to identify RTE (CD103+/CD62L-) and extrathymic T-cells believed to mature in the liver (CD3+/CD25-/CD22+) and the epithelium of the small intestines (CD3+/CD4-/CD8-; TCR$\gamma\delta$+/ CD8$\alpha\alpha$+; CD3-/CD2+/CD7+). Cell populations were analyzed by 4-color flow cytometry and CMV and EBV serostatus was determined by ELISA.

Results: Proportions of RTE among total CD3+/CD4+ or CD3+/CD8+ T-cell subsets did not change immediately after exercise, but was elevated above baseline 1h later due to the preferential egress of T-cells exhibiting a late stage differentiation phenotype. Neither CMV nor EBV status influenced the proportions of RTE in blood or in response to exercise. T-cells reported to mature in the intestinal mucosa (i.e. CD3+/CD4-/CD8- and CD3-/CD2+/CD7+) were found to increase in blood immediately after exercise, however, baseline proportions and the relative mobilization of extrathymic T-cells with exercise was lower in CMV-infected subjects.

Conclusion: Acute exercise elicits the mobilization of T-cells exhibiting phenotype characteristics of extrathymically matured T-cells, suggesting that these cell types contribute to the lymphocytosis associated with acute exercise. The proportions of extrathymic T-cells and their mobilization in response to exercise was blunted in CMV-infected subjects, indicating that latent viruses shape the frequency and exercise response of extrathymically derived T-cells.

P.58. The effect of sodium bicarbonate ingestion prior to high intensity exercise on blood neutrophil functions and cytokine and stress hormone responses in man

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Introduction: The maintenance of acid-base homeostasis is of critical importance for normal cellular responses and physiological integrity. Sodium bicarbonate ingestion before exercise has been proven to increase extracellular buffering capacity, assist the efflux of H^+ ions from muscle and attenuate the metabolic acidosis associated with intense exercise.

Purpose: This study examined the effects of sodium bicarbonate ingestion prior to high intensity exercise on leukocyte redistribution, neutrophil functions, cytokines balance, and stress hormone responses.

Methods: Nine healthy male softball players (age 21.4±1.9 yrs, height 172.6±6.1 cm; body mass 69.0±6.4 kg, peak power 210.3±16.3 watt) from a school team voluntarily participated in this study. All participants were asked to complete one preliminary peak power test and four main trials, which were Trial A (sodium bicarbonate ingestion + exercise), Trial B (sodium bicarbonate ingestion + rest), Trial C (water ingestion + exercise) and Trial D (water + rest) arranged randomly. Participants were asked to ingest 500 mL of sodium bicarbonate drink with 0.3 g/kg body mass or water 90 min before starting a warm-up on a cycle ergometer at 50% peak power at 70 rpm for a 3-min period followed by 95% peak power at 70 rpm until volitional fatigue. Heart rate was recorded continuously during exercise by radiotelemetry. Venous blood samples were taken 5 min before ingesting the drink (pre), 5 min before exercise (pre EX), immediately post-exercise (post EX), and 15 min post-exercise (post 15) for all trials.

Results: There were significant differences between Trial A and Trial C in cycling times (longer in Trial A), blood pH (higher in Trial A at pre EX, post EX, and post 15), plasma concentrations of lactate (higher in Trial A at post EX) and adrenaline (higher in Trial C at post EX), and circulating lymphocyte counts (higher in Trial C at post 15).

Conclusions: These findings suggest that sodium bicarbonate ingestion 90 min before high intensity exercise may affect blood pH and subsequently increase the capacity to buffer metabolic acidosis. However, it appears not to substantially influence stress hormones, leukocyte mobilization and neutrophil functions.

P.59. The impact of latent CMV and EBV infections on the mobilization of viral-specific and senescent T-cells with acute exercise (ECRA)

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Background: Highly-differentiated and senescent T-cells are preferentially mobilized into the blood compartment in response to acute exercise [1]. This response is amplified in individuals with a latent cytomegalovirus (CMV) infection [2], however, the frequency of CMV-specific T-cells among the mobilized cells is not known. Moreover, latent Epstein-Barr virus (EBV) infection also causes changes to the cellular T-cell compartment, but its impact on T-cell mobilization with exercise has not been considered.

Purpose: To examine the frequency of CMV-specific T-cells among highly differentiated and senescent T-cells mobilized by acute exercise and the impact of CMV and EBV serostatus on exercise-induced T-cell mobilization.

Methods: Healthy male subjects (n=24; age: 28 ± 5yrs) cycled for 30-min at 85% maximum power. Blood lymphocytes isolated before, immediatly after and 1 hour after exercise were labelled with MHC class I Pentamers specific to CMV (pp65: NLVPMVATV) or EBV (EBNA: GLCTLVAML) and identified as having a highly differentiated (KLRG1+/CD28+) or senescent (KLRG1+/CD57+) surface phenotype by 4-color flow cytometry. The frequency of naive (CD45RA+/CCR7+), central memory (CM; CD45RA-/CCR7+), effector memory (EM; CD45RA-/CCR7-) and CD45RA+ effector memory (EMRA; CD45RA+/CCR7-) cells was also determined. CMV and EBV serolostatus was determined by ELISA.

Results: Participant physical characteristics (age, BMI, VO2max) and exercise performance (mean power, heart rate) measures were not influenced by CMV or EBV serostatus. The numbers of both CMV and EBV-specific CD8+ T-cells increased with exercise in the infected participants but accounted for less than 5% of the mobilized highly differentiated and senescent cells. The relative egress of highly differentiated, senescent and EMRA CD8+ T-cells during exercise recovery was amplified in CMV-infected participants. EBV infection was associated with a greater frequency of CM CD8+ T-cells but did not influence cellular responses to exercise.

Conclusion: CD8+ T-cells specific to a single epitope of CMV or EBV are mobilized in response to acute exercise, but these account for a very small number of the mobilized T-cells The extravasation of late stage differentiated CD8+ T-cells during the recovery phase of exercise is amplified with CMV, however, EBV serostatus does not appear to influence T-cell mobilization in response to exercise.

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P.60. Regular, moderate exercise attenuates the stress induced increase in plasma IL1beta, but not TNFalpha, IL-6, IL-10, or corticosterone (ECRA)

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Exposure to acute physical or psychological stressors produces a sterile inflammatory response as evidenced by increases in inflammatory proteins and danger associated molecular patterns (DAMPs) in the blood [1]. Regular, moderate physical activity modulates many aspects of the stress response including changes in behavior, brain, endocrine, and antibody and antimicrobial immune responses [2]. The effect of exercise on specifically the sterile inflammatory response, however, is unknown. The purpose of this experiment, therefore, was to examine the impact of regular exercise on the sterile inflammatory response to an intense acute stressor. Because inflammatory proteins function in a network, we investigated both pro-inflammatory (TNFalpha, IL1beta, IL18) and anti-inflammatory (IL6, IL10) stress responsive proteins and a DAMP (extracellular Hsp72). Given prior reports of the "anti-inflammatory" and the "improved eHsp72-dependent antimicrobial" effects of exercise [2,3], we hypothesized that physically active rats would have an optimal sterile inflammatory response, comprised of constrained inflammatory proteins and facilitated eHsp72 responses.

Adult male F344 rats lived for 6 wks with a mobile running wheel (physically active) or locked wheels (sedentary). Sedentary and physically active rats (10 per group) were sacrificed immediately after exposure to a well-characterized laboratory stressor (0, 50 or 100 unpredictable 1.5mA, 5-s tailshocks) or 2 hours following 100 shocks. Plasma inflammatory proteins and eHsp72 concentrations were measured using ELISA. Plasma corticosterone, a reliable indicator of the stress response and modulator of inflammatory cytokines, was measured using EIA.

Rats with mobile wheels ran 2-3 km per night and had a ~10% reduction in body weight gain compared to sedentary controls. Stressor exposure increased every protein measured (ps<.0.05). Physical activity attenuated stress-induced increases in IL1beta and IL18 but not TNFalpha, IL6, IL10, and corticosterone. When the exercise-induced reduction of plasma IL1beta is coupled with the lack of effect on IL10, an anti-inflammatory profile emerges in the exercised rats. Interestingly, physical activity potentiated the stress-induced increase in eHsp72. This is consistent with reports that physically active stressed rats have better eHsp72-mediated antimicrobial responses than sedentary stressed rats [4].

These data demonstrate that regular moderate exercise modulates the sterile inflammatory stress response seen in the blood.

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P.61. Regular moderate exercise modulates the stress-induced expression of inflammatory proteins in white adipose tissue (ECRA)

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Regular, moderate physical activity modulates many consequences of exposure to intense physical and/or psychological stressors including constraint of stress-induced increases in pro-inflammatory proteins in the blood [1]. Although acute stressor exposure can elevate cytokines in immune tissues, it remains unknown whether stress impacts cytokines in white adipose tissue (WAT), and if physical activity status impacts this response. This is important because, in addition to serving as immunological signalling molecules, inflammatory proteins expressed in WAT may also play a role in metabolic homeostasis [2]. This experiment, therefore, tested if 1) stress impacts white WAT inflammatory proteins 2) the response is depot specific and 3) regular moderate exercise impacts these responses.

Adult male F344 rats either lived for 6 wks with a running wheel (physically active, PA) or with locked wheels (sedentary, SED). SED and PA rats (10/group) were sacrificed immediately after exposure to a well-characterized stressor (0 or 100 1.5mA, 5-s tailshocks) or 2 hours post-stressor termination. IL1beta, IL6, and IL10 were measured in visceral (omental, mesenteric) and non-visceral (subcutaneous, retroperitoneal, epididymal) WAT using ELISAs. Plasma norepinephrine (NE) and corticosterone, stress hormones, were assessed via EIA.

PA rats ran ~2-3 km per night and weighed ~7% less than SED rats. Stress elevated NE and corticosterone in both PA and SED rats. Stressor exposure increased inflammatory proteins in all WAT depots. Stress increased IL1beta more in non-visceral than in visceral, depots. IL6 was higher in non-visceral WAT. The IL6 and IL10 stress responses were similar across depots and equal in PA and SED rats. Interestingly, PA rats had potentiated IL1beta responses compared to SED rats in all depots immediately following stress.

In conclusion, acute stressor exposure and physical activity status impact WAT inflammatory proteins. Although the function of stress-induced inflammatory proteins within WAT remains unclear, these data suggest that WAT depots function uniquely based on their anatomical location. Because regular physical activity results in adaptations aimed at glycogen sparing, we hypothesize that PA impacts the stress-induced inflammatory protein response resulting in sparing of glycogen through increased lipid mobilization, reduced lipid uptake, or another route of metabolic regulation.

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P.65. Combined activity of NA and eHsp72 on human neutrophil function during exercise-induced stress: Role of cAMP

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There is evidence suggesting that extracellular heat shock proteins of 72 kDa (eHsp72) [1] and noradrenaline (NA) [2] can act as danger signals during exercise-induced stress, interacting in the activation of neutrophils. Circulating concentrations of NA [2] and Hsp72 [3,4] determined after exercise stimulate neutrophil function (chemotaxis, phagocytosis, and fungicidal capacity). In addition, the post-exercise concentration of NA increases the expression and release of Hsp72 by human neutrophils, which suggests an interaction between the two molecules in the modulation of neutrophils during exercise-induced stress [5]. Given this context, the aim of the present investigation was to study the combined activity of post-exercise circulating concentrations of NA and Hsp72 on the neutrophil phagocytic process, and to evaluate the role of cAMP and Ca^{2+} as intracellular signals in these effects.

Results showed a synergistic stimulation of chemotaxis induced by NA and eHsp72. However, while NA and Hsp72, separately, stimulated *in vitro* the phagocytosis and fungicidal activity of neutrophils, when they act together they do not modify these capacities of neutrophils. NA and eHsp72 did not modify the intracellular levels of Ca^{2+}. Post-exercise concentrations of NA and eHsp72 separately increased the intracellular level of cAMP in parallel with the stimulation of phagocytosis and fungicidal capacity. However, NA and eHsp72 acting together did not modify the intracellular concentration of cAMP.

These results suggest that cAMP is involved in the autocrine/paracrine physiological regulation of phagocytosis and fungicidal capacity of human neutrophils mediated by NA and eHsp72 in the context of exercise-induced stress.

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P.66. Oxidizing and reducing responses before and after Cooper test in healthy subjects (ECRA)

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The present study was designed to determine the effects of exercise on ROS production, PKC activation, AGEs formation, and antioxidant capacity of plasma in healthy well trained male volunteers.

Plasma and leukocytes were purified using a Ficoll-Hypaque gradient method. ROS were quantified by luminol-dependent chemiluminescence. Total plasma antioxidant status was measure in a MTT dye reduction assay For the analysis of the production of AGEs, plasma malondialdhyde (MDA) concentration was measured by TBARS Assay Kit and ROS production with PKC activation was measured using a PDB (phorbol ester) as activator. Statistical analyses were made with Student's t test and F test**.**

Intense exercise (Cooper test) increased (39.0%) the reactive oxygen species (ROS) generation in leukocytes from well trained athletes. Similar experiments were performed *in vitro* with leukocyte collected before and after intense exercise under stimulation without or with phorbol ester (PDB), respectively. The ROS production expressed as Relative Light units/minute (RLU/min) changed from 3,615±306 (L+PBS) to 6,717 ±502(L+PDB)($p<0.05$) in leukocytes collected before exercise and 5.045± 502 versus 18,073± 1,215 ($p<0.05$) after exercise. The serum concentration of MDA was not altered when the quantification performed before and after exercise were compared ($p>0.05$). In contrast, the total plasma antioxidant status evaluated by MTT dye reduction was significantly decreased in plasma collected after exercise.

Our results suggest that the high intensity of exercise (Cooper test) induces an increase in the oxidizing metabolic response evaluated by the ROS production in the presence or in the absence of PDB and a significant decrease in the plasma reducing response to compensate the oxidizing status and to avoid a typical oxidative stress.

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P.67. White blood cells and their subsets counts in different sport disciplines in Polish athletes

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Physical exercise induces changes in the number of leucocytes and their subsets in the circulating blood [1]. These changes can be influenced by many factors such as: catecholamines, increase in cardiac output or decrease adherence of leucocytes to endothelial tissues [2]. Potential differences between the resting values of white blood cells and their subsets in the athletes compared to untrained people were recently determined[3].

Purpose: The purpose of our study was to establish reference ranges of white blood cell counts and their subsets (neutrophils, lymphocytes and monocytes) for Polish athletes which could be used in diagnostics of physical exercise-acquired immunity disorders.

Methods: The samples were collected from 455 Polish athletes (167 females aged 17.8 ± 3.1 (mean ± SD) and 288 males aged 18.9 ± 3.8 (mean±SD) that were representing seven different sport disciplines: canoeing, handball, judo, rowing, swimming, taekwondo and volleyball. Blood samples were taken after an overnight fast from healthy, medically examined athletes. The haematology results were measured using haematology analyzer (ADVIA 120, Siemens).

Results: White blood cells and their subsets counts were found to be lower in athletes in comparison to reference ranges for untrained people [3]. The lowest counts of neutrophils, lymphocytes and monocytes were found for females in canoeing. For males the lowest neutrophil counts were noted in swimming. The lowest lymphocyte and monocyte counts were found in volleyball. Significant changes in neutrophil counts occured in swimmers compared with handball and volleyball players ($p<0.05$).

Conclusions: Changes in white blood cell and their subsets counts, which were found depend on the sport disciplines. It seems likely that the lower values for different white blood cell counts in comparison to reference ranges for untrained people do not indicate acquired disorders, but are an adaptive response to the physical exercise.

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3. Horn PL et al. *Eur J Appl Physiol* 2010: 110(5): 925-932.

P.70. Salivary cortisol, testosterone and IgA responses to high-intensity cycling before and after a 9-day training period (ECRA)

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Basal circulating cortisol (C), testosterone (T) and secretory immunoglobulin A (IgA) concentrations are suggested as markers of training stress. Extensive studies of basal circulating C and T provide contrasting results with increases, decreases and no changes reported with increased training loads. Salivary IgA secretion rate has been shown to decrease in overtrained athletes [1].

A previous study from our laboratory showed that robust elevations of salivary and plasma C and salivary T occur in response to a short duration (30 min) bout of high-intensity cycling that involved alternating blocks of 1 min at 55% maximum power output (W_{max}) and 4 min at 80% W_{max} (55/80)[2]. The aim of the present study was to examine the exercise-induced salivary C, T and IgA response to a high-intensity, short duration cycling protocol before and after a period of increased training load. The cycling protocol in this study was a 55/80 bout followed 2 h later by a 30 min cycle at 70% W_{max} (70).

Ten healthy male participants (mean (±*SD),* age: 25 (4) years, body mass: 75.7 (9.6) kg, height: 176 (8) cm, $\dot{V}O_{2peak}$: *51.6 (5.5)* ml.kg^{-1}.min^{-1}) completed the cycling protocol before and after a 9-day training period involving 1.5 h cycling at 75% $\dot{V}O_{2peak}$ per day (a 125% increase in hours of exercise completed over a normal 9-day period). Saliva samples were collected pre-, post- and 30 min post both the 55/80 and 70 bouts. Recovery-stress Questionnaires (REST-Q) were completed before the 55/80 bout.

Burnout and Fatigue REST-Q scores increased from pre- to post-training. Post-training sIgA secretion rate increased with a blunted salivary C and T response to both the 55/80 and 70 bouts. Pre- to post-training sIgA secretion rate increased 83% (35 to 64 ug.min^{-1}) (55/80) and 18% (55 to 65 ug.min^{-1}). The absolute change of exercise-induced salivary C decreased 72% (11.05 to 3.05 nmol.L^{-1}) (55/80) and 38% (7.02 to 4.38 nmol.L^{-1}) (70) and salivary T decreased 37% (407 to 258 pmol.L^{-1}) (55/80) and 42% (473 to 274 pmol.L^{-1}) (70).

The blunted hormonal response is likely to be due to the development of feedback inhibition of anterior pituitary hormone release arising from repeated exercise-induced elevations of circulating C and T. The sIgA secretion rate increase suggests the participants did not experience depression of mucosal immunity despite the signs of them being overreached following the training period. These results also suggest that the 55/80 exercise bout could be used as a reliable tool in the examination of hormonal changes that occur due to increased training stress.

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P.72. Curcumin attenuates oxidative stress via suppression of MCP-1 production following downhill running induced muscle damage (ECRA)

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Background: Downhill running including eccentric muscle contraction causes muscle damage, and induces oxidative stress and inflammatory reaction. In addition, it is shown that macrophages and neutrophils are present in skeletal muscle after downhill running. Therefore, macrophages and nuetrophils may play an important role in oxidative stress and inflammatory reaction. Recently, curcumin, an important constituent of curcuma longa, possesses anti-oxidant and anti-inflammatory reaction. Interestingly, it is shown that curcumin reduces several inflammatory cytokine concentrations in skeletal muscle after downhill running of mice. However, it is not known whether curcumin affect oxidative stress after downhill running induced-muscle damage. Therefore, the purpose of this study was to investigate the effects of newly developeed nano-particle curcumin (named THRACURMIN) on oxidative stress following downhill running induced-muscle damage. In addition, we investigated whether curcumin affects macrophage infiltration via chemokines such as MCP-1 and CXCL14 expression.

Methods: Male C57BL/6 mice were divided into four groups; rest (n=12), rest+ curcumin (n=12), downhill running (n=14), or downhill running + curcumin (n=14) group. Downhill running mice ran at 22 m/min, −15% grade on the treadmill for 150min. Curcumin (3mg) was administered in oral administration immediately after downhill running. Gene expressions in gastrocnemius were evaluated by real time-RT-PCR.

Results: Creatine kinase (CK) and lactate dehydrogenase (LDH) activity as a muscle damage marker in plasma were significantly affected by downhill running. However, CK and LDH activity were not affected by curcumin administration. Hydrogen peroxide concentration and NADPH-oxidase mRNA expression in the downhill running mice was significantly higher than that in the rest mice. However, hydrogen peroxide concentration and NADPH-oxidase mRNA expression was significantly attenuated by curcumin administration in downhill running mice. In addition, mRNA expressions of MCP-1 and CXCL14 , F4/80 reflecting presence of macrophages in the downhill running mice were significantly higher than those in the rest mice. However, MCP-1 and F4/80 mRNA expression were significantly attenuated by curcumin administration in downhill running mice. On the other hand, CXCL14 mRNA expression was not affected by curcumin administration in downhill running mice.

Conclusion: Curcumin may attenuate oxidative stress via suppression of MCP-1 production following downhill running-induced muscle damage.

P.73. The effect of increased training volume on oxidative stress

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Regular physical activity can confer a number of positive health benefits, including a decrease in the risk of coronary heart disease and improvement in the blood lipid profile [1-3]. The purpose of the present study was to examine the influence of an increased training volume on markers of oxidative stress and antioxidant defence.

Six moderately trained subjects (four males and two females) aged 25 ± 5 years of age (mean ± SD) who prior to the study were undertaking 4.55 ± 1.52 hours of training participated in the eight-week investigation. Training volume increased by 2 hours per week over the research period; subjects completed a 30 km exhaustive cycling time trial (TT_{30}) each fortnight to assess adaptation to the increased training volume. Resting venous blood was sampled at the commencement of the study and fortnightly at rest and post TT_{30}. Blood was analysed for markers of oxidative stress (malandialdehyde; MDA) and antioxidant defences (glutathione peroxidase (GPX), catalase (CAT) and superoxide dismutase (SOD)).

Resting and post TT_{30} concentration of MDA and CAT significantly increased in response to the eight-week training program ($p < 0.05$). However, there was no change in the activity of GPX or SOD at rest or post TT_{30}. The increase in training hours was positively related to the change in both resting levels ($r = 0.44$; $p = 0.015$) and post TT_{30} production of MDA ($r = 0.6$; $p < 0.001$). Time to complete the TT_{30} significantly improved in the first 4 weeks of training but remained unchanged in the last 4 weeks.

These data showed that a sustained increase in the resting concentration of MDA in response to an increase in training volume might have a role in the identification of training maladaption.

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P.74. Prolonged depletion of antioxidant capacity following ultra-endurance exercise (ECRA)

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The purpose of this study was to examine the short and long-term (up to 1-month) impact of an ultra-endurance running event on redox-homeostasis. Markers of oxidative stress and antioxidant capacity in peripheral blood were assessed following a single-stage 233 km (143 mile) running event. Samples were collected from nine men (mean age ± SD; 46.1 ± 5.3 years; body mass index 24.9 ± 2.3 $kg \cdot m^{-2}$, maximal-oxygen uptake 56.3 ± 3.3 $ml \cdot kg^{-1} \cdot min^{-1}$). Peripheral blood lymphocytes were assayed for non-specific DNA damage (frank strand breaks) and damage to DNA caused specifically by oxidative stress (formamidopyrimidine DNA glycosylase (FPG)-dependent damage). Protein carbonyls and lipid peroxides were assessed in plasma. Reduced glutathione (GSH) was measured in whole blood.

Lymphocyte frank strand breaks were elevated above baseline at 24 hours post-race ($p < .001$). FPG-dependent oxidative DNA damage was increased immediately post-race ($p < .05$). Protein carbonyls remained elevated for 7 days following the race ($p < .04$) whereas lipid peroxides were increased for 24 hours ($p < .05$) and fell below baseline 28 days later ($p < .05$). GSH, a measure of antioxidant capacity also showed a biphasic response, increasing by one third post-race ($p < .01$) and falling to one-third of baseline levels 24 hours later ($p < .001$). GSH remained depleted to approximately two thirds of pre-race values 28 days post-race ($p < .01$).

Ultra-endurance exercise causes oxidative stress, which persists for at least 1 calendar month depending on the specific biomarker examined. These results suggest that ultra-endurance events are associated with a prolonged period of reduced protection against oxidative stress.

P.75. Effects of physical exercise on immune function in children with acute lymphoblastic leukemia (ECRA)

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Introduction: Leukemia accounts for 30% of cancer in children 0 to 14 years and 25% of cancers occurring before age 20. Acute lymphoblastic leukemia (ALL) is the most common childhood cancer [1]. Chemotherapy treatments cause immunosuppression, leading to an increase in susceptibility to infections, requiring reconstitution of immune function after treatment. Neutropenia is one of the most common and potentially serious complications of therapy for malignancy. Appropriate supportive interventions are needed to address the immunosuppression in children receiving treatment for cancer [2,3].

Aim: To investigate the effect of different acute bouts of exercise on immune function in children with ALL.

Methods: The study population consists of 20 children and adolescents aged 7 to 18 years receiving maintenance treatment according to the Dutch DCOG ALL 10 protocol, medium risk. In these patients a graded exercise test for measuring peak oxygen uptake and three different exercise training sessions will be performed. Moreover, the physical activity levels will be determined using a questionnaire and an activity monitor. Blood samples will be collected pre-exercise, immediately post-exercise, and 1-hour post-exercise. Blood counts, lymphocytes subsets, Natural Killer cell cytotoxic activity (NKCA) and oxidative burst of neutrophils will be assessed.

Results For now only one pediatric ALL patient has been tested and other patients are scheduled. To test the feasibility of this study, a pilot was performed in five healthy adults. Besides logistic feasibility of the functional assays, increased numbers of lymphocytes and NK cells have been observed, resulting in higher NKCA. No results have yet been obtained regarding oxidative burst of neutrophils.

Conclusion: For children with cancer, physical activity is beneficial, but only if the "exercise dose" does not exacerbate underlying inflammatory status abnormalities. Guidelines for adults, with different exercise modes and intensities might not be usable for the pediatric population. It has become abundantly clear that the biologic mechanisms linking exercise to health in children are multifactorial. Optimal levels and safe limits of exercise in the growing child with cancer should be identified. This would result in a personalized training advice based on immune parameters.

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P.76. Effects of physical exercise on immune function in patients with cancer: A systematic review (ECRA)

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Introduction: Current research shows that exercise training has an important role in the rehabilitation of cancer patients and survivors [1,2]. More evidence on the acute and chronic effects of exercise on immune function is needed to establish the safety of exercise and its optimum to rule out adverse effects on the compromised immune system of patients with cancer.

Aim: To summarize the changes in selected immune parameters after acute and chronic exercise in adult and pediatric cancer patients.

Methods: A systematic literature search was performed in Pubmed (Medline), Embase, Cochrane Library and CINAHL (up to 13th of July 2010). Randomized (un)controlled intervention studies in patients with cancer were included. Interventions consisted of acute bouts of exercise or chronic aerobic and resistance exercise sessions of different frequencies and durations. Immune parameters were the main outcome measures. Two independent researchers assessed the methodological quality of the articles using a modified PEDro scale. A best-evidence synthesis was performed by assigning different levels of evidence to the included studies.

Results: The systematic search yielded 516 articles. In total 17 articles met the inclusion criteria, of which three were in children and 14 in adults. Of these, one investigated acute exercise in children, whereas 15 pertained chronic endurance exercise training studies, and one study combined acute and chronic exercise in adults. Seven studies were of high quality, ten of low quality. Although the findings were not always consistent, the amount of various immune cells (e.g. T lymphocytes, neutrophils) and their functions (e.g. NK cytotoxic activity, lymphocyte proliferation) increased due to exercise.

Conclusion: The results of this review indicate that it is safe and beneficial to conduct exercise interventions in the cancer population. The exercise training-induced response appears highly dependent on the type, duration and intensity of the exercise intervention. Overall, more research is needed to get the insights of the mechanism behind exercise effects on immune function, especially in the pediatric population, and to link immune parameters with clinical outcomes. Future studies should include larger study populations, use appropriate control groups, and a standard collection of outcome measures at different time points to improve comparability between studies.

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P.77. Latent cytomegalovirus infection alters NK-cell phenotypes and blunts their mobilization in response to acute exercise

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NK-cells and γδ T-cells are cytotoxic effectors that are preferentially mobilized in response to acute stress and exercise. While infection history is known to alter the phenotype and exercise responsiveness of CD8+ T-cells, the influence of latent Cytomegalovirus (CMV) and Epstein-Barr virus (EBV) infections on the phenotypes and exercise responsiveness of NK-cells and γδ T-cells are not known.

Purpose: To examine the impact of latent CMV and EBV infections on the mobilization of NK-cells and γδ T-cells in response to acute exercise.

Methods: Twenty healthy males (Age: 28.4±5.4 yrs) cycled for 30-minutes at 85% maximum power. Blood lymphocytes isolated before, immediately after, and 1-hour after exercise were assessed for surface expression of CD3, CD4, CD8, CD56, CD57, CD158a, the killer-cell lectin-like receptor G1 (KLRG1), and γδ-TCR by four-color flow cytometry. CMV and EBV serostatus was determined by ELISA.

Results: Compared to the non-infected, CMV-infected participants mobilized much fewer (+105% vs. +200% increase) numbers of NK-cells in response to exercise, despite having similar NK-cell counts at baseline. CMV-infected also had lower proportions of NK-cells expressing the inhibitory receptors KLRG1+ and CD158a+ and higher proportions of terminally differentiated NK-cells (KLRG1-/CD57+). CMV blunted the mobilization of all NK-cell subsets (CD56dim, CD56bright, KLRG1+ and CD158a+) with exercise ($p<0.05$), except KLRG1-/CD57+ NK-cells ($p = 0.22$). EBV infection was associated with a higher proportion of cytotoxic CD8+ NK-cells but did not influence cellular exercise responses. Neither CMV nor EBV infection influenced the frequency or exercise responsiveness of γδ T-cells. The mobilization of γδ T-cells accounted for 35% and 10% of the exercise-induced increase in KLRG1 expressing CD4+ and CD8+ T-cells respectively.

Conclusion: Latent CMV infection is associated with a down-regulation of the inhibitory receptors KLRG1 and CD158a and blunts NK-cell mobilization in response to acute exercise. This may indicate a compromised immune response to “fight-or-flight” situations in those infected with CMV.

P.79. Lymphocyte subset responses to an acute bout of intense exercise following a night of sleep disruption (ECRA)

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Background: Lymphocyte subsets that demonstrate high cytotoxic capabilities (Natural Killers (NK) cells, KLRG1+/CD8+ T-cells, γδT-cells) are highly responsive to acute exercise, which may suggest a role for cytotoxic T-cells during post-exercise immune surveillance. Sleep disruption/deprivation (DS) is known to alter biological rhythms and catecholamine responses to acute stress. As the mobilisation of cytotoxic lymphocytes with exercise is governed by β^2 adrenergic receptors and catecholamines, DS could alter lymphocyte trafficking in response to exercise.

Purpose: To determine if a night of disrupted sleep (DS) alterers cytotoxic lymphocyte mobilisation and extravasation in response to acute exercise.

Methods: Ten male cyclists (age (27 ± 8 years), height (176 ± 7 cm), mass (74 ± 8 kg) performed a 40k TT on a cycle ergometer. Using a randomised cross-over experimental design, participants completed two further trials cycling for 1hour at 90% of the mean wattage obtained from the 40km TT, following either a night of DS (woken every hour of the night over an 8 hour period) or a night of undisrupted sleep (US) (left undisrupted for an 8hour period). Heart rate (HR) was recorded during trials and the Epworth Sleepiness Scale (ESS) was completed before the trials. Blood lymphocytes were isolated before, immediately after, and 1 h post exercise and assessed for cell surface expression of CD45RA, CD45RO, γδ TCR and KLRG1, and lymphocyte subset markers (CD3, CD4, CD8, CD56) by 4-colour flow cytometry.

Results: Following the DS trial, sleepiness and rating of perceived exertion (RPE) was elevated during exercise, whereas average HR was lowered. Numbers of all lymphocyte subsets (CD3+, CD3+/CD4+, CD3+/CD8+, CD3-/CD56+) increased with exercise. Baseline lymphocyte counts and lymphocyte subset counts were unaffected by DS, however, total lymphocytes and CD3-/CD56+ NK-cells were mobilized in greater numbers in response to exercise following DS compared to US.

Conclusion: One night of sleep disruption lowers the heart rate response and amplifies the mobilisation of NK-cells in response to acute exercise. These data indicate that altered sleep patterns could interfere with the trafficking of cytotoxic lymphocytes in response to acute exercise and might play a role in athlete infection susceptibility.

P.80. Highly differentiated T-cells mobilized by acute exercise have a greater tissue migrating phenotype and an increased susceptibility to apoptosis (ECRA)

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Background: Highly differentiated CD8+ T-cells are preferentially mobilised into the blood compartment in response to acute exercise, which is followed by a rapid egress during the early stages of exercise recovery [1]. It has been postulated that regular exercise may serve as a way to expand the naïve T-cell repertoire by inducing selective apoptosis of highly differentiated T-cells [2]; however, the homing destination and apoptosis susceptibility of these cells following their egress from the blood is unknown.

Purpose: To determine the tissue migratory patterns and apoptosis susceptibility of low (CD28+/CD57-), intermediate (CD28+/CD57+) and highly differentiated (CD28-/CD57+) CD8+ T-cells mobilized by acute exercise.

Methods: Ten healthy males cycled for 30-minutes at 85% maximum power. Lymphocytes isolated from blood samples obtained before, immediately after and 1h after exercise were surface stained for 4-color flow cytometry analysis. The expression of surface homing receptors specific to lymphoid tissue (CD62L), skin (CCR4), intestines (CCR9) and general areas of inflammation (CCR5) were determined on CD8+ T-cells stratified by their stage of differentiation (CD28/CD57 combinations) in response to exercise. The surface expression of CD95 (fas/Apo-1) was also determined on the T-cell subsets as an indirect measure of apoptosis susceptibility.

Results: Exercise evoked a greater mobilization (immediately after) and subsequent egress (1h after) of highly (85% increase; -55% decrease) compared to intermediate (65% increase; -38% decrease) and low (32% increase; -15% decrease) differentiated T-cells. Intermediate and late differentiated T-cells expressed more CCR4, CCR9 and CD95 but less CD62L than early differentiated cells. CCR4 expression increased and CD62L expression decreased on total CD8+ T-cells with exercise, however exercise did not alter their expression on the differentiated T-cell subsets, indicating that this was due to proportional changes in the differentiated cell subsets. CD95 expression was not affected by exercise.

Conclusion: Highly differentiated CD8+ T-cells mobilized by acute exercise display a peripheral tissue (i.e. skin, intestines) migrating phenotype and have a higher surface expression of the apoptosis regulator CD95. However, the expression of these surface receptors was unaffected by exercise suggesting that exercise evokes the preferential mobilization of highly differentiated T-cells with a pre-existing tissue migrating phenotype and increased susceptibility to apoptosis.

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P.82. Exercise enhances the immune response against *Leishmania major* (ECRA)

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The Leishmaniasis represent a group of diseases caused by protozoa from the genus *Leishmania* [1]. During the disease progression, the cellular immune response (Th1) influences the control of infection while the humoral response (Th2) relates to the progression of the disease [2]. Several studies suggest that moderate exercise influences the immune system by stimulating the Th1 response [3,4] that controls parasitic infections [2]. We evaluated the impact of moderate aerobic exercise on the progression of infection by *Leishmania major* in mice.

Animals were grouped into cohorts (N=8) according to the following variables: exercise, infection and treatment with Glucantime®. Infection with *Leishmania* was initiated by an inoculation with 2 x 10^7 promastigotes into the plantar cushion. Moderate exercise consisted of swimming with progressive weights related to the corporal index (25 min, 3 days/week). Exercise intensity was measured by blood lactate levels and TBARS assays. Blood lactate levels after a round of moderate exercise (~3.0 mmol/L) were significantly higher than the resting value (~1.6 mmol/L) and lower than levels after intense exercise (~6.0 mmol/L). TBARS indicated that lipid peroxidation was not significantly different between the exercise and control groups. Glucantime® treatment (5 days/week) commenced 6 weeks after infection using therapeutic doses (8 mg/kg).

After 12 weeks, the lesions in the exercise and Glucantime® treated groups were reduced approximately 93%. Only the trained groups presented a DTH response. The parasitic load in the trained groups was ~1,000 fold less than the infected control group and in the group trained from the beginning of infection, 12,000 fold less. The cytokines IL-12 and IFN-γ also were measured in infected legs. Their concentrations were significantly higher in exercise groups compared with infected control group. These data suggest that exercise modulates the Th1 immune response in mice infected with *L. major* and provides a protective response against this parasitosis.

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P.83. Impaired autonomic nervous system innervation has an impact on the mucosal immune response to exercise in wheelchair athletes (ECRA)

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Introduction: It is well known that saliva composition and secretion of sIgA can be modified by both parasympathetic and sympathetic nerve stimulation. As persons with tetraplegia represent a model with no centrally mediated sympathetic nervous control, it was of particular interest to examine the effects of level of spinal cord injury. Therefore, the purpose of this study was to examine salivary secretory immunoglobulin A (sIgA) responses and α-amylase activity following constant load and intermittent exercise in elite wheelchair athletes with varying levels of disability.

Methods: Twenty-three wheelchair athletes divided into three groups (8 tetraplegic (TETRA), 7 paraplegic (PARA) and 8 non-spinally injured (NON-SCI) performed two randomised and counterbalanced 60-min bouts of exercise on a treadmill. These consisted of a constant load (60% peak oxygen uptake ($\dot{V}O_{2peak}$)) and an intermittent (80% and 40% $\dot{V}O_{2peak}$) exercise bout. Timed, unstimulated saliva samples were obtained pre, mid, post, and 30 min post exercise and analysed for sIgA and α-amylase. Furthermore, oxygen uptake, blood lactate concentration and rating of perceived exertion (RPE) were measured during both sessions.

Results: SIgA secretion rate and α-amylase activity were increased during exercise in all groups ($p<0.05$). However, the increase of sIgA secretion rate during exercise was greater in TETRA individuals (post exercise average data for both trials in comparison to pre: TETRA +60±31%, PARA +30±35%, NON-SCI +11±25%, $p<0.05$). Yet, groups were comparable with respect to the α-amylase response, blood lactate concentration and RPE for both conditions.

Discussion: The disruption of autonomic salivary gland innervation in TETRA athletes seems to result in an altered sIgA response. However, their ability to increase sIgA secretion rate is comparable with wheelchair athletes with intact autonomic salivary gland innervation. This may stem from sympathetic reflex activity during exercise and/or a predominant contribution of parasympathetic activity to increase sIgA, as these are still intact systems in the TETRA population. The similar α-amylase responses between groups indicate that α-amylase may not be suitable to be used as a biomarker for sympathetic central drive. Finally, these results support the positive role of acute exercise on oral immune function in wheelchair athletes independent of disability type.

Posters: Technical Advances and Novel Approaches in Immune Assessment

P.84. Multiplexed measurement of cytokines in saliva with a high sensitivity Evidence biochip array

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Cytokines normally function as part of a complex network and are investigated in multiple fields of research as they play central roles in many biological processes. Cytokines are present in saliva, its use as analytical medium is attractive, as the collection methods are simple and non-invasive. Biochip array technology enables multi-analyte profiling with a single sample, which provides more information on the cytokine network than single-analyte determinations. The miniaturization of the simultaneous assays on the biochip surface, leads to the reduction of sample/reagent consumption and implications in the cost-effectiveness of the tests. We report here the applicability of a high sensitivity cytokines biochip array to the multiplexed measurement of 12 cytokines in oral fluid.

Simultaneous chemiluminescent immunoassays are employed and the capture antibodies, specific for the analytes, are bound and stabilised in predefined positions on the biochip surface (9mm x 9mm) defining microarrays of test sites. Assay specific reagents and sample are applied to the biochip and incubated under controlled conditions. Signal detection, data processing and storage were carried out using the semi-automated bench top analyser Evidence Investigator.™

All the analytes were measured in picogram quantities with sensitivity ranging from 0.12pg/ml for IL-6 (calibration range 0-400pg/ml) to 2.12pg/ml for IL-4 (calibration range 0-450pg/ml). The intra-assay and inter-assay precision (n=20) expressed as %CV were typically $\leq$12 for three multianalyte control levels. When 20 apparently normal oral fluid samples were analysed, ten out of twelve cytokines were simultaneously measured in all the samples. Levels of the two remaining cytokines (IL-2, IFNγ) were reported in 90% and 45% of the samples, respectively.

The data indicates that this technology is applicable to the simultaneous high sensitivity measurement of cytokines in oral fluid, maintaining a broad calibration range with good precision. It represents a very applicable tool for research as a sample profile can be obtained at a single point in time.

P.85. Investigating the use of a Point of Care sIgA test in the sporting environment

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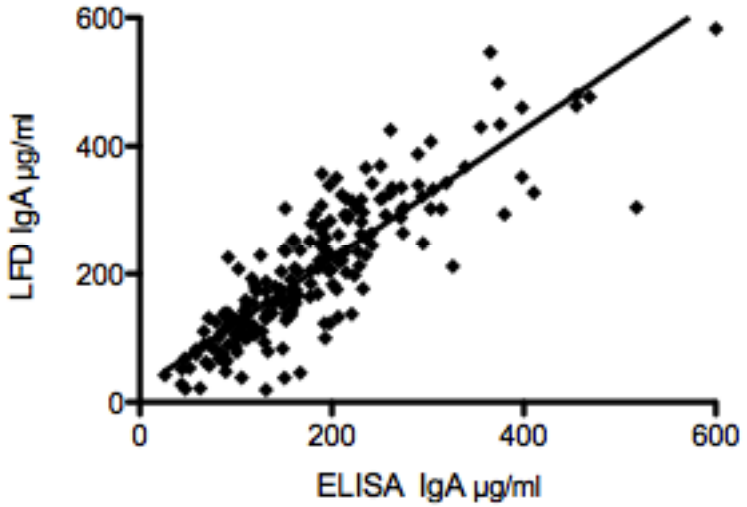
Introduction: The use of salivary diagnostics within the sporting community has gathered momentum in recent years; identifying hormone levels to assist in the optimisation of workloads, or antibodies such as sIgA to assess individual recovery status and potential immune suppression. Immediate feedback for coaching and support staff via a Point of Care test would give a significant time advantage over standard laboratory techniques, which often reveal data to sporting squads only days later. This paper assesses a new point of care product for the rapid determination of sIgA in comparison to standard laboratory ELISA determination.

Methods: A total of 208 saliva samples were taken from a cohort of 21 English Premier League soccer players (26.4 ±4.2 yrs) using IPRO OFC collection kits. The samples were taken during routine monitoring: before training sessions, as well as before, straight after and the day after Premier League matches, thus giving a wide range of concentrations. The same samples were assessed to determine sIgA concentrations via laboratory ELISA and a real-time Lateral Flow Device (LFD), which can give a quantitative reading within 6 minutes of sample collection. In both methods a variety of batch Lot Numbers were used.

Results: sIgA concentrations measured via ELISA ranged from 43.9-598.8 µg/ml and with the LFD from 27.7-628.0 µg/ml, with the mean difference 23.22 µg/ml. The relationship between the sIgA values obtained using the ELISA and LFD was represented by the formula: $y = 1.011x + 30.5$, with R^2 0.757.



Conclusion: The point of care test shows good agreement with the ELISA method for the determination of sIgA. Given the quick data turnaround and efficiency in terms of cost, it represents a suitable alternative method for use in sports teams.

Posters: Age & Gender Issues in Exercise Immunology

P.86. Association of exercise-mediated fitness and body composition with immune response to influenza vaccine in elderly women

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Although Immune function declines with advancing age [1], exercise may be able to improve immune function [2]. This research analyzed the changes of blood and saliva IgA and cortisol concentration, and physical fitness by administering respectively for 12 week yoga exercise or recreation dance after influenza vaccination to healthy aged women. Additionally, this study was to examine whether cardiovascular fitness was associated with immune responsiveness of T-lymphocyte to influenza vaccine after regular aerobic exercise in older women.

Subjects were composed of 27 healthy aged divided into recreation dance group (N=10), yoga group (N=7), and control group (N=10). Recreation dance group practiced exercise program three times a week for totally 12 weeks, and yoga group performed yoga motions with focus on stretching 3 times a week for totally 12 weeks, but control group did not perform exercise program. Subjects were measured body composition, VO_2max of graded treadmill testing, blood concentration of IgA, IgG1, IgG2, IL-10 and IFN-γ concentration, saliva IgA and cortisol concentration at pre, mid and post-exercise. The relationship of change of cardiovascular fitness with immune responsiveness of T-lymphocyte to influenza vaccine after regular aerobic exercise program was analyzed by Pearson's correlation coefficients.

Body composition items, IgA and cortisol concentrations showed no significant changes after exercise intervention in all groups. However, yoga group showed more positive changes of S-flow rate and SIgA secretion rate than recreation dance group. Regarding health-related physical fitness items, grip strength and balance tended to increase in recreation dance group compared to yoga group, and muscular endurance, agility and heart rate recovery rate etc tended to increase in both exercise groups. Blood concentration of IFN-γ and IL-10 showed no significant difference after 12 week exercise program in all groups. Exercise groups showed the increased tendency of blood concentration of IgG1 and IgG2 after exercise program. Recreation group showed a significant increase of VO_2max after exercise program. This increase of VO_2max showed a significant relationship with the activation of blood IgG1 concentration [3,4] as compared to no activation cell-medited response [5]. Although recreation dance and yoga programs are judged to make no significant effects on immunity variables, yoga program showed positive effects to S-flow rate and SIgA secretion rate.

In conclusion, the improvement of cardiovascular fitness was associated with cell-mediated immune responsiveness of T-lymphocyte to influenza vaccine after regular aerobic exercise in older women.

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P.87. Intestinal lymphocyte apoptotic protein expression after acute treadmill exercise in young and old mice (ECRA)

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Introduction: Variations in apoptotic protein expression may contribute to acute exercise-induced immunosuppression. In young mice, acute treadmill exercise increases pro-apoptotic protein expression in intestinal lymphocytes (IL) due to oxidant stress [1]. Whether a similar response occurs in the IL of old animals is not known.

Purpose: We examined the effects of a single bout of treadmill running in young and old mice on IL expression of the apoptosis-inducing cytokine TNF-α, the pro-apoptotic proteins caspase-3 and 7, the anti-apoptotic protein Bcl-2, and IL apoptotic status (AnnexinV^+).

Methods: Young (3-4 months, n= 44) and old (13-14 months, n= 45) female C57BL/6 mice were randomized to treadmill exercise (10 min warm-up, 20 min at 22 m/min, 30 min at 25 m/min, 30 min at 28 m/min, 2º slope) with sacrifice immediately (IMM) or 2hr after (2Hr), or to a non-exercised control (SED). IL were removed and prepared for analysis of % apoptosis (flow cytometry) and determination of apoptotic protein and cytokine expression (Western blotting). Plasma corticosterone and 8-isoprostanes were measured by EIA.

Results: Exercise was associated with a higher IL expression of caspase-3 in IMM and 2Hr vs. SED ($p<0.001$), a higher expression of TNF-α in IMM vs. SED ($p<0.001$), and a lower Bcl-2 expression in IMM and 2Hr vs. SED ($p<0.01$) mice. There was a trend ($p=0.07$) for increased caspase-7 expression after exercise. IL caspases 3 and 7 and TNF-α expression did not differ by age whereas Bcl-2 expression was lower ($p<0.001$) and % Annexin V^+ IL was higher ($p<0.05$) in old vs. young mice. Plasma corticosterone and 8-isoprostanes were higher ($p<0.001$ and $p<0.05$) in IMM vs. SED mice but did not differ by age.

Conclusions: The expression of the pro-apoptotic proteins, caspase-3 and 7, and the apoptosis-inducing cytokine, TNF-α, in mouse IL does not differ by age in response to a single treadmill challenge. However, older mice had a lower expression of the 'protective' anti-apoptotic protein Bcl-2 and a higher percentage of early apoptotic IL. Whether repeated exercise challenge compared to a single bout results in less IL resiliency in old compared with young mice remains to be determined. [Support-NSERC Canada]

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P.88. The effect of exercise training on gastrointestinal tract in aged mice (ECRA)

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Age-associated dysreguretion of the immune system of gastrointestinal (GI) tract has been well documented for secretory (S)-IgA immunity [1]. Exercise training prevent age-associated dysreguretion of the immune system, especially many reports has about saliva S-IgA [2] and it thought it is similar in the GI tract [3]. Therefore, we hypothesize exercise training prevent age-associated dysreguretion of the immune system of GI tract as mucosal immunity in oral. In this study, we focused on cryptdin-4 (Crp4) which are secretrd into the intestinal lumen and has the most potent microbicidal activity [4]. However, the effect of exercise training in Crp4 are poorly undrestood. Thus, the aim of this study is whatever exercise training prevent age-associated dysreguretion of the immune system of GI tract.

Young adult (8 weeks old) and aged at 15-16 momths C57BL/6 mice were divided into four groups: Young adult sedentary (YS), Young adult training (YT), Aged sedentary (AS), Aged training (AT). Training group performed treadmill running (20m/min, 30min, 3 times/week). The mice were euthanized at 48 h after the last training session, and the intestine was removed. The expression of Crp4, IgA and caspase-3 in intestine was assessed by real-time polymerase chain reaction, ELISA and western blot assay. The mRNA level of Crp4 in Young mice significantly was higher than the level in Adult ($P< 0.05$). Furthermore, AT tended to increase compared with AS (P=0.09, effect size d=.1.1 large). IgA in intestine was not significantly changed by either training or age. Caspase-3 in AT was significantly lower than AS ($P< 0.05$).

It suggested Cry4 reduced by aging, exercise training in aged mice may prevent this decrease. In addition, the inhibit of apoptosis by exercise training protect the apoptosis of paneth cell secreted Cry4. Therefore, the mRNA of Cry4 in AT might be higher than that of Cry4 in AS.

Exercise training could be helpful for age-associated dysreguretion of the immune system of GI tract.

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(**S** = Symposium, **O** = Oral Presentation, **P** = Poster Presentation)

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