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平成 28 年度杉山産業化学研究所研究助成報告

龍谷大学農学部 石原健吾 准教授

(学会発表)

- 1) 石原健吾、秋山奈生、畑ひろみ、伏木亨、「摂取した糖質の最大利用能力についてのマウスを用いた検討」、第 70 回 日本栄養・食糧学会大会、2016. 5. 15 武庫川女子大学中央キャンパス  
URL: [https://member.jsnfs.or.jp/eishoku\\_db/index.php?t\\_search\\_seq=6591](https://member.jsnfs.or.jp/eishoku_db/index.php?t_search_seq=6591)  
<添付書類> 講演スライド (タイトル及び謝辞)
- 2) 石原健吾、「滑り止め処理を施工したトレッドミルでのランニングエコノミー、最大酸素摂取量の測定」、第 71 回 日本栄養・食糧学会大会、2017. 5. 20 沖縄コンベンションセンター  
URL: [https://member.jsnfs.or.jp/eishoku\\_db/index.php?t\\_search\\_seq=9355](https://member.jsnfs.or.jp/eishoku_db/index.php?t_search_seq=9355)
- 3) K Ishihara, "FatMax test during hilly running exercise in mice", International Sports and Exercise and Nutrition Conference, Newcastle upon Tyne, UK, 2016. 12. 18  
International Journal of Sport Nutrition and Exercise Metabolism, 2017, 27, S1-S20  
URL: <http://journals.humankinetics.com/doi/pdf/10.1123/ijsnem.27.s1>
- 4) Ishihara K, Taniguchi H, Asami Y, "Rice gel as a carbohydrate source during endurance exercise", International Sports and Exercise and Nutrition Conference, Newcastle upon Tyne, UK, 2017. 12. 18  
International Journal of Sport Nutrition and Exercise Metabolism, 2018, *in press*

以上

以下、参考資料

(投稿、審査中論文)

- 5) Ishihara K and Taniguchi H, Fat<sub>max</sub> as an index of aerobic exercise performance in mice during uphill running.

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## 講演詳細

分類	一般演題
年度	2016
講演日時	2016年5月15日 10:36-10:48
会場名	C
発表番号	3C-09a
セッション名	栄養生理・応用栄養：運動栄養・生理（2）
タイトル	摂取した糖質の最大利用可能量についてのマウスを用いた検討 Estimation of maximum oxidation of exogenously administered glucose in mice during treadmill running.
著者	○石原 健吾 <sup>1</sup> 、秋山 奈生 <sup>1</sup> 、畑 ひろみ <sup>1</sup> 、伏木 亨 <sup>1</sup> Oshihara Kengo <sup>1</sup> , Akiyama Nao <sup>1</sup> , Hata Hiromi <sup>1</sup> , Fushiki Tohru <sup>1</sup>
所属	<sup>1</sup> 龍谷大・農・食品栄養、
要旨	<p>【目的】 運動強度が増加するにつれて、糖質エネルギーへの依存度が高くなり、グリコーゲンの消耗が速くなる。持久運動トレーニングの主な目的は脂質酸化能力の向上であるが、補給した糖質の最大利用能力がトレーニング標的となりうるかは知られていない。そこで、運動中に補給した糖質溶液の利用速度を測定し、その上限を求めることを目的とした。</p> <p>【方法】 呼気ガス中の<sup>13</sup>C<sub>2</sub>O<sub>2</sub>排出量を高感度かつ速いレスポンスで検出できるようにするために、呼気ガス分析装置（アルコ2000、アルコシステム社）の測定条件検討を十分に行った。ICR雄性マウスを用い、安定同位体<sup>13</sup>Cで標識したグルコース溶液を経口投与して、呼気中への<sup>13</sup>C<sub>2</sub>O<sub>2</sub>排出量を経時的に測定した。グルコース溶液の濃度は10, 20, 30, 36%の4種、投与量は5 uL/g体重とし、全ての溶液が同一量（2mg）のU-<sup>13</sup>C-グルコースを含むように調製した。</p> <p>【結果】 U-<sup>13</sup>C-グルコースは10～12分で呼気への排出が開始され、26～30分でピークに達した。ピークでの<sup>13</sup>C<sub>2</sub>O<sub>2</sub>/<sup>13</sup>C<sub>2</sub>O<sub>2</sub>比は1.5～1.7であり、十分に検出範囲内であった。4種の糖質溶液の間で、<sup>13</sup>C<sub>2</sub>O<sub>2</sub>/<sup>13</sup>C<sub>2</sub>O<sub>2</sub>比に全く違いは認められなかった。</p> <p>【考察】 本実験条件の範囲では、糖質投与量に比例して糖質の酸化量が増加した。すなわち本グルコース濃度範囲では、糖質溶液の利用速度に上限は認められなかった。そこで低濃度および高濃度の糖質溶液についても、同様の試験を行ない糖質溶液の利用速度の上限について検討を行い発表する予定である。</p>
キーワード1	安定同位体
キーワード2	<sup>13</sup> Cグルコース
キーワード3	呼気ガス

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## 講演詳細

分類	一般演題
年度	2017
講演日時	2日目(5/20・土) 13:30~14:30
会場名	ポスター会場・エリアB
発表番号	2P-B39
セッション名	B：運動栄養・生理(2)
タイトル	滑り止め処理を施工したトレッドミルでのランニングエコノミー、最大酸素摂取量の測定 Anti-slip coating to treadmill for measurement of running economy and Vo2max
著者	○石原 健吾 <sup>1</sup> Oshihara Kengo <sup>1</sup>
所属	<sup>1</sup> 龍谷大 農・食品栄養、
要旨	マウスをトレッドミルで運動させる場合、走行速度を上昇させると酸素消費量は増大するものの、ヒトに比べるとその増加は小さく、今日でもマウスの最高酸素摂取量（PeakVo2）が真の最大酸素摂取量（Vo2max）に相当するの議論がある。著者は走行時の骨格筋への負荷を高めることによって、真のVo2maxに近い値が得られると考えた。実験には雄性ICRマウスを用い、半数はランニングソーサーによる7週間の自発走行トレーニングを行い、半数は安静群とした。呼気ガス測定は、トレッドミルの傾斜面の角度を0度から50度まで10度刻みの6条件で実施した。それぞれの傾斜でPeakVo2、ランニングエコノミーを評価し、トレーニング効果を検出できる運動負荷プロトコルがヒトの運動負荷条件を反映していると考えた。呼気ガス測定にはARCO2000（アルコシステム）を用い、トレッドミルの走行面には独自の滑り止め処理を施工した。滑り止め処理によって20度以上の傾斜での走行能力が向上し、40度の傾斜での疲労困憊までの走行時間は未処理の場合と比較して、14.9±8.6SD分から22.1±4.2SD分に増加した（P<0.05）。以下は滑り止め処理をしたトレッドミルでの結果である。傾斜に比例して、走行速度に対する酸素消費量の増分が大きくなった。PeakVo2も傾斜に比例して増加し、トレーニング群では0度の傾斜でのPeakVo2は平均125 mL/min/kgであったが、40度の傾斜で最も高い値（176 ±18SD mL/min/kg）に到達した。30および40度の傾斜で測定したPeakVo2は、トレーニング群と安静群の間で有意差が認められた。走行中の酸素消費量を走行速度で除して算出したランニングエコノミーは0度から40度までの傾斜でトレーニングによる向上が認められた（P<0.05）。本研究課題は、 <b>杉山産業化学研究所の助成金によって実施された。</b>
キーワード1	最大酸素摂取量
キーワード2	呼吸持久力
キーワード3	ランニングエコノミー

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## Abstracts From the December 2016 International Sports and Exercise Nutrition Conference in Newcastle upon Tyne

**Five days exposure to high dietary fat intake has no impact on the performance of a preloaded 5km treadmill based time trial in endurance-trained women.**

G Fletcher<sup>1</sup>, EI Glover<sup>2</sup>, JL Thompson<sup>1</sup>, GA Wallis<sup>1</sup>

<sup>1</sup> School of Sport, Exercise and Rehabilitation Sciences, University of Birmingham, UK

<sup>2</sup> GlaxoSmithKline, Consumer Healthcare, UK

Manipulating the dietary intake of carbohydrate and fat results in differences in the circulating hormonal and metabolic milieu alongside differences in the myocellular substrate storage profile. These divergent profiles can dramatically impact substrate utilisation during exercise, potentially impacting endurance performance. However, at least in men, improvements in endurance performance following a high fat diet are equivocal. Women compared to men, however, demonstrate a greater ability to oxidise fat and so may respond more favourably to high dietary fat intake. Previous studies employing a high fat diet are typically restricted in carbohydrate, and so a change in performance could be attributed to the manipulation of either macronutrient. The current investigation sought to determine if a high fat diet both with (HF) and without restricting carbohydrate intake (N+HF) impacts endurance performance in women compared to a control diet that reflects normal intake (N). Over three separate periods of 5 days, in a randomised counterbalanced order, women (means  $\pm$  SD: age  $34 \pm 8$  y;  $\text{VO}_2\text{max}$   $55.1 \pm 2.5$  ml/kg/min) were provided with 3 diets designed with the following macronutrient composition (% of energy intake [carbohydrate/fat/protein]); N (50/35/15); HF (20/65/15), and a hypercaloric (130% energy intake) N+HF (50/65/15). Post-diet intervention, in the overnight fasted state, subjects completed a 90min treadmill run at 65%  $\text{VO}_2\text{max}$  immediately followed by a self-paced 5km time trial (TT). Data was assessed for differences using a repeated-measures one-way ANOVA. There were no significant differences in the time taken to complete the TT (N -  $1328 \pm 83$ s; HF -  $1349 \pm 76$ s and NHF -  $1333 \pm 76$ s). Within the ranges of macronutrients provided, the total amount and proportion of dietary fat or carbohydrate consumed over five days had no impact on preloaded 5km treadmill running performance in endurance-trained women runners. This work was funded through a BBSRCi CASE studentship with GlaxoSmithKline the industrial partner.

**Unclear effect of omega-3 polyunsaturated fatty acid supplementation on speed-skating performance**

DA Noordhof<sup>1</sup>, SL van Ginkel<sup>1</sup>, K Levels<sup>1</sup>, S van der Zwaard<sup>1</sup>, MJ Hofmijster<sup>1,2</sup>, WJ van der Laarse<sup>3</sup>, CJ de Ruiter<sup>1</sup>, RT Jaspers<sup>1</sup>, and JJ de Koning<sup>1,4</sup>

<sup>1</sup> Department of Human Movement Sciences, Faculty of Behavioural and Movement Sciences, Vrije Universiteit Amsterdam, MOVE Research Institute Amsterdam, the Netherlands

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<sup>3</sup> Department of Physiology, VU University Medical Center Amsterdam, The Netherlands

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Elite speed skaters need to be able to generate a high peak power output and to sustain a high power output for the duration of the race; they therefore combine resistance-type training with endurance training. Combining these training modalities results in a smaller increment in muscle strength than when only resistance-type training is performed. An increase in, among others, myoglobin concentration can possibly reduce this interference effect. Omega-3 fatty acid supplementation and hypoxia may increase myoglobin expression. As, speed skating is by nature a sport in which the active leg muscles become easily hypoxic; the goal of this study was to investigate the effect of omega-3 fatty acids on speed-skating performance and performance-determining variables. Competitive speed skaters ( $n=27$ ) were, based on performance and, randomly assigned to the placebo (corn oil) or experimental group (2400 mg omega-3 fatty acids per day). Speed skaters performed a maximal incremental test, jump tests, a Wingate, and a 3000-m skating race before and after a ~7-weeks supplementation period. The magnitude-based inferences approach was used for the statistical analysis. The average change in 3000-m speed-skating performance time, due to the supplementation period, was  $-0.5 \pm 2.2\%$  in the placebo group and  $-0.7 \pm 2.3\%$  in the experimental group. The difference in change scores between groups was unclear ( $-0.2\%$ , 90% CL  $2.2\%$ ). A likely positive effect of the intervention was found on squat jump height (difference in change scores  $4.9\%$ , 90% CL  $4.2\%$ ), with likely and very likely negative effects on at

the ventilatory threshold (difference in change scores -6.9%, 90% CL 6.4%) and (difference in change scores -5.5%, 90% CL 3.4%), respectively. In conclusion, omega-3 fatty acid supplementation resulted in an unclear effect on speed-skating performance. The chances on a possibly negative effect, trivial effect, and positive effect on performance were 32%, 27%, and 41%, respectively. In addition, omega-3 supplementation resulted in negative effects on aerobic performance-determining variables and variable effects (unclear, trivial or likely positive) on anaerobic performance-determining variables. Therefore, speed skaters are advised not to consume omega-3 supplements in a dose of 2400 mg per day.

#### **Leucine availability regulates p70S6K activity when carbohydrate availability is sufficient in recovery from low carbohydrate training.**

SG Impey<sup>1</sup>, KM Hammond<sup>1</sup>, R Naughton<sup>1</sup>, C Langan-Evens<sup>1</sup>, SO Shepherd<sup>1</sup>, AP Sharples<sup>1</sup>, J Cegielski<sup>2</sup>, K Smith<sup>2</sup>, S Jeromson<sup>3</sup>, DL Hamilton<sup>3</sup>, JP Morton<sup>1</sup>

<sup>1</sup> Research Institute for Sport and Exercise Sciences, Liverpool John Moores University, Liverpool, UK

<sup>2</sup> MRC-ARC Centre for Musculoskeletal Aging, University of Nottingham, Derby, UK

<sup>3</sup> Health and Exercise Sciences Research Group, University of Stirling, Stirling, UK

We assessed the ability of protein with augmented leucine content to reactivate p70S6K following low CHO training. In a repeated measures cross-over design eight males completed 75 minutes of cycling exercise comprised of HIT and steady state intensity in a low carbohydrate but high protein state. Protein was provided before, during and after (20g per hour) in the form of collagen gel (GEL) or conventional whey protein (WHEY). Following exercise carbohydrate was provided in addition to protein so as to facilitate muscle glycogen synthesis. Muscle glycogen was comparable before (GEL: 354 ± 44; WHEY: 339 ± 66 mmol.kg dw) exercise and decreased post exercise (GEL: 141 ± 25; WHEY: 158 ± 80 mmol.kg dw) ( $P < 0.05$ ). Exercise induced comparable PGC-1 $\alpha$  (8-fold), ATG12 (1.3-fold) and Parkin (1.3-fold) increases in gene expression at 90 minutes' post-exercise. Suppression of p70S6K activity was comparable between conditions post-exercise (~ 25 fmol.min<sup>-1</sup>.mg<sup>-1</sup>) ( $P < 0.05$ ), while provision of carbohydrates and collagen protein was sufficient to re-activate p70S6K activity (GEL: 73 ± 42 fmol.min<sup>-1</sup>.mg<sup>-1</sup>), provision of leucine rich whey protein further augmented activity at 90 min post exercise (WHEY: 180 ± 105 fmol.min<sup>-1</sup>.mg<sup>-1</sup>) ( $P < 0.05$ ). This augmented response occurred independent of insulin (WHEY AUC: 5296 ± 1478; GEL AUC: 3925 ± 602 AU) ( $P > 0.05$ ) and PKB (WHEY: 47.7 ± 21.9; GEL: 40.4 ± 19.8 fmol.min<sup>-1</sup>.mg<sup>-1</sup>) ( $P > 0.05$ ). These data support the notion of a critical glycogen threshold for augmented oxidative adaptations in addition to identifying optimal nutritional recommendations to augment signalling to facilitate recovery processes.

#### **The effect of milk on recovery from repeated sprinting and jumping in female team-sport athletes**

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<sup>3</sup>London Sport Institute, School of Science and Technology, Middlesex University, UK.

Research has shown that consumption of milk post-eccentric exercise can attenuate the effects of exercise-induced muscle damage (EIMD) in team-sport athletes. However, these studies evoked damage using an intense eccentric protocol quite different to the exercise stress experienced by athletes during and after team sport training or competition. The aim of this study was to examine the effects of post-exercise milk consumption on recovery from activities that simulate those of team sport participation. Eighteen female team-sport athletes participated in an independent-groups investigation, completing 15x20m maximal sprints stopping within a 10m deceleration zone immediately after the sprint. Following a rest period, participants completed eight sets of 10 countermovement jumps, with one minute recovery between sets. On completion of the protocol participants consumed 500ml of milk or 500ml of an energy-matched carbohydrate drink. Muscle function (peak torque, rate of force development (RFD), 5, 10 and 20m sprint, countermovement jump (CMJ), reactive strength index (RSI)), muscle soreness and tiredness, perception of recovery, and serum creatine kinase (CK) and high-sensitivity C-reactive protein (hsCRP) were determined pre- and 24h, 48h and 72h post-exercise. Results were analysed using magnitude based inferences.

Results indicated that consumption of milk had a likely beneficial effect on peak torque extension at 60°/s from baseline (B) to 24h (6.0 ± 7.7, Mean effect ± 90% CI), flexion at 60°/s from B- 24h (8.7 ± 13.5) and B-72h (9.4 ± 12.3). A likely benefit was seen for extension at 180°/s B-24h (7.2 ± 6.4), B-48 (10.8 ± 9.6) and a very likely benefit B-72h (9.9 ± 6.8). A likely benefit of milk was seen for flexion at 180°/s B-24h (12.4 ± 13.8) and B-72h (10.2 ± 12.1). Milk had a possible/likely benefit in limiting losses in 5m sprint time (2.7 ± 4.0) and RFD (22.7 ± 31.7) from B-24h, a very likely beneficial effect on perception of recovery (2.0 ± 2.4) from B- 24h and a likely beneficial effect on muscle soreness (1.0 ± 1.3) from B- 72h. Comparison of the effects of milk and carbohydrate on 10m and 20m sprint time, RSI, muscle tiredness, CK and hsCRP were unclear. In conclusion, consumption of milk can limit decrements in muscle function, soreness and perception of recovery following repeated sprinting and jumping exercise.

#### **Changes in dietary intake, immune function and performance monitors throughout a season in professional rugby league players**

E Tester<sup>1</sup>, B Jones<sup>1</sup>, L Duckworth<sup>1</sup>, T Ispoglou<sup>1</sup>

<sup>1</sup> Centre for Sports Performance, Institute for Sport, Physical Activity and Leisure, Leeds Beckett University, UK

Throughout a rugby league (RL) season players are exposed to a high volume of competitive fixtures. Cumulative loads and inadequate nutrition may suppress immune function, which would have negative consequences for health and performance. As no study has monitored immune function and dietary intake throughout a RL season, the aim of the study was to identify the pattern of dietary intake and critical time points where immune function and selected performance monitors are compromised. Following ethics approval, 20 male volunteer professional RL players (25.4±3.2 y, 98.2±8.4 kg) were monitored the day prior to a competitive fixture for 25 weeks of the season. Weekly changes in neuromuscular function (CMJ), wellbeing (5-point questionnaire), training loads (sRPE), salivary testosterone (sTest) and immunoglobulin A (sIgA) were recorded. Dietary intake (4-day food diary) was assessed at the start, middle and end of the season. Changes from the overall mean were inferred via Cohen's *d* effect sizes using means and standard deviations which were calculated from a linear mixed model to account for missing data. Moderate increases in training load occurred in weeks 3, 5, 9, 10, 12 and 20 and a very large increase occurred in week 21. Moderate decreases in CMJ flight time occurred in week 14 and small decreases occurred in weeks 6, 7, 20, 22, 24, 25. For all wellbeing parameters small decreases occurred in weeks 7 and 17 with small increases in stress, soreness and fatigue also occurring in week 20. Mean sTest was 119.8±55.5 pg·ml<sup>-1</sup> with small declines occurring in week 7, 10, 13, 20 and 22. Overall sIgA concentrations ranged from 1.03-1.18 µg·min<sup>-1</sup>, but compared to the overall mean (1.12 ± 0.17 µg·min<sup>-1</sup>) small decreases were observed in weeks 6, 9, 16, 21, 23, 24. Dietary intakes were consistent during the recording periods with mean energy intake ranging from 2811-3149 kcal·day<sup>-1</sup>. Carbohydrate, protein, and fat intakes ranged from 2.9-3.2 g·kg·BM<sup>-1</sup>, 2.0-2.1 g·kg·BM<sup>-1</sup>, and 1.1-1.4 g·kg·BM<sup>-1</sup> respectively. The findings suggest that a consistent intake throughout a competitive season did not prevent reductions in immune function and selected performance monitors. Further research to develop appropriate periodised nutritional strategies to maintain immune function and support player health and performance is needed.

#### Heart rate variability and common health measures

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Recent reports in small populations suggest a relationship between body mass index (BMI) and heart rate variability (HRV). The aim of the present study was to investigate this relationship, together with other common indices of health, in a

large population of healthy men and women. Between 2006 and 2015 four hundred and ninety-three volunteers (Men 218; 29±11 y, 1.83±0.11 m, 77.6±11.3 kg; Women 275; 31±12 y, 1.66±0.07 m, 64.4±11.1 kg) had heart rate variability measured (R-R intervals, ms) (RS 800, Polar, Kempele, Finland), during 10-15 minutes of quiet supine rest, health measures included BMI (kg/m<sup>2</sup>), %body fat (BC418 Segmental Body Composition Analyzer), systolic (SBP) and diastolic (DBP) blood pressure (mmHg), aerobic fitness (VO<sub>2</sub>max) and physical activity (Baecke Questionnaire). Simple correlational analysis revealed measures of heart rate variability, particularly vagal function were negatively associated with BMI ( $r=-0.164$ ), SBP ( $r=-0.195$ ), DPB ( $r=-0.456$ ), resting heart rate ( $r=-0.577$ ) ( $P<0.01$ ) body fatness ( $r=-0.195$ ,  $P<0.05$ ) and positively associated with Baecke sport index ( $r=0.130$ ), aerobic fitness ( $r=0.145$ ) ( $P<0.05$ ) and the fat free mass ( $r=0.192$ ,  $P<0.01$ ). Stepwise regression analysis indicated that in this population of normal healthy men and women, age and sex acted as confounding variables on selected health measures, except for BMI and body fatness with HRV ( $P=0.054$  and  $P=0.058$  respectively) The present study illustrates the importance of non-invasive HRV analysis to complement traditional health measures.

#### The effect of tart cherry (prunus cerasus) on sleep and fat oxidation during steady state exercise

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Melatonin influences human circadian rhythms and mediates the sleep-wake cycle. Accumulating evidence suggests a relationship between sleep duration/quality and fat oxidation (FO). Montmorency tart cherries (MTC) are reported to contain high amounts of naturally occurring melatonin, but the effects of MTC supplementation on sleep duration/quality and on subsequent exercise are unknown. Therefore, the aim of this study was twofold: 1) to determine the effects of 1-week MTC supplementation on sleep duration/quality and; 2) to investigate subsequent FO and physiological parameters during a single bout of exercise. Following a randomised, double blind, placebo controlled, cross over design, ten males (age: 27±7 years; body mass index: 24.2±4.5 kg/m<sup>2</sup>; O<sub>2max</sub>: 43.8±5.6 ml·kg<sup>-1</sup>·min<sup>-1</sup>) underwent two experimental trials separated by 14 days to ensure washout. Under both conditions, participants ingested a beverage twice daily for 7 days, either with (MTC) or without (PLA) the addition of 30ml MTC concentrate. Under both conditions, participants completed 30-minutes of standardised steady state cycling at baseline (prior to experimentation) and post-supplementation; workload was set to elicit individualised predetermined maximal FO rates to account for inter-individual

variation in FO. Sleep wake cycles were continuously assessed during each 7-day supplementation phase using wrist actigraphy and FO determined via indirect calorimetry. Sleep onset latency was reduced under MTC, but not under PLA (MTC  $4.3 \pm 0.6$  vs. PLA  $10.0 \pm 1.9$  minutes;  $P = 0.01$ ). Correspondingly, total sleep time under MTC was greater than PLA (MTC  $430 \pm 14$  vs. PLA  $398 \pm 13$  min;  $P = 0.01$ ). Under MTC the number of awakenings was less (MTC  $18 \pm 2$  vs. PLA  $23 \pm 3$ ;  $P < 0.01$ ), and sleep efficiency increased (MTC  $89 \pm 1$  vs. PLA  $84 \pm 1$  %;  $P < 0.01$ ). No conditional differences or time effects were observed in FO ( $P > 0.05$ ), blood lactate ( $P > 0.05$ ), core temperature ( $P > 0.05$ ), cutaneous vascular conductance ( $P > 0.05$ ), or rating of perceived exertion ( $P > 0.05$ ) during steady state exercise. In conclusion, 7-day TC supplementation improved sleep duration/quality but failed to influence subsequent FO or physiological parameters during a single bout of exercise.

#### Effect of caffeinated gum on a battery of soccer-specific tests in trained university-standard male players

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The purpose of this study was to determine whether caffeinated gum would improve performance in a battery of soccer-specific tests typically used by teams to assess performance and fitness. In a double-blind, randomised design, ten trained university-standard soccer players (age  $19 \pm 1$  years; body mass  $75.5 \pm 4.8$  kg; stature  $1.80 \pm 0.10$  m) chewed a caffeinated gum (Military Energy Gum, Chicago, IL) that contained 200 mg of caffeine and a placebo gum with the same appearance on two separate occasions separated by 7 days. After a standardised warm up, the gum was chewed for 5 minutes before participants performed a 20 m sprint, a maximal countermovement jump test and the Yo-Yo intermittent recovery test level 1 (Yo-Yo IR1). Performance on 20 m sprint times was not different (caffeine =  $3.2 \pm 0.3$  vs placebo =  $3.1 \pm 0.3$  s;  $P = 0.567$ ), but the caffeinated gum did allow players to cover a greater distance on the Yo-Yo IR1 (caffeine =  $1754 \pm 156$  vs placebo =  $1719 \pm 139$  m;  $P = 0.016$ ) and increased maximal countermovement jump height (caffeine =  $47.1 \pm 3.4$  vs placebo =  $46.1 \pm 3.2$  cm;  $P = 0.008$ ). Chewing caffeine gum for 5 minutes immediately before performing a battery of soccer specific tests enhanced performance by 2.0% on the Yo-Yo IR1 and by 2.2% on the maximal countermovement jump test but had no effect on 20 m sprint times in trained university-standard soccer players.

#### Sodium bicarbonate ingestion improves cycling performance, irrespective of single or split dose ingestion

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Split dose sodium bicarbonate (SB) supplementation protocols may minimise the gastrointestinal discomfort associated with single dose ingestion. In consideration of athletic competition protocols, the most practical time for a second SB dose is following the warm-up. This study examined the effect of single vs split dose SB ingestion on blood bicarbonate and cycling performance following a high-intensity warm-up. Twelve physically active males (age  $21 \pm 2$  y; body mass,  $79 \pm 4$  kg) completed four experimental sessions. Individuals consumed either a single ( $0.3 \text{ g} \cdot \text{kg}^{-1}$  body mass (BM)) or split ( $0.2 \text{ g} \cdot \text{kg}^{-1}$  BM) dose of placebo (PLA) or SB 3 h prior to a high-intensity warm-up. During the split dose condition an additional  $0.1 \text{ g} \cdot \text{kg}^{-1}$  BM was provided immediately following the warm-up. Participants then rested for 30 min before performing a cycling capacity test at  $110\% W_{\max}$  (CCT<sub>110%</sub>), until volitional exhaustion. The Institutional Ethics Review Committee approved the study. Fingertip capillary blood samples were taken throughout the sessions, with total work done (TWD) recorded as the outcome measure for the CCT<sub>110%</sub>. SB ingestion raised blood bicarbonate levels from baseline ( $P < 0.001$ ) with an additional  $2 \text{ mmol} \cdot \text{L}^{-1}$  increase following the single dose compared to the split dose ( $P < 0.001$ ). After the HI warm-up, blood bicarbonate levels were greater than PLA only during the single dose SB condition ( $P = 0.003$ ). The decline in blood bicarbonate following the CCT<sub>110%</sub> (PLA:  $7.1 \text{ mmol} \cdot \text{L}^{-1}$ ; Split:  $7.4 \text{ mmol} \cdot \text{L}^{-1}$ ; Single:  $9.4 \text{ mmol} \cdot \text{L}^{-1}$ ) was influenced by SB ingestion, but not dosing strategy. SB ingestion improved TWD by  $\sim 8 \text{ kJ}$  compared to placebo following both dosing strategies (Single: +17%; Split +23%;  $P < 0.001$ ). These data suggest that high-intensity cycling performance is increased following SB supplementation, irrespective of single or split dose ingestion protocols, despite greater increases in blood bicarbonate with the single dose.

#### Warm-up intensity does not influence the beneficial effect of sodium bicarbonate ingestion on cycling performance

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Increasing blood bicarbonate concentration via sodium bicarbonate (SB) ingestion is associated with improved high-intensity (HI) exercise performance. SB ingestion is commonplace with competitive athletes to aid HI performance, yet these individuals also complete HI warm-up (WU) protocols prior

to exercise. The impact of WU intensity on blood bicarbonate concentrations and performance following SB ingestion remains unknown. The study examined the effect of WU intensity on blood bicarbonate and cycling performance following ingestion of 0.3 g·kg<sup>-1</sup> body mass placebo (PLA) or SB. Twelve physically active males (age, 21±2 y; body mass, 79±4 kg) completed four experimental trials. Individuals consumed either PLA or SB 3 h prior to a HI or low-intensity (LI) WU. Participants rested for 30 min before performing a cycling capacity test at 110% W<sub>max</sub> (CCT<sub>110%</sub>), until volitional exhaustion. The Institutional Ethics Review Committee approved the study. Fingertip blood samples were taken throughout the sessions, with total work done (TWD; in kJ) recorded as the outcome measure for the CCT<sub>110%</sub>. SB consumption raised blood bicarbonate levels from baseline (LI: +6.8±1.6 mmol·L<sup>-1</sup>; HI: +6.0±1.6 mmol·L<sup>-1</sup>; P<0.001) with no change following PLA. Completion of the WU reduced blood bicarbonate in all conditions (P<0.001), with greater declines evident following the HI WU (LI: -8.5±3.0 mmol·L<sup>-1</sup>; HI: -12.7±3.9 mmol·L<sup>-1</sup>; P=0.005). SB ingestion also resulted in greater declines in blood bicarbonate following WU when compared to PLA (P=0.02). During the 30 min rest period the blood bicarbonate recovered, dependent upon WU intensity and SB ingestion. TWD was unaffected by WU intensity, yet was improved following SB ingestion (~7kJ; P<0.001) during both the LI (+22%) and HI (+17%) conditions. These data suggest that WU intensity does not influence the subsequent benefit of SB ingestion to cycling performance.

#### The additive ergogenic effect of β-Alanine and sodium bicarbonate supplementation on repeated bouts of high intensity cycle ergometry in male collegiate athletes

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β-Alanine (β A) and sodium bicarbonate (SB) ingestion may provide performance enhancing benefits by enhancing concentrations of their respective physiochemical buffers, counteracting acidosis during intense exercise. Whilst there is extensive literature on the individual performance enhancing effects of these supplements there is little research on their combined effects. With institutional ethical approval, twelve male collegiate athletes (mean±SD: age 20±1y; body mass 94.5±16.8 kg; height 1.81±0.81 m) volunteered to take part in the study. Subjects performed 4 x 30 s high intensity sprints at 5% body mass on a cycle ergometer interspersed with a 180s passive recovery period, before and after a 28 day supplementation period. A computerised software package linked to the cycle ergometer was used to record peak power output (PPO) and mean power output (MPO) during each sprint. Blood lactate (BL) and rate of perceived exertion (RPE) were measured at the end of each sprint. Using a double-blind, randomized, matched paired experimental design, subjects were allocated to either a glucose (placebo- PL)/ SB or β A/ SB supplementation group. The β A/ SB group consumed 6.4g·day<sup>-1</sup> of β A (split between four servings) for 28 days and 0.3g·kg<sup>-1</sup> body mass of SB 90 minutes prior to post supplementation testing. The PL/ SB group followed the same protocol but consumed 6.4g

day<sup>-1</sup> of PL for 28 days. Delta changes for pre and post supplementation were calculated for PPO, MPO, BL and RPE. Data were then analyzed using a two way mixed design analysis of variance (ANOVA). There was no difference (p> 0.05) in PPO between the groups for sprint one (1134±58 vs 1104±40 W), two (1039±54 vs 1008±45 W), three (1020±53 vs 1017±40W) or four (998±42 vs 1005±49W). There was also no difference (p> 0.05) in MPO between the groups for sprint one (603±38 vs 609±40W), two (607±36 vs 600±37W), three (617±35 vs 604±42W) or four (596±36 vs 582±45W). β A/ SB supplementation led to increases in BL and RPE but this was not different from the PL/ SB group. In conclusion the combination of β A and SB did not have an additive effect on repeated bouts of high intensity cycle ergometry in male collegiate athletes.

#### Do female dance students meet the recommended dietary intakes for athletes?

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Dancers undergo huge pressure from the industry to meet a specific body size and shape, which can lead to eating disorders and poor nutritional health. Numerous studies have identified a higher prevalence of low body image satisfaction and negative eating attitudes in dancers than in non-dancers. Often accompanied with these issues is a failure to meet nutritional requirements, which can negatively impact on performance. Previous studies have focused on dancers, but research into university dance students is limited and there is a lack of investigation into the nutritional status of dancers. The aim of this study was to investigate whether female dance students meet the nutritional recommendations for athletes. After gaining ethical approval, female dance students (n=15) were recruited from Liverpool Hope University. Participants completed a three-day food diary, Eating Attitudes Test-26 (EAT-26) questionnaire and were measured for height, weight and body fat percentage. The dietary intake data was analysed using Microdiet software (v3) and statistical analysis was conducted using SPSS Statistics (v22). The statistical significance level was set at 0.05. The results showed that the dance students' carbohydrate intake was 3.1±1.2 (mean ± SD) g/kg BW/day, which is lower than the ACSM recommendation of 5-7g/kg BW/day according to their activity level of ≥5 hours of dance per week. The guideline for protein intake for athletes is 1.2-2.0g/kg BW/day, but some dancers were consuming less than this, with a mean intake of 1.1±0.4 g/kg BW/day. The dancers also presented very low energy intake of 1608±4 kcal/day. In addition, 5 of the 15 of the dance students scored above the cut-off in the EAT-26 for being at risk of an eating disorder. The failure to meet the nutritional guidelines for athletes may be due to the pressure the dancers feel to adhere to a specific body image. This is likely to have detrimental effects as insufficient energy, carbohydrate and protein intake can compromise performance, lead to loss of muscle mass, cause menstrual dysfunction, decrease bone density and increase fatigue and injury. This study emphasises the need to detect and intervene with disordered eating patterns and low nutritional intakes that are present in athletes in University settings.



### Sweating rate and dehydration levels in professional soccer players after an official league match (Spanish Second Division "B")

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One of the main reasons of performance loss is the dehydration caused by inadequate liquid intake during physical exercise. When 2% total body weight is lost due to the fluid loss, this compromises many physiological and cognitive functions, resulting in a decrease in performance. In soccer, to determine hydration strategies it is very important to maintain the proper water balance and avoid a performance fall. Our aim was to describe the sweating rate and the dehydration levels in professional soccer players after an official league match of the Spanish Second Division "B". Thirteen professional soccer players (20±2 years old, body weight 72±8 kg, height 175±5 cm) were analyzed before and after the match. The goalkeeper was excluded. The temperature during the match was 31±1°C and relative humidity 61±4%. To assess weight loss, players were weighed nude before and after the match. The fluid intake calculation was determined by the difference in volume of the bottles from the start to the final of the game (2 bottles per player, one with 500 ml of mineral water, and another with 500 ml of a liquid with a 20% carbohydrate and 0.6% sodium). The measurement was done with a measuring cylinder. No player urinated during the game. Sweating rate was calculated as: Sweating rate = (weight loss + liquid intake - urine excreted) / minutes of activity. We registered an activity time of 85±33 minutes. Weight loss was 929±473 g (range 0 - 1800 g), equal 1.3±0.6% (range 0.0-2.3%) of body mass. The liquid intake was 678±395 ml (range 290 - 1620 ml), and the sweating rate was 20±10 ml/min<sup>-1</sup> (range 0-37 ml/min<sup>-1</sup>). The liquid intake from the studied sample was not enough for remedying the liquid loss caused by the dehydration. Therefore, it would be necessary to incorporate new hydration strategies that aim to achieve and maintain a proper water balance.

### FatMax test during hilly running exercise in mice

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Fat oxidation is of interest to endurance athletes and scientists. FatMax is the exercise intensity that elicits maximal fat oxidation. However, measurement of FatMax is rarely conducted in small rodent research, because fat oxidation of trained mice keeps increasing and does not clearly show a decrease phase during running. The present study investigated the effect of incline of the treadmill on FatMax and maximum oxygen consumption in trained and untrained mice. Male

ICR mice were divided into two groups. Each group of mice was housed in standard cages with or without running wheel. Mice ran to their maximum speed (5 to 40 m/min) on motor driven treadmill after 7 weeks of spontaneous running training period. Oxygen consumption (VO<sub>2</sub>) and carbon dioxide production (VCO<sub>2</sub>) were measured during running using mass spectrometer for respiratory analysis (ARCO-2000, Japan). Maximum running test was repeated six times and incline of the slope was differed (0, 10, 20, 30, 40, and 50) in each running test. Peak oxygen consumption (VO<sub>2peak</sub>) was correlated the incline of slope (P < 0.05). VO<sub>2peak</sub> was different among training and sedentary group at the incline of 30 and 40°. Peak of fat oxidation was observed at the incline of 20, 30 and 40°, and exercise intensity of maximum fat oxidation (FatMax) was higher in training group than sedentary group (P < 0.05). Serum lactic acid level was lower in training group than sedentary group at the exercise intensity of FatMax (P < 0.05). Repeated measurement of VO<sub>2peak</sub> at the incline of 40° showed a correlation (P < 0.05). In conclusion, measurement of FatMax and VO<sub>2peak</sub> should be conducted at an incline of 30 or 40° in mice. This work was supported by a grant from Sugiyama Sangyo Kagaku Institute.

### A systematic review and meta-analysis of randomized clinical trials of the herbal extract EPs 7630 in acute respiratory tract infections with relevance to adult performance athletes

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Athletes are at higher risk of contracting upper respiratory tract infections (URTI) during periods of intense training and following competition. A recent placebo-controlled study showed that prior 28 day treatment with the licensed herbal medicine EPs 7630 (*Pelargonium sidoides* extract) increased production of secretory immunoglobulin A in saliva, reduced interleukin-6 in serum, and likewise interleukin-15 in the serum and nasal mucosa in marathon runners subjected to an intense running session. This suggests EPs 7630 can exert a modulating influence on the immune response in the upper airway mucosa in athletes subjected to intense physical activity with the authors suggesting it is potentially able to prevent upper respiratory illness in athletes. To consider these observations in a clinical context, a review of double-blind, randomized, placebo-controlled trials of the efficacy and safety of *Pelargonium sidoides* preparation EPs 7630 in adult patients with acute bronchitis (AB) or acute rhinosinusitis (ARS) was completed. 10 trials met selection criteria, and 8 were identified as suitable for initial review, with a total of 3,392 participants (AB: 6/8 trials; ARS: 2/2 trials) In ARS, all trials included adults only whereas in AB, patient groups also included children and adolescents, both of which were excluded for the purpose of this analysis. 3 trials of 809 adults experiencing AB were subsequently analysed and 2 trials of

375 participants with ARS. In the meta-analyses of efficacy, EPs 7630 was superior to placebo in reducing both symptom severity and time until complete recovery for all indications investigated. Significant advantages for EPs 7630 were also observed for time until the onset of a meaningful treatment effect, global therapy outcome, and days of incapacity. No serious adverse drug reactions were reported. EPs 7630 is efficacious, safe, and well-tolerated in the symptomatic management of acute respiratory tract infections such as AB, ARS, in adults. It is well suited for use in performance athletes having been screened in UKAS testing Laboratory 1187 and found to contain no prohibited contaminants.

#### Habitual meal frequency, body composition and blood lipid profile in non-competitive bodybuilders

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The ultimate aim of bodybuilding is to achieve an aesthetically pleasing physique through gains in lean body mass (LBM) and reductions in fat mass (FM). Favourable blood lipid profile (BLP) adaptations have been reported but research is equivocal. Total energy intake (EI) has been suggested to be one of the biggest dietary predictors for optimum body composition with daily distribution of meals less important. However, high quality protein per meal as a means to maintain muscle protein synthesis suggests that higher daily meal frequency (MF) may be a more appropriate dietary strategy. Our aim was to investigate the interplay between habitual MF, body composition and BLP in non-competitive bodybuilders. Following ethics approval, 44 males and 10 females met participation criteria. Upper and lower 25<sup>th</sup> percentiles of response to number of eating occasions were calculated. Arranged into a low (LFG, 2.6±0.8: n=12, 28±5 years, 80.9±17.8 kg) or high (HFG, 6.6±0.8: n=12, 27±7 years, 85.2±16.8 kg) daily MF group, participants completed a 3-day diet diary, had a dual energy X-ray absorptiometry scan, and blood lipids measured. The HFG (13.9±3.8%) had lower (P=0.024) %body fat than the LFG (19.2±6.7%) but LBM in the HFG (70.2±14.4 kg) was not different from that of the LFG (62.1±14.5 kg). Blood lipids were within healthy range, while the HFG completed more (P=0.000) weekly training sessions (4.3±0.8) than the LFG (5.5±0.7). There was no difference in energy intake between the groups: HFG 2564±681 kcal; LFG 2215±533. Protein intake in the HFG was higher (P=0.054) than the LFG (2.6±1.0 vs 1.9±0.5 g/kg<sup>-1</sup>/BW/d<sup>-1</sup>). Differences were not observed in fat (1.2±0.6 and 1.4±0.6 g/kg<sup>-1</sup>/BW/d<sup>-1</sup>) or carbohydrate (2.5±1.4 and 1.9±1.1 g/kg<sup>-1</sup>/BW/d<sup>-1</sup> in LFG and HFG respectively) intakes. In relative terms, the carbohydrate intake in the HFG (25±9%) was lower (P=0.027) than that of the LFG (35±12%). In conclusion, BLP was within the healthy range in both groups. Furthermore, higher MF was associated with better sport-specific body composition outcomes. This is potentially

due to higher consumption of dietary proteins (35% of daily EI) resulting in optimisation of muscle synthetic response and training capacity.

#### Free-living 24-hour dietary intake and glycaemic control following experimental high versus low fat feeding in people with type 1 diabetes

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This study assessed the influence of high versus low fat feeding on subsequent dietary intake, insulin administration, and glycaemic control in ten men with type 1 diabetes (HbA<sub>1c</sub>: 7.0 ± 0.5%). Subjects completed two experimental morning-time trials (~07:30AM) in a randomised and counter-balanced fashion. On both occasions participants consumed a meal matched for carbohydrate and protein content, but containing either low fat (5g fat: LF), or high fat (55g fat: HF). Insulin dose was standardised and calculated according to the carbohydrate content of the meal. Continuous glucose monitoring captured interstitial glucose for 6-hours under laboratory conditions and a further 18-hours under free-living conditions. Free-living self-selected dietary intake and insulin administration was captured using weighed food diaries. Following meal ingestion, interstitial glucose peaked similarly at 60-mins under both conditions (P≤0.05). Beyond this time, concentrations under LF returned to fasted, resting levels, whereas concentrations remained elevated under HF throughout the remaining 5-hour laboratory observation period. Having left the laboratory and under free-living conditions, interstitial glucose under HF was on average higher than LF across the entire 18-hour period (HF 9.0±2.6 vs LF 6.4±1.0 mmol.l<sup>-1</sup>; P≤0.05); compared to LF, participants under HF spent a greater percentage of this time in hyperglycaemic ranges (HF 71% vs LF 5%; P≤0.05) and less time in euglycaemia (HF 28% vs LF 93%; P≤0.05) despite similar incidence of hypoglycaemia (P≥0.05). Self-selected dietary intake was similar between conditions with similar energy (HF 1974±584 vs LF 2028±657 Kcal; P≥0.05), macronutrient composition (Carbohydrate: HF 45% vs LF 43%, Fat: HF 40% vs LF 42%; Protein: HF 15% vs LF 15%; P≥0.05) and bolus insulin (HF 9.8±1.6 vs LF 9.7±1.5 IU; P≥0.05). Insulin administered as a corrective bolus was greater under HF (HF 5.2±0.7 vs LF 1.3±0.6 IU; P≤0.05). In conclusion, meals with a high-fat content predispose people with type 1 diabetes to sustained hyperglycaemia for up to 24-hours following feeding which may require additional insulin units to correct glycaemia. Subsequent dietary intake under free-living conditions does not seem to be influenced by acute high-fat feeding.

### Adding Fish Oil to Whey Protein, Carbohydrate and Vitamin D Improves Eccentric-Exercise Recovery in Soccer Players

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Soccer players often experience eccentric exercise-induced muscle damage due to the physical demands of soccer match-play. An improved muscle functional recovery from eccentric exercise has been shown with protein feeding. Since fish oil derived n-3 polyunsaturated fatty acids (n-3PUFA) exhibit anti-inflammatory properties, combining fish oil with protein may facilitate recovery. Therefore, the primary aim of the present study was to assess the influence of 6 wk supplementation with a multi-ingredient beverage containing a combination of n-3PUFA, whey protein and vitamin D on muscle function and soreness, inflammation and soccer performance during exercise recovery. Competitive soccer players (n=30) were assigned to one of three conditions. The experimental condition (FO) contained n-3PUFA (550 mg DHA, 550 mg EPA), whey protein (15 g) and vitamin D (3 µg). The protein control (PRO) contained whey protein (15 g) and vitamin D (3.9 µg) and the eucaloric control condition (CHO) contained carbohydrate. Eccentric exercise consisted of 12 sets of unilateral knee extension/flexion contractions on both legs separately. Eccentric exercise impaired maximal force and increased ratings of muscle soreness ( $p < 0.05$ ). Post supplementation, blood %n-3PUFA/total PUFA was greater in FO ( $36 \pm 6$ ) vs. PRO ( $24 \pm 3$ ,  $p < 0.05$ ) and CHO ( $23 \pm 2$ ,  $p < 0.05$ ). Maximum force loss during recovery tended ( $p = 0.10$ ) to be less in FO ( $-19 \pm 22\%$ ) vs. CHO ( $-30 \pm 10\%$ ). Muscle soreness (dominant leg), expressed as AUC during 72 h exercise recovery, tended to be less in FO ( $791 \pm 549$  mm × 72 h) vs. PRO ( $1403 \pm 779$  mm × 72 h,  $p = 0.08$ ) and CHO ( $1298 \pm 621$  mm × 72 h  $p = 0.10$ ). The blood concentration of c-reactive protein tended ( $p = 0.09$ ) to be lower in FO (AUC:  $42 \pm 11$  mg/L × 72 h) vs. CHO (AUC:  $83 \pm 42$  mg/L × 72 h,  $p < 0.05$ ). FO elicited a possibly (82-85%) beneficial effect on counter movement jump performance vs. PRO and CHO. In summary, a juice-based multi-ingredient beverage, containing n-3PUFA, whey protein and vitamin D, appears to mediate a reduced feeling of muscle soreness after eccentric exercise and translates into improved muscle function and soccer-specific performance during acute recovery. Hence, the benefit of whey protein ingestion to exercise recovery in competitive soccer players is enhanced by adding n-3PUFA and vitamin D.

#### Case study: the use of smartphone application technology to improve dietary practices, nutrition knowledge and body composition of a female international rugby union player

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Nutrition application (app) technology may offer a cost-effective and novel method of maintaining better communication between athletes and practitioners at times of the season when such interactions may be limited (i.e. periods separating live-in training camps). In contrast to clinical populations, limited research has profiled the use of nutrition app technology within athletic cohorts. With indices of nutrition knowledge, body composition and dietary intake of primary interest, this case study reports the effects of implementing nutrition app usage with a female international rugby union player (19 years old, 6 international U20s caps). Anthropometric data, a general nutrition knowledge questionnaire (GKNQ) and a 3-day food diary were collected before and after an 8-week in-season period that separated two consecutive training camps. Normal communication methods between camps were complemented by the use of app technology (MealLogger, New York) which allowed photo and text commentary interactions between the athlete and performance nutritionist. Daily energy intake increased ( $2029$  kCal.d<sup>-1</sup> to  $2213$  kCal.d<sup>-1</sup>), GKNQ scores improved ( $68$  to  $76$ ;  $+11\%$ ) and body composition benefitted (8-site skinfold sum  $133$ mm to  $112$ mm) from the 8-week period that required the upload, and subsequent commentary on, 161 meal photographs (~2.8 meals a day). By means of comparison, such changes were not observed in a comparable control player who completed the 8 week programme but did not receive the additional support via nutrition app technology. In scenarios where athlete-practitioner communication may be limited, the use of a nutrition app may be of benefit to the performance nutritionist to increase nutrition knowledge and body composition in team sport players.

Keywords: Rugby Union, Female, Smartphone App, Body Composition

#### Association between muscle strength and immune system biomarkers in older adults

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Aging is associated with immune system dysfunction which may increase the risk of infections, autoimmune diseases, and cardiovascular or neurodegenerative disorders. Little is known about the association between muscle strength and immune system biomarkers in older-aged individuals. In 153 individuals (96 females, 57 males), aged 65-100 years, concentrations of neopterin, tumor necrosis factor (TNF)-α, serum soluble tumor necrosis factor receptor type2 (sTNF-R75), interleukin

6, C-reactive protein (CRP), kynurenine to tryptophan ratio (Kyn/Trp), and muscle strength using hand-grip dynamometry were determined. The cutoff for considering low grip strength, adjusted for gender and body mass index, was established based on the Fried definition of frailty (lowest 20%). One-year mortality risk was estimated using the Charlson's comorbidity index. Individuals with low grip strength (n=40; 16% females, 44% males) showed higher levels of TNF- $\alpha$  (p<0.05) and sTNF-R75 (p<0.001) concentrations compared to individuals with normal grip strength. In addition, grip strength correlated with sTNF-R75 (p<0.01), neopterin (p<0.05), and CRP (p<0.05) concentrations, whereas Kyn/Trp was only weakly associated. After adjustment for age, only the association between sTNF-R75 and grip strength remained significant. One year mortality risk was significantly associated with biomarkers of immune activation (all p<0.001): sTNF-R75 (r=0.417), neopterin (r=0.278), Kyn/Trp (r=0.277), and kynurenine (r=0.230). Increasing concentrations of immune system biomarkers with older age indicate that the process of aging in healthy individuals is associated with immune activation especially of the T-cell/macrophage system. The immunobiological alterations correlated with muscle strength of individuals. Randomized intervention studies aiming to increase muscular strength in older adults, have to show whether the observed associations are causal and whether immunosenescence can be prevented by increased muscular strength.

#### Body water assessment using bioimpedance spectroscopy during rapid weight loss and recovery in Japanese wrestlers

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Athletes in weight-category sports conduct rapid weight loss (RWL) followed by weight regain (WR) before competition. Body water assessment during RWL and WR is important for monitoring changes in body composition and for predicting excessive dehydration. Bioimpedance spectroscopy (BIS) is an easy tool for assessing body water status. In contrast to the single-frequency impedance technique, BIS can assess extra- and intra-cellular water (ECW and ICW) content, and total body water (TBW) content separately. However, assessment of body water change, using BIS during RWL and WR, has not been well validated. This study aimed to determine the validity of BIS method, as compared to the stable isotope dilution technique, for measuring changes in TBW in Japanese wrestlers. Ten male Japanese collegiate wrestlers were asked to conduct RWL (i.e., loss of 6% of body mass in 53 hours) by a self-selected method, followed by WR in 13 hours, with

a prescribed diet (energy 2886 $\pm$ 310 kcal, protein 64 $\pm$ 9 g, fat 60 $\pm$ 10 g, and carbohydrate 522 $\pm$ 47 g). Body composition using an air plethysmography displacement system, TBW using a stable isotope dilution technique (TBW<sub>DIW</sub>), and ECW and ICW using BIS were determined at baseline, post-RWL, and post-WR. TBW assessed by BIS (TBW<sub>BIS</sub>) was calculated as the sum of ECW and ICW. Body mass, fat mass, and fat-free mass decreased post-RWL (73.68 $\pm$ 7.99 to 68.96 $\pm$ 7.65 kg, 8.7 $\pm$ 2.2 to 7.2 $\pm$ 2.1 kg, and 65.0 $\pm$ 7.0 to 61.8 $\pm$ 6.3 kg, respectively; P < 0.05). Body mass and fat-free mass recovered post-WR (71.84 $\pm$ 7.71 and 64.31 $\pm$ 6.42 kg, respectively; P < 0.05 compared with post-RWL). The ECW/ICW ratio assessed by BIS decreased post-RWL (0.68  $\pm$  0.03 to 0.65  $\pm$  0.03, P < 0.05), and recovered post-WR (0.69  $\pm$  0.03 P < 0.05 compared with post-RWL). Both TBW<sub>DIW</sub> and TBW<sub>BIS</sub> decreased post-RWL (46.4 $\pm$ 5.2 to 43.2 $\pm$ 4.8 L and 48.6 $\pm$ 6.9 to 45.3 $\pm$ 6.0 L, respectively; P < 0.05) and recovered post-WR (46.5 $\pm$ 4.9 L and 47.5 $\pm$ 6.6 L, P < 0.05 compared with post-RWL). The change in TBW<sub>BIS</sub> from baseline to post-RWL was similar to that in TBW<sub>DIW</sub> (-3.3 $\pm$ 0.3 and -3.2 $\pm$ 0.2 L, P = 0.720), but the change in TBW<sub>BIS</sub> from post-RWL to post-WR was smaller than was observed for TBW<sub>DIW</sub> (2.1 $\pm$ 0.3 and 3.2 $\pm$ 0.2 L, P < 0.05). In conclusion, BIS provides accurate assessment of dehydration status during RWL, but underestimates the rehydration status during WR.

#### The effect of an acute dose of inorganic dietary nitrate on intermittent sport-related performance in heat.

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Dietary nitrate (NO<sub>3</sub><sup>-</sup>) has been shown to improve endurance, self-paced events and intermittent, high-intensity exercise in temperate environments through its effect on enhanced blood flow, reduced O<sub>2</sub> and improved skeletal muscle efficiency. However, the ergogenic effects on sport-related performance in the heat has yet to be studied. The impact of heat stress on aerobic performance is well documented, where a rise in cardiovascular strain mediates a decrease in O<sub>2max</sub>. Twelve trained males (Mean $\pm$ SD age 22 $\pm$ 4 y; body mass 80.4 $\pm$ 5.8 kg; O<sub>2max</sub> 46 $\pm$ 6 ml $\cdot$ kg<sup>-1</sup> $\cdot$ min<sup>-1</sup>) took part in a randomised, double-blind crossover study. Participants ingested a nitrate rich (NR) beetroot juice shot (8.2 mmol NO<sub>3</sub><sup>-</sup>) or a placebo (PLA) (<0.004 mmol NO<sub>3</sub><sup>-</sup>) 3-hours prior to performing an intermittent sprint test in both hot (30°C, 70% RH) and temperate (22°C, 35% RH) conditions. The cycle ergometer protocol consisted of twenty maximal 6s sprints interspersed by 114s of active recovery, where heart rate (HR), RPE and core temperature (T<sub>c</sub>) were measured throughout. No significant treatment effects were found on sprint performance in hot or temperate environments (p > 0.05). Mean peak power in heat following NR was not reduced (p = 0.056; d = 0.28) compared to the PLA (659 $\pm$ 100 W vs. 693 $\pm$ 139 W, respectively). Similarly, total work done was not reduced in heat following the NR supplement (66 $\pm$ 11 kJ vs. 70 $\pm$ 15 kJ, respectively; p = 0.101, d = 0.28). The NR supplement did not reduce (p = 0.081, d = 0.27) mean power in heat (543 $\pm$ 101 W) compared to PLA (575 $\pm$ 133 W). No differences were found in HR, T<sub>c</sub> and RPE between treatments in either environmental condition (p > 0.05). There were no performance differences

between supplements in temperate conditions ( $p > 0.05$ ). In heat, acute dietary  $\text{NO}_3^-$  supplementation did not significantly reduce high-intensity, intermittent performance, though eight of the twelve subjects experienced performance decreases. An acute dose in heat may or may not affect performance negatively but it is hypothesised that a longer, loaded dose would elicit a detrimental effect on performance. An acute dose of dietary  $\text{NO}_3^-$  offered no aid to intermittent, sport-related exercise in ambient conditions.

### A Paleolithic hiking intervention improves acute but not chronic clinical markers of health in athletic male adults

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Imitation of Paleolithic lifestyle is associated with health-improving effects. Previous studies mainly described the benefit of a Paleolithic-like diet (PD) or Paleolithic-like movement pattern (PM) for overweight or diabetic people with a high cardiovascular risk. Aim of the study was to investigate the health-improving potential of a Paleolithic-like intervention for healthy athletic male adults. A group of 12 healthy athletic young adults (age=22±2 y; BMI=23.4±2.0 kg/m<sup>2</sup>;  $\text{VO}_2\text{max}$ =57.9±4.1 mL/min/kg) was exposed to the rural area of Lapland including PM and PD for 10 days (daily hiking distance: 13.0±2.5 km, 1 day rest). Testing occurred 6 days before, 6 days after the start of, and one day after (d-6, d6, d+1) the intervention, and again 3 months after the end of the intervention (d+91). Parameters included serum glucose (GLU), total cholesterol (CHO), high density lipoprotein cholesterol (HDL), and triglycerides (TG). Further, body mass (BM), fat mass (FM) and systolic blood pressure (SBP) were measured. We found a decline of BM, FM, SBP, CHO, GLU and TG during and directly after the intervention ( $p < .05$ ) (BM: d-6=76.0±2.1, d6=74.0±1.9, d+1=73.1±1.9 kg; FM: d-6=12.5±0.4, d+1=11.1±0.4 %; SBP: d-6=124±11, d+1=113±8 mmHg; CHO: d-6=3.8±0.2, d6=3.4±0.2, d+1=3.6±0.2 mmol/L; GLU: d-6=5.1±0.1, d6=2.8±0.7, d+1=4.7±0.1 mmol/L; TG: d-6=0.8±0.1, d6=0.6±0.0, d+1=0.6±0.0 mmol/L, respectively). HDL increased on d+1 but remained unchanged during and 91 days after the intervention ( $p > .05$ ) (d-6=1.3±0.1, d6=1.2±0.1, d+1=1.5±0.1, d+91=1.4±0.1 mmol/L, respectively). 3 months after the intervention BM increased and GLU decreased compared to d-6 ( $p < .05$ ) (d+91: BM= 77.8±2.1 kg; GLU=3.7±0.2; mmol/L, respectively). FM, SBP, CHO and TG regained the same values compared to pre-intervention ( $p > .05$ ) (d+91: FM=12.7±0.5 %; SBP=123±10 mmHg; CHO=3.7±0.2; TG=0.8±0.1 mmol/L, respectively). 10-day Paleolithic-like lifestyle intervention had acute small beneficial effects on some health related parameters in healthy physically active male adults. The intervention did not result in any longer-lasting effects on the parameters under investigation. However, the regain of BM 3 months after the intervention was

not due to an increase in FM, indicating some lasting positive effects in young and healthy male adults.

### Evolution of the body fat percentage of a professional soccer team over three seasons. (Spanish First Division "La Liga")

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Nowadays, Soccer is the sport which has the most practitioners, besides a lot of supporters. Professional football clubs, which play in the biggest leagues and championships, count on an increasingly high budget. This has led to soccer players receiving the best medical care, aiming to maintain an appropriate health and performance status, among other aspects. To provide these services, it is necessary that doctors, fitness coaches, rehab fitness coaches, physiotherapists, psychologists and nutritionists work closely together. The multidisciplinary approach of all these specialists can bring the players towards success. At the beginning of the season, one of the factors to monitor is the nutritional assessment, where the body composition measure is. The body fat percentage is one of the factors to consider, since the reference values in elite soccer players have proved a better performance and lower injury risks. For this reason, it is vitally important to know this percentage, in order to provide an ideal competition weight to the players. It will contribute to improve the nutritional strategies which will help them to enhance their performance. The aim of this study was to describe the evolution of the body fat percentage of a professional soccer team, over three seasons. Twenty five players from several nationalities and from the same team were followed for three seasons. Their ages, body weight and height were 24±4, 28±2 and 29±3 years old, 75±7, 77±5 and 76±6 kg, and 179±6, 178±6, 179±6 cm in the season 13/14, 14/15 and 15/16 respectively. We analyzed the triceps, subscapular, supraspinale and abdominal skinfolds following the guidelines of The International Society for the Advancement of Kinanthropometry (ISAK), by accredited researchers, calculating the body fat percentage using the Faulkner formula. The highest body fat percentage was found in July, at the beginning of the pre-season (12±2%, 12±1% and 11±1% in the season 13/14, 14/15 and 15/16 respectively). Comparisons between measurements were performed by analysis of variance (ANOVA), for checking the level of significance of the mean fat percentage of each of the monthly measurements per season. No differences ( $p > 0.05$ ) between the means monthly measurements per season were found. Despite the absence of significant differences, we concluded that the body fat percentage can change due to different factors, such as the ups and downs of the team in the championships or injuries. In the pre-season, players come with the highest levels of body fat percentage. Therefore, dietary monitoring of the players must be the key to improving their performance during the season.

### Effect of acute NO pathway supplementation on flow mediated dilation of the lower limb assessed by near infrared spectroscopy.

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Nitric oxide (NO) has several important implications in the body especially at the vascular level. It is implicated in the vasomotor mechanisms that regulate the diameter of the vessels in different physiological and pathological conditions. There are two main NO production pathways, one is NO synthase (NOS) independent with reduction of nitrate to nitrite and then nitrite to NO, and the other is NOS dependent by converting arginine to NO and citrulline. The objective of the study was to evaluate the effect of acute nitrate and citrulline supplementation on post-ischemic vascular response in healthy subjects. Fourteen subjects performed two leg vascular occlusion tests, 3 days apart. They were randomly assigned to consume a drink containing either 300 ml of beetroot juice which supplied 1200 mg of nitrate added with 6 g of citrulline (N+C) or a placebo (Pl). Changes in oxyhemoglobin (HbO<sub>2</sub>) and total hemoglobin (Hbtot) concentration were recorded by near infrared spectroscopy (NIRS) on the thigh and calf muscles. The reperfusion period was characterized by the difference between the baseline or the value at the end of occlusion period and the post-occlusion maximal values ( $\Delta$ max/baseline and  $\Delta$ max/min respectively) and area under the curve (AUC). Hbtot increased to a larger extent during the reperfusion period for the thigh ( $\Delta$ max/baseline 21.3 versus 19.5  $\mu$ mol,  $p=0.030$ ;  $\Delta$ max/min 20.5 versus 17.6  $\mu$ mol,  $p=0.010$ ; AUC 821 versus 627 mmol.s,  $p=0.003$ ) and the calf ( $\Delta$ max/baseline 20.0 versus 16.8  $\mu$ mol,  $p=0.010$ ;  $\Delta$ max/min 15.3 versus 12.9  $\mu$ mol,  $p=0.010$ ; AUC 515 versus 400 mmol.s,  $p=0.029$ ) in the N+C versus Pl condition. Similar results were found for HbO<sub>2</sub> for the thigh ( $\Delta$ max/baseline 13.7 versus 12.5  $\mu$ mol,  $p=0.010$ ;  $\Delta$ max/min 30.2 versus 28.7  $\mu$ mol,  $p=0.020$ ). Calf HbO<sub>2</sub>  $\Delta$ max/baseline only was different in N+C versus Pl (15.0 versus 13.6  $\mu$ mol,  $p=0.010$ ). The largest post-occlusive HbO<sub>2</sub> and Hbtot responses observed after N+C intake suggest a greater vasodilation which may be due to an increased NO availability via the activation of the two main NO production pathways.

### Vitamin B<sub>1</sub> and B<sub>6</sub> nutritional status in elite Polish athletes

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Vitamins B<sub>1</sub> and B<sub>6</sub> are necessary in the energy-producing pathways of the body. There is evidence that marginal or subclinical deficiencies of these vitamins might cause a decline in athletic performance; therefore the purpose of the present study was to

examine of B<sub>1</sub> and B<sub>6</sub> vitamin status and estimate the frequency of the risk of a deficiency of both vitamins in elite Polish athletes representing different sports. In the period of 2000-2014, the risk of vitamin B<sub>1</sub> deficiency was determined in 814 healthy athletes: 515 males and 299 females (2875 total measurements, 1896 in males and 979 in females), and risk of vitamin B<sub>6</sub> deficiency in 894 athletes: 567 males and 327 females (3290 total measurements, 2142 in males and 1148 in females). To assess vitamin B<sub>1</sub> and B<sub>6</sub> status the enzymatically determined activation coefficients (AC) of erythrocyte transketolase (ETK) for vitamin B<sub>1</sub> and of erythrocyte aspartic aminotransferase (EAST) for vitamin B<sub>6</sub>, were used. The cutoff values  $> 1.25$  for ETK AC and  $> 1.85$  for EAST AC were accepted as criteria of deficit of vitamin B<sub>1</sub> and B<sub>6</sub>, respectively. The ranges of ETK AC 1.20-1.25 for vitamin B<sub>1</sub> and of EAST AC 1.7-1.85 for vitamin B<sub>6</sub> were classified as marginal deficiency. The Shapiro-Wilk test was applied to test normality of the data and the ANOVA with the post hoc Tukey's HSD test was used for statistical analysis. The risk of vitamin B<sub>1</sub> deficiency was found in only 2% of all athletes. In the case of vitamin B<sub>6</sub> the risk of deficiency was higher, 11%, while its marginal deficiency was observed in 22% of subjects. The risk of deficiency of vitamin B<sub>6</sub> was higher than vitamin B<sub>1</sub>, both in individual years and in different sports. Athletes practising anaerobic sports were more exposed to the risk of vitamin B<sub>6</sub> deficiency than those practising endurance disciplines. Furthermore, women were more exposed to the risk of deficiency of both vitamins than men. The relatively high prevalence of vitamin B<sub>6</sub> risk among athletes confirms the need to monitor the nutritional status of this vitamin. The study was supported by the Ministry of Sport and Tourism of the Republic of Poland (2000-2014)

Relationships between vitamin D and iron status in male and female athletes

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Recent evidence suggests that vitamin D may affect iron regulation and erythropoiesis by its influence on hepcidin as well as on erythroid precursors. For these reasons deficit of vitamin D may deteriorate iron status and increase the risk of anaemia. We aimed to investigate whether vitamin D deficiencies have an impact on iron status in normal and iron-deficient athletes. The study included 290 healthy subjects (175 males and 115 females), aged 15-34 years, representing volleyball, cycling, canoeing, rowing, hockey, taekwondo and badminton. Blood for analysis was withdrawn in different seasons. Vitamin D status was assessed by measurements of 25(OH)D concentrations in serum. To assess iron status the concentrations of

ferritin, soluble transferrin receptor (sTfR), iron and total iron binding capacity (TIBC) were determined in serum. In addition to blood morphology, the mean erythrocyte and reticulocyte haemoglobin content (CH and CHr) and the percentage of hypochromic erythrocytes (HYPOm, LowCHm), reticulocytes (HYPOr, LowCHr) and microcytic erythrocytes (MICROm) were determined. Concentrations of 25(OH)D <10 ng/ml were classified as a deficit, values within 10-30 ng/ml as an insufficient concentration, and values > 30 ng/ml as normal. The frequencies of subjects with 25(OH)D < 30 ng/ml were 48% and 60% in females and males respectively, wherein deficit of vitamin D (<10 ng/ml) was observed in 6% of females and 12.6% of males only. Iron deficiency (without anaemia symptoms) was identified in 42% of female and 13.7% of male subjects. Despite the high prevalence of low concentrations of vitamin D and iron deficiencies, regardless of gender there were no associations between vitamin D concentration and iron status indices concerning either serum variables (ferritin, iron, TIBC and sTfR) or any haematological, i.e. red blood cell and reticulocyte, indices. These results indicate that vitamin D status does not have an impact on indices of iron status at least in athletes with normal and latent iron deficiency. It cannot be excluded that more profound deficiencies of both vitamin D and iron are required to observe the relationship between them. The study was supported by the Ministry of Sport and Tourism of the Republic of Poland (2014-2016)

#### Effect of physical activity intervention on overall physical fitness in women with and without Polycystic Ovarian Syndrome (PCOS)

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Polycystic ovary syndrome (PCOS) is a complex endocrine disorder in women of reproductive age. It is associated with increased risk of type 2 diabetes, hypertension, coronary heart disease, gestational diabetes and endometrial cancer. Physical activity/exercise intervention is the first line approach to manage PCOS. The present study evaluated the effect of a physical activity intervention on outcome of PCOS and physical fitness in women with and without PCOS. The study participants comprised of 24 young women aged 20-30y. These women were divided in three groups- Control group-1(n=8, women without PCOS with exercise intervention), Control group-2(n=8 women with PCOS without exercise intervention) and Experimental group (n=8 women with PCOS with exercise intervention). Exercise intervention was given for 8 weeks to Control group 1 and Experimental group. The exercise included 30min of brisk walking, 10-15min of stretching and muscular related activities 5 days in a week. Dietary nutrient intake (3days recall), body fat (4 site-skin fold; Durnin Womersely equation), body circumferences (waist, hip, thigh, calf), Hemoglobin and physical fitness assessment (Bruce protocol, muscular strength and endurance and flexibility) was done at baseline, 4 weeks and 8 weeks. The data were analyzed using SPSS version 16, with paired t test, ANOVA and Pearson's correlation. Exercise intervention

improved body composition in Control 1 and Experimental group in terms of waist circumference ( $p<0.01$ ) and percent body fat, but weight loss was not noticeable. Both these groups showed improvement in aerobic capacity ( $VO_{2max}$   $32\pm 9$  to  $36\pm 6$  ml/Kg/min) ( $p<0.01$ ), muscular endurance ( $p<0.05$ ) and flexibility, with greater improvements in Experiment Group ( $p<0.01$ ). Exercise also improved the overall condition of women with PCOS. In conclusion, exercise intervention for 8 weeks improved the overall fitness of women with and without PCOS and also had a positive impact on PCOS condition.

#### Physiological effects of acute intake of Irish blackcurrant juice during an incremental cycling test in endurance athletes.

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The polyphenol compound anthocyanin present in blackcurrant juice (BC) has been found to elicit positive biological action in terms of cardiovascular and metabolic functions. Supplementation of BC may also enhance aerobic exercise performance. With seven days intake of New Zealand blackcurrant powder, a shift of the lactate curve was observed with 4 mmol/l of lactate obtained at higher power output. Acute effects of BC intake on responses during exercise are not fully known. Therefore, the actions of an acute intake of anthocyanins in BC may affect exercise performance benefits in relation to blood lactate, power output, heart rate and time in a standard exercise ramp test. This study set out to ascertain if acute supplementation of a high dose of BC affects responses during an incremental cycling protocol in healthy trained cyclists. The primary purpose of this study was to examine whether a single dose of BC ingested prior to the endurance ramp protocol may affect parameters at a blood lactate (BLA) value of 4mmol/l. The design was a randomized controlled trial using blinding. The study sample was 14 male endurance athlete: height ( $175\pm 7$  cm), weight ( $71.3 \pm 8.9$  kg), age ( $36\pm 6$  y). Each subject was blinded and received either a BC or placebo (PLa) juice in 500ml liquid with 300ml of BC containing 105mg of anthocyanin or coloured water 2h prior to testing. The endurance ramp protocol test to 4mmol/l BLA was performed on a Watt bike ergometer starting at 125W with 3 min stages of 25W increments to reach 4mmol/l BLA. Power, time and heart rate were measured at each 3 min interval; all metrics were recorded, compared using t-test and analysed on Graphpad Prism statistical software ( $P<0.05$ ). There was difference after a high dose of BC in power (BC:  $227\pm 5$  W (n=6), PLa:  $222\pm 5$  W (n=6),  $P=0.54$ ), time (BC:  $15.3\pm 0.6$  min (n=6), PLa:  $14.2\pm 0.9$  min(n=6),  $P=0.37$ ) and heart rate (BC:  $155\pm 2$ beats/min (n=6), PLa:  $160\pm 3$ beats/min (n=6),  $P=0.29$ ) to reach a blood lactate value of 4 mmol/l. A single dose of BC had no effect on power, time or heart rate in an endurance ramp protocol test to reach a blood lactate value of 4mmol/l. Future studies may examine how many days intake of blackcurrant are required to observe changes in physiological and performance responses during exercise testing in trained endurance athletes.

A2 milk enhances dynamic muscle function following repeated sprint exercise

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Hyperaminoacidemia following ingestion of dairy-milk may stimulate muscle anabolism and attenuate exercise-induced muscle damage (EIMD). However, athletes who are dairy intolerant do not get the reported benefits from consuming milk-based products; but A2 milk may offer an alternative as it does not contain the A1-Protein. This study aimed to determine the effect of A2 milk on recovery from a sports-specific muscle damage model (Keane et al., 2015). Twenty-one male team sport players (n=21) were randomly allocated to three independent groups: (A2 milk n=7, regular milk n=7, CHO-matched placebo (PLA) n=7). Immediately following muscle damaging exercise participants consumed either A2 milk, regular milk or PLA (3 x 500ml). Visual analogue scale (muscle soreness), maximal voluntary isometric contraction (MVIC), countermovement jump (CMJ) and 20m sprint were measured pre, 24, 48, and 72 h post-EIMD. A2 and regular milk demonstrated a beneficial effect at 48h in minimising decrements in CMJ (p=0.02 and p=0.02, respectively) and 20m sprint (p=0.04 and p=0.03, respectively) vs. PLA. Relative to baseline, 48h CMJ and 20m sprint recovered quicker in A2 (33.4±9.6 and 3.3±1.4%, respectively) and regular milk (33.1±9.5 and 3.3±1.2%, respectively) vs. PLA (29.2±14.8 and 3.6±2.9%, respectively). Ingestion of 500ml of A2 or regular milk after muscle damaging exercise can limit decrements in dynamic muscle function in male athletes, thus hastening recovery, and increase subsequent performance; but evidently not isometric strength performance or active muscle soreness (p>0.05). The findings propose A2 milk as a suitable recovery beverage following EIMD, and may offer an alternative to athletes who experience gastrointestinal discomfort with the digestion of A1-Protein.

#### Matcha green tea drinks enhance fat oxidation during brisk walking in females

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Epigallocatechin gallate (EGCG) is present in tea and considered an essential bioactive catechin. Intake of capsules of EGCG has been shown to enhance fat oxidation during exercise. Matcha green tea powder contains catechins and caffeine and is consumed as a drink. We examined the effect of Matcha green tea drinks on metabolic, physiological and perceived intensity

responses during brisk walking in females. Thirteen females (mean±SD age: 27±8 y, body mass: 65±7 kg, height: 166±6 cm, BMI: 23.5±2.6 (range 19.2-30.1) volunteered. In the 1<sup>st</sup> visit, resting metabolic equivalent (1-MET) using Douglas bags was measured (1-MET: 3.43±0.27 ml·kg<sup>-1</sup>·min<sup>-1</sup>) and participants completed an incremental walking protocol to establish the relationship between walking speed and MET. A randomized cross-over design was used with participants tested between day 9 and 11 of the menstrual cycle (follicular phase). In the Matcha condition, participants consumed 3 drinks (each drink made with 1 gram of Matcha premium grade, OMGTea Ltd, UK) the day before and 1 drink 2 hours before the 30 min walk at 5 (n=10) or 6 METs (walking speed: 5.8±0.4 km·hr<sup>-1</sup>, range 5.3-6.5 km·hr<sup>-1</sup>) with responses measured at 8-10, 18-20 and 28-30 min and averaged. Matcha had no effect on heart rate (control: 119±18, Matcha: 120±17 beats·min<sup>-1</sup>), minute ventilation (STPD) (control: 25.7±3.3, Matcha: 25.2±3.3 L·min<sup>-1</sup>), oxygen uptake (STPD) (control: 18.10±2.79, Matcha: 18.09±2.76 ml·kg<sup>-1</sup>·min<sup>-1</sup>) and rating of perceived exertion (control: 11±1, Matcha: 11±3). Matcha drinking resulted in lower respiratory exchange ratio (control: 0.84±0.04, Matcha: 0.82±0.04, t-test, P=0.004) and enhanced fat oxidation by 18.4±29.2% (range -10.1-98.0%, P=0.004). Absolute fat oxidation for the 30 min brisk walks were 9.1±3.1 and 10.3±3.1 gram for the control and Matcha condition (P=0.004). Matcha green tea drinks do not result in adverse physiological responses and are beneficial to substantially enhance fat oxidation during brisk walking in females. Future studies should examine effects of Matcha green tea drinks over a longer period in combination with regular brisk walking on fat oxidation during exercise in females. Matcha green tea may have health implications in females when regular exercise is undertaken as part of a weight loss program. Acknowledgement: We thank OMGTea Ltd (UK) for Matcha green tea powder and support for conference attendance.

#### Flaxseed oil supplementation does not attenuate the 48-hour inflammatory response associated with a simulated 16-km fell run

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This study assessed the influence of a six-week flaxseed oil supplementation on inflammatory markers and subjective muscle soreness for 48-hours following a simulated 16-km fell run. Twenty well-trained male runners (age: 33±8 years, O<sub>2</sub>peak of 61±9 ml·kg<sup>-1</sup>·min<sup>-1</sup>) were randomly assigned to one of two conditions; a flaxseed oil supplementation group (FLAX), or placebo control (PLA). FLAX received a daily dose of 15ml/25kg·BM<sup>-1</sup> delivered via addition to a fruit-based beverage. PLA received the same beverage but without the addition of the flaxseed oil supplement. Both groups completed a 16-km simulated fell run on a motorised treadmill at baseline, and following 6-weeks of supplementation or placebo. Periodic blood sampling captured C-reactive protein (CRP), creatine-kinase



(CK), and interleukin-6 (IL-6) up to 48-hours post-exercise. Muscle soreness was concurrently assessed using the Borg CR10 scale. Pre- and post-exercise baseline blood measures and indices of muscle soreness taken prior to the intervention were similar between groups ( $P>0.05$ ). Following the 6-week intervention, pre-exercise blood measures and indices of muscle soreness remained similar to baseline values ( $P>0.05$ ). IL-6 concentrations were greatest immediately after exercise, increasing 7-fold under both conditions; CK and CRP peaked at 24-hours post-exercise. IL-6, CK, and CRP under both FLAX and PLA remained similarly elevated from pre-exercise concentrations at 48-hours post-exercise. There were no condition ( $P>0.05$ ) or time ( $P>0.05$ ) effects, nor a condition-X-time interaction ( $P>0.05$ ) in post-exercise inflammatory markers or muscle soreness indices. Six-week supplementation of 15ml/25kg. BM<sup>-1</sup> flaxseed oil prior to a 16-km simulated fell run does not attenuate associated elevations in CRP, CK, IL-6, or subjective muscle soreness over 48-hours post-exercise.

#### Effect of a one week alpine trek on submaximal and maximal physiological measures

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Previous studies, typically over a 6-8 week time frame, have shown lower resting and submaximal exercise heart rates and improved aerobic fitness in men and women. The aim was to investigate any change in physiology resulting from a 7-day alpine trek (Tour du Mont Blanc, TMB; 150-km). Between 2009 and 2015, 92 men (mean±SD) (age 21±1 y; height 1.80±0.07 m; body mass 77.8±14.3 kg) and 78 women (age 21.2±1.9 years; height 1.67±0.06 m; body mass 63.3±10.4 kg) participated. A submaximal treadmill test (Woodway, PPS 55) at 1.50 m s<sup>-1</sup> (4-min at each of 0, 4, 8, 12 and 16% gradient) assessed walking economy (Cortex Metalyzer). A separate test to volitional exhaustion measured the maximal aerobic power. Tests were repeated at the same time of day 10-days later. Pre- versus post-TMB differences were assessed using Student's t-test for paired

samples (SPSS version 22). Maximal aerobic power increased in both men (52.9±6.4 vs. 54.1±6.2) and women (43.7±5.7 vs. 46.2±5.4 mL kg<sup>-1</sup> min<sup>-1</sup>) ( $P<0.01$ ). Submaximal measures for men and women are shown in Table 1.

The present study shows significant improvements in walking economy, decreased heart rate at the same treadmill gradient, and improved oxygen delivery after compared with before TMB. There was no change in substrate utilization assessed by the respiratory exchange ratio.

#### Effect of the time of carbohydrate intake on muscle glycogen recovery post-exercise

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Muscle glycogen synthesis rate is lowered when post-exercise CHO ingestion is delayed 2 hours compared with CHO ingestion immediately post-exercise. However, effect of CHO intake at 1 hour post-exercise on muscle glycogen recovery, is not clear. The purpose of this study was to evaluate the effect of the time of CHO intake on post-exercise muscle glycogen recovery. Six male endurance-trained athletes (age: 21±2 y; height: 170±8 cm; body mass: 61.3 ± 5.7 kg) consumed CHO (2.0 g/kg body weight) either immediately post-exercise (P-EX) or 60 minutes post-exercise (60P-EX) after a time trial (TT) on a cycle ergometer at 70% maximum power for approximately 60 min (842 ± 50 kJ). They were directed to complete the TT as soon as possible. The glycogen contents of thigh muscle group of the right leg: the vastus lateralis and vastus intermedius muscles were measured by carbon magnetic resonance spectroscopy (<sup>13</sup>C-MRS) before exercise and post-exercise, at 1, 2, 3, and 4 hours after exercise. Plasma glucose, insulin, and free fatty acid concentrations were also measured at the same time points. The TT times were 67.7±3.6 minutes for P-EX, and 65.8±5.4 minutes for 60P-EX. Muscle glycogen levels were decreased by exercise to 54.8 ± 2.4 % of pre-exercise levels for P-EX, and recovered to 68.0 ± 1.5 % of pre-exercise levels 4 hours

**Table 1: Physiological responses before and after a one-week alpine trek**

Gradient (%)	Oxygen uptake		Heart rate		Oxygen pulse		RER	
	(mL kg <sup>-1</sup> min <sup>-1</sup> )		(b min <sup>-1</sup> )		(mL beat <sup>-1</sup> )			
	pre-	post-	pre-	post-	pre-	post-	pre-	post-
0	15.9±1.9	15.1±1.9**	107±17	101±14**	15.2±2.8	15.1±2.2	0.82±0.07	0.84±0.07**
4	19.1±2.1	18.6±2.3**	116±17	111±15**	16.7±3.0	16.8±2.0	0.86±0.06	0.87±0.06
8	24.1±2.6	23.6±2.8*	133±17	127±18**	18.4±2.4	18.7±2.3*	0.89±0.06	0.90±0.06
12	29.7±2.9	29.3±3.1	151±19	145±18**	19.9±3.0	20.4±3.0**	0.94±0.06	0.93±0.07
16	35.3±3.7	34.8±3.2*	166±18	160±17**	21.5±2.9	21.9±2.6**	0.98±0.08	0.98±0.07

\* $P<0.05$  and \*\* $P<0.01$ , significant difference between pre- and post-TMB values; RER, respiratory exchange ratio

after exercise for 60P-EX. At 1 h after exercise, the recovery rate of muscle glycogen was higher for P-EX than for 60P-EX ( $P < 0.05$ ). At the other time points, there was no difference between P-EX and 60P-EX for the muscle glycogen recovery. Increased plasma glucose levels were more frequent for P-EX compared with 60P-EX at 1-2 hours after exercise ( $P < 0.05$ ), and at 3 hours after exercise, for 60P-EX. Insulin levels were higher for P-EX compared with 60P-EX at 1 hour ( $P < 0.05$ ), and for 60P-EX compared with P-EX at 3 hours ( $P < 0.01$ ). Increased levels of plasma free fatty acids were observed for 60P-EX compared with P-EX at 1 hour after exercise ( $P < 0.05$ ), and for P-EX compared with 60P-EX at 4 hours ( $P < 0.01$ ). In conclusion, increased muscle glycogen recovery occurs 1 hour post-exercise when CHO intake is immediately after exercise, compared with a 60-minute delay; however, 4 hours after exercise, the influence of the timing of CHO intake is small.

#### **Magnesium status in Olympic & Paralympic athletes compared with a sedentary population and validation of a FFQ as an assessment tool for dietary magnesium intake**

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Magnesium (Mg) is an essential mineral involved in over 350 enzymatic reactions within the human body and plays a fundamental role in electrolyte balance, cellular growth and glucose homeostasis. Literature suggests Mg deficiencies may be prevalent within athletic populations, but it is not clear whether this is due to reduced dietary intakes or increased utilisation of Mg due to high levels of physical activity. The aims of the study were two-fold. Firstly to validate a food frequency questionnaire (FFQ) against a 4-day diet diary and to compare intakes with blood samples for its potential to predict status. Secondly, to compare dietary intakes and plasma Mg status in elite athletes compared with a sedentary population. Thirty-eight individuals participated in the study; 24 elite athletes (Olympic & Paralympic track and field) and 14 non-athletes (age 18-30 y). Participants completed a FFQ and provided a venous blood sample. In addition, the non-athlete group completed a 4-day diet diary. Analysis of plasma Mg was carried out using ICP-MS. Plasma Mg status was lower in elite athletes compared with non-athletes ( $1.2 \pm 0.1$  vs  $1.4 \pm 0.3$  mg/dL;  $p=0.03$ ), despite similar intakes of dietary Mg ( $431 \pm 140$  vs  $458 \pm 165$  mg;  $p>0.05$ ). Self-reported Mg intake assessed by FFQ was correlated to intakes from diet diaries from the non-athlete group ( $p<0.01$ ), suggesting FFQ to be a valid tool for dietary assessment. Dietary intakes were not correlated with plasma Mg status. Lower plasma Mg levels in athletes compared with non-athletes suggest enhanced nutrient demands during exercise may be causing a transient shift of Mg from the plasma to sites of metabolic demand. Red blood cell (RBC) analysis may provide greater insight into overall Mg status in athletic populations, despite plasma analysis cur-

rently portrayed as the gold standard measurement for clinical investigations. The lack of correlation between Mg intake and status was likely the result of the transient nature of plasma Mg analysis. Intestinal absorption and excretion of Mg is critical in the maintenance of plasma status, therefore high consumption does not necessarily result in elevated plasma levels. Repeated analysis of Mg across a season is important for maintaining health and performance in athletes and should be assessed in RBCs where possible.

#### **The effects of coconut water vs. sports drink on physiological performance in male soccer players**

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To compare a commercially manufactured sports drink versus coconut water in maintaining endurance sport performance. From the relevant research, the hypothesis of this study is: Coconut water, a more natural alternative beverage will elicit the same enhancing properties on performance as that of a manufactured sports drink. The null hypothesis is that no difference will be noted in maintenance of endurance performance after consumption of either a sports drink or coconut water. Eight healthy male collegian soccer players participated in this study. Prior to testing, subjects were required to attend the laboratory to establish reliability and for familiarisation. On two other occasions the research trial protocol was carried out, where each subject consumed either a sports drink or coconut water. Each subject carried out two trials on separate days of a modified Loughborough Intermittent Shuttle run Test, in which they ingested one of the fluids throughout 15 minute intervals of the trial. The variables that were investigated include total distance covered, heart rate, blood glucose levels and urine osmolality. Shapiro-Wilk Test found that the data was not normally distributed ( $p<0.05$ ). Greenhouse-Geisser and Huynh-Feldt testes were used to assess the effect of violating the assumption of severity. A Two-Way Repeated Measures Anova showed no significant difference between testing days or between variables: performance distance ( $p>0.222$ ), urine osmolality ( $p>.153$ ), heart rate ( $p>.412$ ), blood glucose ( $p>0.447$ ) for both coconut water and sports drink. Sports drinks are widely used to promote rapid fluid replacement because their electrolyte content helps keep ingested water in the body while their taste and sodium content stimulate the thirst mechanism and promote voluntary uptake, ensuring the athlete drinks until fully rehydrated. It was discovered in this study that 66.7%, ( $=4/6$  SD) observed a lower distance covered with consumption of the sports drink. This may due to the fact coconut water is more palatable due to its less sodium and higher potassium content than the sports drink and the subjects found it easier to consume, therefore enhancing their ability of physiological performance maintenance. Despite performance distance not being significantly different, ( $p>0.222$ ) the subjects still had an overall higher distance covered (4.6%) ingesting coconut water when compared to the ingestion of sports drink. Coconut water deserves more

commercial attention as a natural alternative to sports drink as it has proven to demonstrate the same enhancing effects on physiological performance as that of a sports drink yet it is a healthier alternative.

**Bi-monthly highdose vitamin D supplementation maintains vitamin D status in military recruits over the winter months in the UK; A double blind, randomised, placebo-controlled trial.**

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A growing body of evidence links vitamin D status with risk of injury and illness in athletic and military populations. Training days lost through musculoskeletal injury and illness represent a significant financial and operational burden to the Armed Forces, resulting in lengthy rehabilitation, medical downgrading and failure to complete military training. The Vitamin D Supplementation in the Armed Forces (D\_SAF) study is investigating whether vitamin D supplementation reduces illness and injury occurrence in Royal Marines (RM) recruits undertaking the 32-week Commando training course. The Pilot Study phase of this project assessed the efficacy of intermittent high-dose supplementation to maintain vitamin D status (assessed by measuring serum 25-hydroxy-vitamin D (25OHD) concentration), where daily doses were deemed unfeasible during military training. Volunteer recruits (*n* 534), were randomly assigned to receive either placebo (PLA), 25,000 IU (625 µg) vitamin D3 (LOW), or 50,000 IU (1250 µg) vitamin D3 (HIGH), in a double-blind approach, at weeks 1, 6, 15 and 24 of training. Blood samples were drawn at weeks 1, 6, 15, 24 and 31 of training and analysed for calcium, phosphate, albumin and 25OHD status. Week-6 and 15 blood sample data for recruits who started training in Dec and Jan (*n* 77) were compared to assess the impact of vitamin D supplementation during winter months. ANOVA identified differences in 25OHD between supplement groups at week-6 [ $F(2,75)=8.9, p<0.001$ ] and week-15 [ $F(2,75)=6.1, p<0.005$ ]. Mean (SD) 25OHD (nmol.L<sup>-1</sup>): at Week 6: PLA 53.2 (4.2), LOW 59.6 (3.1), HIGH 72.8 (2.7); and Week 15: PLA 53.2 (8.9), LOW 61.2 (4.7), HIGH 68.4 (1.5). Post-hoc tests (Tukey HSD) identified that the significant differences in 25OHD occurred between the PLA and LOW vs. HIGH groups. Calcium and phosphate remained within normal ranges, and no adverse reactions were reported from supplementation. Intermittent high-dose vitamin D supplementation (50,000 IU) is an effective and practical strategy for maintaining vitamin D status in military recruits. Further research is under way to investigate concomitant health benefits.

**The effects of caffeinated gum and caffeine capsules on running sprint performance**

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Use of anhydrous caffeine is an established and widely used ergogenic method. In sprinting events, optimum performance is highly dependent on the simultaneous peak functioning of a host of physiological systems. Therefore, caffeine supplementation protocols need to be perfectly timed in order to achieve culmination in sprint performance parameters within a narrow time window. Typically, caffeine capsules are ingested approximately 1-hour before exercise, but absorption rates may be highly variable. An alternative mode of ingestion is through caffeinated gum where caffeine is rapidly absorbed through the buccal mucosa. Our aim was to investigate the acute effects of two distinct modes of caffeine ingestion on sprint performance. Following ethics approval, eight trained male sprinters aged 20±1 y took part in a screening and familiarisation session before they completed four trials (3x40 m sprints with 4 min recovery between runs) a week apart. A double-blind randomized crossover design was adopted where, during the trials, participants received: 1) Caffeine gum (CAFG, 6 mg.kg<sup>-1</sup> of body weight), 2) CAFG placebo (CAFPG), 3) Caffeine capsules (CAFC, mg.kg<sup>-1</sup> of body weight), 4) CAFC placebo (CAFPC). General and sport-specific warm-up commenced 15 minutes before sprint one. Capsules were given 45 minutes and chewing gums 15 minutes before sprint one. The gums were chewed for 5 minutes. Blood lactate and glucose concentration, heart rate, arousal and feeling levels were recorded at baseline and different time points during testing. Mean times to complete the three sprints were 5.00±0.23, 5.03±0.17, 5.10±0.15, and 5.10±0.14 s for the CAFG, CAFC, CAFPG and CAFPC conditions respectively. Participants ran faster ( $P<0.05$ ) during the caffeine than the placebo conditions. Additionally, sprint two in the CAFG (2.25±0.45 % faster than CAFPG) was faster ( $P=0.022$ ) than in the CAFC (1.40±0.32 % faster than CAFPC). Blood glucose and arousal levels were also higher during the caffeine trials. Our data confirm that caffeine is an effective ergogenic strategy for sprinters. Furthermore, the greater performance gains in sprint two suggest that caffeinated gum may be a more efficacious mode of ingestion than traditional methods of caffeine ingestion.

**Estimates of energy intake and expenditure in elite female Touch players assessed during an international tournament**

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Touch Rugby (Touch) is an intermittent high-intensity sport. At an international level, players are required to compete in

up to three 40-minute matches per day over four consecutive days. Players must be adequately fuelled to achieve optimum performance and recovery. Studies are yet to estimate the total daily energy expenditure (TDEE) and energy intakes (EI) of elite Touch players. Therefore, this study aimed to assess the TDEE, EI and macronutrient intakes of elite female Touch players during an international competition. Sixteen elite female Touch players (age:  $27.4 \pm 6.2$  years, stature:  $1.63 \pm 0.06$  m, body mass:  $60.0 \pm 6.5$  kg, BMI:  $22.5 \pm 2.3$  kg·m<sup>-2</sup>) from the 2016 England Women's squad were recruited. Participants wore GPS technology during matches and accelerometers outside of matches over four consecutive days at the 2016 European Touch Championships. These data were combined with estimates of resting energy expenditure to estimate TDEE. Participants completed four-day self-reported food diaries to estimate EI and macronutrient intakes. TDEE ( $43.6 \pm 3.1$  kcal·kg<sup>-1</sup>) was not different ( $p > 0.05$ ) from EI ( $39.9 \pm 9.4$  kcal·kg<sup>-1</sup>). Carbohydrate intakes ( $4.4 \pm 0.6$  g·kg<sup>-1</sup>, 45% total kcal) were below ( $p < 0.05$ ) the current recommended intakes (6–10 g·kg<sup>-1</sup>). Protein ( $2.0 \pm 0.8$  g·kg<sup>-1</sup>, 20% total kcal) and fat ( $1.7 \pm 0.5$  g·kg<sup>-1</sup>, 37% total kcal) intakes were in line with the recommendations (protein: 1.2–2 g·kg<sup>-1</sup>, fat: 20–35% total kcal) whereas saturated fat intakes ( $13 \pm 3\%$  total kcal) were greater ( $p < 0.05$ ) than the recommendations (10% total kcal). For the first time, this study reports that elite female Touch players maintain energy balance during an international tournament. Carbohydrate intakes appear insufficient when compared to the current sports nutrition recommendations, but future work should look to establish whether these recommendations are applicable to Touch. Further research is also required to determine if findings here are representative of elite level Touch or unique to the cohort studied.

#### Vitamin D status is an important predictor of aerobic performance in male and female Army recruits

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Reports are conflicting regarding the association between physical performance and vitamin D status (total 25-hydroxyvitamin D [25(OH)D] concentration in blood). Our aim was to determine if an association exists between 25(OH)D concentration and physical performance in a large sample of male and female Army recruits, whilst controlling for important covariates (e.g. body composition). While previous investigations have focused on strength and power measurements, we also investigated

aerobic performance. Participants included 969 British Army recruits aged  $22 \pm 3$  years commencing 13-weeks training (64% male; 36% female). Performance assessments completed at week 1 were a 1.5-mile run, maximal dynamic lift strength, and maximal vertical jump. A blood sample was obtained for 25(OH)D measurement by LC-MS/MS, and haemoglobin by a coulter counter. Body composition was assessed by dual-energy x-ray absorptiometry (DXA), and smoking by a questionnaire. After 13-weeks training, a follow-up cohort ( $n = 331$ , 51% male; 49% female) repeated these measurements. After controlling for fat mass, smoking and season in a hierarchical multiple linear regression, 25(OH)D explained 4–6% of variance in 1.5-mile run time ( $P < 0.001$ ). 25(OH)D was a better predictor of 1.5-mile run time than body mass index (2–3%), lean body mass (2–4%), haemoglobin (0–2%), and smoking (0–1%). In practical terms, recruits with 25(OH)D  $\geq 75$  nmol/L ran 1.5-miles ~30 s faster than recruits with 25(OH)D concentration  $< 30$  nmol/L:  $614 \pm 51$  vs.  $638 \pm 50$  s for males;  $715 \pm 69$  vs.  $750 \pm 73$  s for females ( $P < 0.05$ ). After 13-weeks of military training under similar conditions of activity, nutrition, and sleep, 25(OH)D once again explained 3–6% of variance in 1.5-mile run time, indicating the robust nature of this association. No significant associations were observed between 25(OH)D and strength or power. These findings show that vitamin D status as assessed by 25(OH)D concentration is an important predictor of aerobic performance in a large sample of young, healthy male and female Army recruits. Future studies should investigate the effect of vitamin D supplementation on aerobic physical performance.

#### Effects of a pre-training rich CHO/protein snack intake on S-RPE, fatigue and physical performance of adolescent athletes.

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Scientific evidence shows that adolescent athletes usually omit lunch intake, especially when they train in the afternoon. However, a high carbohydrate (CHO)/protein (P) snack 2–4 h before training could supply the nutrients not consumed at lunch, optimizing glycogen stores. Thus, fatigue (F) and internal load training, measure as session rating of perceived exertion (S-RPE) could decrease, improving recovery and physical performance (PP). Our aim was to assess the effects of a pre-training rich CHO/P snack intake on S-RPE, F and PP of adolescent athletes. A 7-weeks experimental study was carried out with 26 healthy runners (13–16 years) belonging to a Spanish club team. Athletes were equitably randomized into an experimental group (EG) or a control group (CG). EG

received a rich CHO/P snack (1g CHO/Kg body weight (BW) + 0.25g P/Kg BW) and water (5-7 ml/Kg BW) 2 hs before each training while CG only drank water (5-7 ml/Kg BW). Pre and post intervention (INT): food intake was assessed (3 day food diary), anthropometric measures were taken (ISAK), PP tests were done (day 1: CMJ test, long jump test, sprint test-40 m and 60 m; day 2: mile test) and S-RPE (0-10 scale) (Foster et al., 2001) and fatigue before (FB) and after training (FA) (1-10 scale) (Del Campo, 2005) were recorded. Statistic: the groups were compared pre and post INT using the independent T-Test and U-Mann-Whitney test for parametric (PV) and non-parametric variables (NPV) respectively. Pre and Post INT data of each group were compared using paired T-Test (PV) and Wilcoxon test (NPV). After INT, S-RPE decreased in EG (day 1:  $4.5 \pm 1.3$  vs  $3.3 \pm 1.6$ ;  $t_{(12)} = 6.1$ ;  $p < 0.05$ ) and increased in CG (day 1:  $4.1 \pm 1.4$  vs  $5.3 \pm 1.4$ ;  $t_{(12)} = -7.4$ ;  $p < 0.05$ ) with differences between groups (day 1:  $5.3 \pm 1.4$  vs  $3.3 \pm 1.6$ ;  $t_{(24)} = 4.1$ ;  $p < 0.05$ ). FB increased in CG (day 1:  $1.2 \pm 0.4$  vs  $2.0 \pm 1.0$ ;  $Z = -2.7$ ;  $p < 0.05$ ; day 2:  $2.2 \pm 0.9$  vs  $2.7 \pm 1.3$ ;  $Z = -2.1$ ;  $p < 0.05$ ) being higher than EG (day 1:  $2.0 \pm 1.0$  vs  $1.2 \pm 0.4$ ;  $Z = -2.2$ ;  $p < 0.05$ ; day 2:  $2.7 \pm 1.3$  vs  $1.3 \pm 0.5$ ;  $Z = 3.1$ ;  $p < 0.05$ ). EG also decreased FA (day 1:  $5.1 \pm 1.3$  vs  $3.9 \pm 1.4$ ;  $t_{(12)} = 4.2$ ;  $p < 0.05$ ) and improved the Mile test ( $8.3 \pm 0.9$  min-seg vs  $7.8 \pm 0.9$  min-seg;  $t_{(12)} = 4.7$ ;  $p < 0.05$ ). The intake of a pre-training rich CHO/P snack would improve aerobic capacity of adolescent athletes, reducing S-RPE and fatigue after training while its omission could increase S-RPE and fatigue before training.

#### Nitrate-rich beetroot juice supplementation enhances short but not longer duration running time-trial performance in trained males

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This study evaluated the effects of nitrate-rich beetroot juice supplementation on exercise performance in trained males conducting short and longer duration self-paced treadmill running time-trials (TT). Eight male runners or triathletes (age:  $28.3 \pm 5.8$  years, body mass:  $74.7 \pm 10.1$  kg, height:  $179.1 \pm 2.4$  cm,  $\dot{V}O_{2max}$ :  $62.3 \pm 8.1$  ml·kg<sup>-1</sup>·min<sup>-1</sup>) completed four exercise performance tests in a randomised order, comprising 10 minutes of moderate-intensity exercise followed by either a 1500 m or 10,000 m treadmill TT. Participants were supplemented double-blind with either 140 ml concentrated nitrate-rich (BRJ; ~ 12.5 mmol nitrate) or nitrate-deplete placebo (PLA; ~ 0.01 mmol nitrate) beetroot juice, 3 hours before the start of exercise. Pre-exercise plasma nitrite concentrations were significantly elevated in BRJ compared with PLA prior to both the 1500 m ( $661.3 \pm 263.0$  vs.  $114.8 \pm 15.8$  nM;  $p =$

$0.001$ ) and 10,000 m ( $500.5 \pm 210.5$  vs.  $102.8 \pm 16.1$  nM;  $p = 0.002$ ) TTs. The increase in plasma nitrite concentrations in BRJ compared with PLA was not significantly different between 1500 m and 10,000 m TTs ( $p > 0.05$ ). Post-exercise plasma nitrite concentrations significantly decreased in the BRJ 1500 m condition compared to pre-exercise levels ( $\Delta 272.0 \pm 229.4$  nM;  $p = 0.01$ ), but did not change relative to pre-exercise levels in all other conditions (all  $p > 0.05$ ). Post-exercise blood lactate concentrations were significantly greater in BRJ compared with PLA following the 1500 m TT ( $6.6 \pm 1.2$  vs.  $6.1 \pm 1.5$  mM;  $p = 0.02$ ), but not significantly different between conditions following the 10,000 m TT ( $p = 0.96$ ). Resting blood pressure and exercise  $\dot{V}O_2$  were not significantly different between BRJ and PLA (all  $p > 0.05$ ). Performance in the 1500 m TT was significantly faster in BRJ versus PLA ( $319.6 \pm 36.2$  vs.  $325.7 \pm 38.8$  s;  $p = 0.03$ ). Magnitude based inferences indicated a 'very likely beneficial' effect of BRJ on 1500 m TT performance. Conversely, there was no significant difference in 10,000 m TT performance between conditions ( $2643.1 \pm 324.1$  vs.  $2649.9 \pm 319.8$  s;  $p = 0.77$ ). Magnitude based inferences indicated an 'unclear' effect of BRJ on 10,000 m TT performance. These data suggest that BRJ may be ergogenic in trained males conducting short but not longer duration exercise tests.

#### Effects of a 68-day military course on body composition and performance

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The Singapore Ranger Course exposes trainees to a combination of high physical activity, food restriction, sleep deprivation, mental and psychological stress, unfamiliar environment and extreme weather conditions. This study aimed to understand the impact of these demands on physical and cognitive performance before (PRE) and after (POST) the course. Mean  $\pm$  SD total daily energy expenditure during the course was  $4697 \pm 682$  kcal/d, while mean energy intake was  $4610 \pm 8$  kcal/d and  $1541 \pm 211$  kcal/d from fresh and field rations, respectively. Energy deficit was 18%, which led to a mean  $6.7 \pm 3.8$  % reduction from PRE body mass. Daily sleep ranged from 2.3–4.0 h/d. Handgrip strength declined by  $6.2 \pm 13.9$  % ( $p < 0.05$ ) and mean standing broad jump distance declined by  $12.6 \pm 6.6$  % ( $p < 0.01$ ). A positive association was observed between the lean mass and handgrip strength at PRE ( $r = 0.50$ ,  $p = 0.01$ ,  $N = 30$ ) and POST ( $r = 0.55$ ,  $p = 0.02$ ,  $N = 17$ ). The combined lean mass in both legs was positively correlated with standing broad jump distance at PRE ( $r = 0.55$ ,  $p < 0.01$ ,  $N = 29$ ) but not POST ( $r = 0.48$ ,  $p = 0.20$ ,  $N = 9$ ). Mean Simple Reaction Test (SRT) reaction time increased

by  $11.1 \pm 7.2\%$  ( $p = 0.001$ ) and the mean number of lapses increased from  $0.6 \pm 1.2$  to  $2.4 \pm 2.2$  ( $p < 0.05$ ). Although mean Number Pair Test (NPT) reaction time did not change ( $p > 0.05$ ), the mean percentage correct responses fell from  $96 \pm 2\%$  to  $93 \pm 4\%$  ( $p < 0.01$ ). An association was observed between the amount of daily sleep over Phase 2 and 3 and the number of lapses at POST assessed by SRT ( $p < 0.01$ ) and the percentage of correct responses at POST assessed by NPT ( $p < 0.05$ ). In summary, upper body strength, lower body explosive power, vigilance and accuracy in decision making were affected under a multi-stressor environment. Future work should investigate protein supplementation to preserve physical performance.

Metabolic and performance effects of *Ilex paraguariensis* (Yerba mate) during submaximal exercise and laboratory-based time trials in well-trained cyclists.

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*Ilex paraguariensis* (IP) is a plant rich in polyphenols, saponins and xanthines consumed in beverages by millions of people world-wide, but its metabolic effects remain unknown. Traditionally IP has been considered a stimulant and recent research suggests that it increases fat oxidation during exercise. Our aim was to test the effect of IP on substrate utilisation during submaximal exercise and endurance performance in athletes. Eleven well-trained male cyclists (Age, mean $\pm$ SD 30 $\pm$ 3 y; Weight, 75 $\pm$ 7 kg;  $\dot{V}O_{2max}$  71 $\pm$ 6 ml/kg/min; PPO 403 $\pm$ 32 W) participated in a double-blind counterbalanced crossover study design. Participants ingested 5 g of IP or placebo (PL; maltodextrin) daily for 5 days, and 1 h prior to an experimental trial. Prior to the study, IP samples were characterized phytochemically. Experimental trials were conducted in the morning, with fasted participants, after 48 h of exercise control and 24 h of dietary provision (8 g CHO/kg). Tests included a submaximal step-test (SST) at 30,40,50,60,70 and 80% of  $\dot{V}O_{2max}$  for 5 min per stage, followed by 10 min recovery and an ergometer-based time-trial (TT; n=9) to complete a predetermined amount of mechanical work and last ~30 min. During SST and selected TT time-points, respiratory gases were collected and analysed through an automated gas analysis system. Venous blood samples were taken prior to ingestion of treatment, pre-SST, post-SST and every 1/3 of TT. Plasma was analysed for free fatty acids (FFA), glycerol, glucose and lactate. Results show a small (ES=.38) but significant ( $p < 0.05$ ) improvement of IP in time to complete TT from 30.1 $\pm$ 1.8 to 29.4 $\pm$ 1.4 min, and a concomitant small increase (2.3%) in power output (ES=.2;  $p < 0.05$ ). Blood parameters remained unchanged at rest. [FFA] was lower from rest and IP in PL pre-SST ( $p < 0.05$ ). [Glycerol] increased above rest at all time-points after SST in both treatments ( $p < 0.05$ ) and in IP vs PL post-SST and at 1/3 TT ( $p < 0.05$ ). [Lactate] increased above

rest at all time-points post-SST ( $p < 0.001$ ) and was higher in IP vs PL at 2/3 and 3/3 TT ( $p < 0.02$ ). Respiratory gas analyses show lower fat oxidation in PL vs IP at 30, 40, 50 and 60% of  $\dot{V}O_{2max}$  ( $p < 0.05$ ) but no differences during TT. In conclusion, there was a small but significant increase in performance with IP and markers of fat utilization were lower in PL during SST compared to IP.

#### Effect of New Zealand blackcurrant extract on performance during repeated 4 km cycling time trials

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New Zealand blackcurrant (NZBC) extract increased 16.1 km cycling time trial performance. It was hypothesized that NZBC extract would enhance repeated 4 km time trial performance. Ten males (age: 30 $\pm$ 12 y, body mass: 74 $\pm$ 9 kg, height: 179 $\pm$ 7 cm, body fat: 11 $\pm$ 3%,  $\dot{V}O_{2max}$ : 55 $\pm$ 7 ml $\cdot$ kg<sup>-1</sup> $\cdot$ min<sup>-1</sup>, mean $\pm$ SD) volunteered. In the first visit, participants completed a cycling test to exhaustion to establish  $\dot{V}O_{2max}$  and were then familiarized with the time trials. Prior to visits two and three, participants consumed capsulated NZBC extract (300 mg $\cdot$ day<sup>-1</sup> CurraNZ<sup>TM</sup>; containing 105 mg anthocyanin) or placebo (P) (300 mg $\cdot$ day<sup>-1</sup> microcrystalline cellulose M102) for 7 days (double blind, randomised, cross-over design, wash-out at least 7 days). Participants performed a warm-up protocol by the British Cycling Federation, before 2 x 4 km time trials (10 min active self-paced recovery between trials) with personalized similar gear settings (SRM ergometer, SRM International, Germany) and distance feedback. Heart rate was recorded and blood lactate sampled immediately after each trial and 8 min into recovery between the trials. Times over comparable one km distances in each 4 km time trial were similar. No effect was observed for the time to complete the first (PL: 380 $\pm$ 28 s, NZBC: 377 $\pm$ 27 s) and second 4 km of cycling (PL: 391 $\pm$ 32 s, NZBC: 387 $\pm$ 30 s), with times for the second 4 km of cycling slower by 11 $\pm$ 8 s and 11 $\pm$ 9 for placebo and NZBC, respectively. However, the total time of the two 4 km cycling trials was 0.8% faster with NZBC extract (PL: 771 $\pm$ 60 s, NZBC 764 $\pm$ 56 s,  $P = 0.034$ , one-tailed paired t-test) with 7 participants having faster total times. There was no effect of NZBC extract on the mean power for completion of the first (PL: 390 $\pm$ 66 W, NZBC: 390 $\pm$ 69 W) and second 4 km of cycling (PL: 357 $\pm$ 72 W, NZBC 365 $\pm$ 71), with slower mean power values for the second 4 km of cycling in both groups. There was a trend for mean power to be 7 Watts higher over the two 4 km cycling trials (PL: 747 $\pm$ 138 W, NZBC 754 $\pm$ 140 W,  $P = 0.098$ , one-tailed paired t-test) with 7 participants showing higher mean power. There was no effect of NZBC on heart rate and lactate values at identical time points. It is concluded that New Zealand blackcurrant extract (CurraNZ<sup>TM</sup>) seems to be somewhat beneficial in repeated high-intensity cycling time trials for overall performance. Acknowledgement: We thank Health Currancy Ltd (UK) for CurraNZ<sup>TM</sup> supplementation and support for conference attendance.

**Six-weeks of flaxseed oil supplementation does not alter substrate oxidation or energy expenditure during a simulated 16-km fell run**

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This double blind, randomised study investigated the effects of a six-week, omega-3 rich flaxseed oil supplementation on substrate oxidation and energy expenditure during a simulated 16-km fell run. Twenty well trained male runners (age:  $33 \pm 8$  years,  $\text{VO}_2\text{peak}$ :  $60.5 \pm 8.5$  ml.kg<sup>-1</sup>.min<sup>-1</sup>) were randomly assigned to either a flaxseed (FLAX) or placebo (PLA) supplementation group. On a daily basis, FLAX consumed 15ml/25kg.BM<sup>-1</sup> flaxseed oil mixed with a fruit beverage whereas PLA consumed an identical fruit beverage without the addition of flaxseed oil. A standardised 16-km simulated

fell run was completed on a motorised treadmill prior to and following the six week supplementation period. Online gas analysis was conducted throughout exercise to assess trial and time based differences in substrate oxidation and energy expenditure. The proportional reliance on carbohydrate and fat as a fuel was similar between groups before supplementation ( $P = 0.63$ ,  $ES = 0.23$ ). There were no significant differences in proportional carbohydrate reliance between pre and post supplementation in either group (FLAX:  $78.8 \pm 7.7$  % vs  $76.1 \pm 11.0$ ,  $P = 0.42$ ,  $ES = 0.28$ ; PLA:  $76.5 \pm 12.0$  % vs  $75.4 \pm 11.8$  %,  $P = 0.85$ ,  $ES = 0.09$ ). Furthermore, energy expenditure was similar between groups prior to supplementation ( $P = 0.486$ ;  $ES = 0.36$ ). There were also no significant differences in energy expenditure between pre and post supplementation in either group (FLAX:  $1318 \pm 78$  vs  $1260 \pm 77$  kcal,  $P = 0.141$ ,  $ES = 0.75$ ; PLA:  $1275 \pm 152$  vs  $1246 \pm 99$  kcal,  $P = 0.232$ ,  $ES = 0.23$ ). Six-weeks of omega-3 rich flaxseed oil supplementation does not alter substrate oxidation or energy expenditure during a 16-km simulated fell run in well trained male runners.

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Rice gel as a carbohydrate source during endurance exercise.

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Carbohydrates in solution or solid are generally used as energy supply to increase endurance exercise performance. But there are few studies in relation to foods whose physical properties are between solution and solid, which athletes can chew or swallow without chewing. We made an original rice product (Rice gel) and investigated its effects on thirsty, hunger, palatability, and exercise performance. The main ingredient of rice gel was rice plant containing 100% amylopectin. Six male university students cycled at  $65\% \pm 2\%$  VO<sub>2</sub>max for 60 min plus time trial (400 kJ) while receiving one of the following two treatments in randomized order: rice gel (Gel) or carbohydrate drink containing soft jelly (Jelly). The Jelly and Gel was taken three times (80 kcal each) at 0, 30 min of exercise, and immediately after time trial. The VAS score of thirst and hunger were significantly suppressed in Gel compared to Jelly ( $P < 0.05$ ). The VAS score also revealed that there observed no difficulties in swallowing Gel compared to Jelly even immediately after time trial. Neither of them showed stomach upset or like that. There was no significant difference in blood sugar level, Vo<sub>2</sub>, respiratory exchange ratio, lactic acid level, heart rate and time trial score (7'51" and 7'42' in Gel and Jelly, respectively). In conclusion, rice gel could be a supplementary meal which is not stuck in the throat during the endurance exercise of moderate strength, and suppressed the feelings of thirst and hunger during exercise. This work was supported by Sugiyama Sangyo Kagaku with the approval of the Human Test Ethics Committee of Ryukoku University.