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The time-course of creatine mediated augmentation of skeletal muscle glycogen storage following exhaustive exercise in man

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Muscle glycogen availability is a determinant of endurance exercise performance and its depletion results in fatigue. We have demonstrated that 5 days of creatine (Cre; 20g/day) and simple carbohydrate (CHO) ingestion augmented muscle glycogen storage following exhaustive exercise in man when compared to CHO feeding alone (Robinson *et al.* 1999). This study characterises the time-course of this Cre-induced glycogen super-compensatory response, and points to mechanisms underpinning this phenomenon.

Table 1 – Muscle glycogen & total-creatine concentrations (mmol kg dry muscle^{-1}) & area under the serum insulin curve (mU/l/min) in exercised legs before (Exhaustion) and after 1, 3 and 6 days of Creatine+CHO or Placebo+CHO supplementation.

	Treatment	Exhaustion	1 Day	3 Days	6 Days
Glycogen	Placebo	69.9 ± 22.2	294.8 ± 39.4*	512.1 ± 38.6*	788.7 ± 38.6*
	Creatine	77.2 ± 19.2	486.6 ± 35.4*†	620.4 ± 49.3*	944.8 ± 76.7*†
Total Creatine	Placebo	127.9 ± 4.5	128.0 ± 4.3	130.9 ± 3.8	129.0 ± 5.3
	Creatine	127.1 ± 5.0	138.1 ± 1.8*†	144.8 ± 2.5*†	157.9 ± 2.7*†
Area Under Insulin Curve	Placebo	4070 ± 813	4310 ± 642	4858 ± 912	4070 ± 797
	Creatine	4361 ± 962	5995 ± 733	6491 ± 1706	6226 ± 1508

**Different ($P < 0.05$) from pre-treatment (Exhaustion) time-point within group, † different ($P < 0.05$) between groups at corresponding time-point.*

Fourteen recreationally-trained males (age 26.4 ± 2.0 years, BMI 24.5 ± 1.0, $\dot{V}_{O_2,max}$ 44.4 ± 1.5 ml kg^{-1} min^{-1}) participated in this study, which was approved by the local Ethics Committee. Subjects reported fasted to the laboratory, whereupon they underwent a 2 h oral-glucose tolerance test (GTT, 90 g CHO; blood sampling every 15 min for serum insulin analysis). Following GTT, subjects exercised at 70 % $\dot{V}_{O_2,max}$ on a cycle ergometer until exhaustion. Following exercise, a muscle biopsy sample was obtained from the subject's non-dominant leg (Exhaustion). Subjects were then randomly assigned to a Cre ($n = 7$) or glycine (placebo, $n = 7$) treatment group and ingested 5 g of Cre or 5 g of glycine dissolved in 250 ml of a warm sugar-free solution followed by 500 ml of a CHO-containing solution (90 g simple sugars). Subjects continued to ingest these solutions, at equally spaced intervals, on 4 occasions per day over 6 days. A high CHO diet (37.5 kcal kg^{-1} day-1, >80 % calorific intake CHO) was provided and ingested during the supplementation period. Subjects reported fasted to the laboratory on days 1, 3 and 6 of supplementation for further GTTs and muscle biopsy sampling from their non-dominant leg. Muscle samples were immediately frozen in liquid nitrogen, freeze dried and used for subsequent biochemical analysis (Table 1). Data are expressed as means ± S.E.M. Statistical analysis was performed using two-way ANOVA with LSD post-hoc analysis.

The present study unequivocally establishes Cre's glycogen super-compensatory properties and shows that this marked response occurs within 24-hours post-exercise, during which the muscle total-Cre stores had increased < 10 %. Furthermore, Cre augments muscular glycogen stores independent of any clear increase in serum insulin AUC (area under the curve) during 6 days of supplementation, which points towards some other mechanism as being causative.

Robinson TM *et al.* (1999). *J Appl Physiol* **87**, 598–604.

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All procedures accord with local guidelines and the Declaration of Helsinki

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Temporal relationship between human muscle protein synthesis, protein kinase B (PKB) and p70^{S6} kinase (p70^{S6k}) phosphorylation after shortening or stretching exercise

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We have examined the relationship between the post-exercise stimulation of muscle protein synthesis and the activities of p70^{S6k} and PKB, members of the anabolic signalling pathway.

Eight healthy males (25 ± 5 y, body mass index, 26 ± 3 kg.m^{-2} mean ± S.E.M.) were studied. Carrying 25 % body weight, the subjects stepped up onto a knee-high box with one leg and stepped down with the other at ~1 Hz for repeated bouts of 6, 3 and 3 min with 2 min rest between. Quadriceps muscle biopsies were taken from both legs (using 1 % lignocaine anaesthesia) before exercise and 3, 6 and 24 h post exercise. In the 2 h before each biopsy, subjects were fed intermittently 45 g essential amino acids and 135 g sucrose. We infused [1-^{13}C]leucine (prime 1.0 mg kg^{-1}; 0.8 mg kg^{-1} h^{-1}) from rest to 6 h post-exercise and [1^{13}C]valine (prime 1.2 mg kg^{-1}; 1.0 mg kg^{-1} h^{-1}) between 21–24 h post exercise. Rates of myofibrillar and sarcoplasmic protein synthesis were measured by standard techniques using gas chromatography-combustion-isotope ratio mass spectrometry. Phosphorylation (activation) of p70^{S6k} and PKB was quantified by Western blotting.

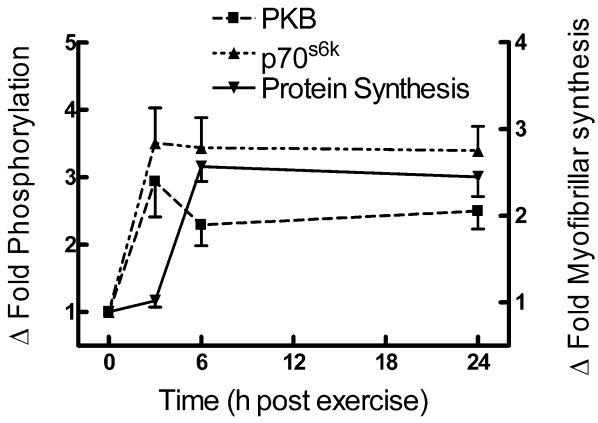

Figure 1. Changes in muscle PKB and p70^{S6k} phosphorylation and myofibrillar protein synthesis after exercise (mean ± S.E.M.) . All changes $P < 0.05$ vs. rest values except protein synthesis at 3 h. Lengthening and shortening contractions have been meaned together as there was no significant diffrerence between them.

With this protocol there were no differences observed in any of the measured variables after lengthening or shortening contractions. PKB phosphorylation (Serine 473) rose to a maximum value (3.2 fold above baseline) at 3 h post exercise but was still significantly elevated at 24 h. However, phosphorylation of p70^{S6k} (basal 23 % phosphorylation), increased to a constant value of ~3.5 fold above basal between 3 and 24 h. Muscle

protein synthesis rose rapidly between 3 and 6 h and remained elevated for 24 h (basal myofibrillar and sarcoplasmic synthesis rates 0.042 ± 0.012 % h^{-1} and 0.061 ± 0.007 % h^{-1} respectively), the relative increase being 62 % greater for myofibrillar than sarcoplasmic protein at 24 h. The results are consistent with a sequence of events in which muscle contraction causes a sustained activation of PKB and $p70^{S6k}$ with a subsequent prolonged stimulation of protein synthesis. This is in contrast to the transient rise in PKB and $p70^{S6k}$ activity seen after acute exercise in the rat (Bolster *et al.* 2003).

Bolster DR *et al.* (2003). *J Physiol* **553**, 213–220.

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All procedures accord with current local guidelines and the Declaration of Helsinki

C61

Sympatho-adrenal responses to repeated brief maximal exercise in man: influence of training type

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The sympatho-adrenal responses to physical exercise following long-term training are unclear. Previous work has demonstrated similar noradrenaline but greater adrenaline responses to high intensity exercise in endurance-trained compared to untrained subjects (Kjaer & Galbo 1988). Other research has reported no differences in sympatho-adrenal responses following maximal exercise between endurance-trained and untrained subjects but found a greater adrenaline response in sprint-trained compared to both endurance-trained and untrained subjects (Zouhal *et al.* 2001). Therefore, the aim of this study was to investigate the sympatho-adrenal responses of untrained, sprint- and endurance-trained subjects performing repeated brief maximal cycle sprints.

With local ethics committee approval and informed consent sprint-trained (ST $n = 7$), endurance-trained (ET $n = 8$) and untrained (UT $n = 10$) subjects participated in this study. Physical and physiological characteristics of ST, ET and UT were (mean ± S.D.); age 24 ± 7, 30 ± 10, 21 ± 1 years, body mass 73 ± 7, 68 ± 6, 76 ± 10 kg and $\dot{V}_{O_2max}$ 58 ± 3, 52 ± 5, 67 ± 5 ml kg^{-1} min^{-1}. Subjects performed ten 6 s maximal cycle sprints against a frictional load of 0.075 kg kg^{-1} body mass with 30 s rest between sprints. Power output was corrected for flywheel acceleration. Blood samples (10 ml) taken from an antecubital vein at rest, immediately following each sprint and 2 and 5 min post-exercise were first used to determine blood lactate, then analysed for plasma catecholamines (adrenaline; AD, noradrenaline; NA and dopamine; DA) by HPLC with electrochemical detection (Davies *et al.* 1981). Data were analysed using one-way and repeated measures ANOVA with Tukey *post-hoc* where appropriate. Significance was established at $P < 0.05$.

ET demonstrated greater resting NA concentration compared to UT (2.0 ± 0.4 *vs.* 1.3 ± 0.5 nmol l^{-1}, $P < 0.05$). Peak NA concentrations occurred immediately following Sprint 10 in all groups (ST 28.3 ± 7.0, ET 16.5 ± 6.2 and UT 17.9 ± 7.0 nmol l^{-1}) with ST demonstrating greater NA from Sprint 5 onwards compared to ET ($P < 0.05$) and Sprint 8 onwards compared to UT ($P < 0.05$). There was an 8 fold increase in AD concentration from rest following sprint 10 in ST but a 3–4 fold increase in ET and UT (ST 5.0 ± 1.7, ET 2.7 ± 1.1 and UT 2.4 ± 1.0 nmol l^{-1}, $P < 0.05$). ST demonstrated greater AD following all sprints compared to ET and UT ($P < 0.05$) with no differences between ET and UT. A greater plasma DA concentration following sprint 9 was found in ST compared to UT (1.7 ± 0.9 *vs.* 0.8 ± 0.2 nmol l^{-1}, $P < 0.05$). Total work done over the ten sprints was greater in ST compared to ET and UT (ST 48195 ± 4897, ET 40597 ± 5890, UT 41706 ± 4131 J, $P < 0.05$). Blood lactate concentrations were greater 5 min post-exercise in ST (10.6 ± 1.7 mmol l^{-1}) and UT (10.6 ± 1.4 mmol l^{-1}) compared to ET (8.4 ± 0.9 mmol l^{-1}, $P < 0.05$).

The results of this investigation suggest that sprint but not endurance training may alter the magnitude of the plasma catecholamine response to repetitive brief maximal cycle exercise in man. Additionally, endurance training may alter the resting plasma NA concentration.

Davies CL *et al* (1981). *Ann Chem* **53**, 156–159.

Kjaer M & Galbo H (1988). *Am J Physiol* **254**, R197–R203.

Zouhal H *et al* (2001). *J Sp Med & Physical Fitness* **41,** 330–336.

All procedures accord with current local guidelines and the Declaration of Helsinki

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Myofibrillar protein synthesis in human quadriceps muscle rises more quickly after maximal lengthening than maximal shortening contractions

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We aimed to determine if there was any difference in the extent and time course of myofibrillar protein synthesis (MPS) after maximal muscle shortening (SC, average peak torque = 225 ± 7 N.m, means ± S.E.M.) or lengthening contractions (LC, average peak torque = 299 ± 18 N.m) with equivalent work performed in each mode. Eight healthy young men (21.9 ± 0.6 y, BMI 24.9 ± 1.3 $kg.m^{-2}$) performed 6 sets of 10 maximal unilateral LC of the knee extensors on an isokinetic dynamometer. Then, with the contralateral leg they performed 6 sets of maximal unilateral SC with work matched to the total work performed during LC (10.9 ± 0.7 *vs.* 10.9 ± 0.8 kJ, $P = 0.83$). Whole body oxygen consumption (VO_2) was recorded during both LC and SC to indicate oxidative ATP production. On a separate day the subjects were studied at rest; on both rest and exercise days the subjects were fed small intermittent meals of Myoplex Lite^{R} (EAS^{TM}) to provide 0.1 g kg^{-1} h^{-1} of protein and 0.1 g kg^{-1} h^{-1} of carbohydrate. Quadriceps muscle biopsies were taken (2 % xylocaine anaesthesia) using the Bergstrom technique at rest and at 4.5 h and 8.5 h post-exercise. To determine protein synthesis we infused [1, 2-^{13}C] leucine (prime 1.0 mg kg^{-1}; 1.0 mg kg^{-1} h^{-1}) and measured incorporation into myofibrillar protein by gas chromatography-combustion mass spectrometry using our standard methods.

Prior exercise elevated MPS above rest (0.067 ± 0.003 % h^{-1}, $P < 0.01$) in both LC and SC, but was greater in LC than SC at 4.5 h (Fig. 1). At 8.5 h post-exercise, MPS remained elevated above rest but was not different between legs. VO_2 was greater ($P < 0.01$) with SC (14.0 ± 1.4 l) as compared to LC (10.2 ± 0.6 l). Despite matching the total work performed in both the LC and SC protocols, maximal LC were accompanied by a more rapid rise in the post-exercise synthesis of myofibrillar protein than SC. This observation may be related to the lower energy cost of performing LC *versus*SC or some other as yet

unknown factor.

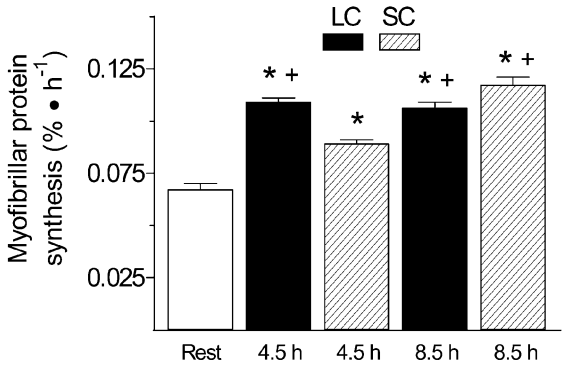

Figure 1. Muscle myofibrillar protein synthesis following maximal lengthening (LC) and shortening contractions (SC). * Significantly different from rest (P < 0.01). + Significantly different from SC at 4.5 h (P < 0.01).

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All procedures accord with current local guidelines and the Declaration of Helsinki

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The effects of carbohydrate ingestion on muscle glycogen utilisation during exhaustive high-intensity intermittent running

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The ingestion of carbohydrate-electrolyte (CHO-E) solutions has been shown to improve capacity during high-intensity intermittent running (LIST) (Nicholas *et al.* 1995). Nicholas *et al.* (1999) reported that this was due to a sparing of muscle glycogen. Initial muscle glycogen concentrations in that study were moderate (approx 350 mmol kg^{-1} dry matter), therefore the purpose of the present study was to examine the effect of CHO ingestion on muscle glycogen utilisation and intermittent running capacity on subjects with high pre-exercise muscle glycogen concentrations.

Six men (mean ± (S.E.M.); age 23.0 years (± 0.4); body mass 75.0 kg (± 1.2); $\dot{V}_{O_2max}$ 60.0 ml kg^{-1} min^{-1} (± 0.7)) volunteered, with informed consent, to participate in this study. Subjects performed two trials separated by 14 days in a randomised crossover design, consuming either a 6.4 % hypotonic CHO-E solution (HYP) or a placebo (PLA) in a double-blind fashion prior to the trials (8ml kg^{-1}BM) and at 15 min intervals (3ml kg^{-1}BM) until fatigue. Following a glycogen depleting trial and 48 h high CHO diet (10g kg^{-1}BM day^{-1}) subjects ran LIST to fatigue. Muscle biopsy samples were obtained pre-LIST, 90 min and fatigue.

All subjects ran longer in HYP (158.0 min ± 11.6) compared to PLA (131.0 min ± 8.0; $P = 0.03$; Wilcoxon). There were no differences in muscle glycogen concentrations pre-exercise (HYP 533 ± 31 mmol kg^{-1} DM v PLA 512 ± 41 mmol kg^{-1} DM) or at 90 min (HYP 344 ± 37 mmol kg^{-1} DM v PLA 359 ± 24 mmol kg^{-1} DM; paired Student's *t* test). There was a trend for muscle glycogen utilisation to be greater in PLA post 90 min (4.2 ± 1.2 mmol kg^{-1} DM min^{-1}) compared to HYP (2.5 ± 0.3 mmol kg^{-1} DM min^{-1}; $P = 0.1$) although this failed to reach statistical significance. Plasma glucose concentrations were higher at fatigue in HYP ($P < 0.001$; ANOVA) although hypoglycaemia did not occur in PLA.

These data suggest that the ingestion of a 6.4 % hypotonic CHO-E solution will improve endurance capacity during intermittent exercise (LIST) in subjects with elevated pre-exercise muscle glycogen concentrations. This appears to occur through higher plasma glucose concentrations which may result in elevated rates of exogenous CHO oxidation late in exercise as indicated by the tendency towards a lower rate of muscle glycogen utilisation.

Nicholas CW *et al.* (1995). *J Sports Sci* **13**, 283–290.
Nicholas CW *et al.* (1999). *MSSE* **31**, 1280–6.

All procedures accord with current local guidelines and the Declaration of Helsinki

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The CSF and arterial to internal jugular venous hormonal differences during exercise in humans

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During and especially after intense exercise the brain takes up carbohydrate in excess of O_2 (Dalsgaard *et al.* 2002). This study evaluated the arterial to internal jugular venous differences and the cerebrospinal fluid (CSF) levels of hormones that may influence carbohydrate metabolism.

After approval by the Ethics Committee of Copenhagen (KF 01–034/02) and written informed consent, nine healthy subjects of median age 26 years (range 23–28) performed about 12 min exhaustive exercise. The CSF was drained immediately post-exercise by a lumbar puncture, while CSF at rest was obtained from 6 other age matched subjects. Included hormones were noradrenaline (NA), adrenaline (A), insulin, insulin-like growth factor (IGF) I, and cortisol. Also determined were interleukin (IL) 6, tumour necrosis factor (TNF) α, as well as ammonium (NH_4^+).

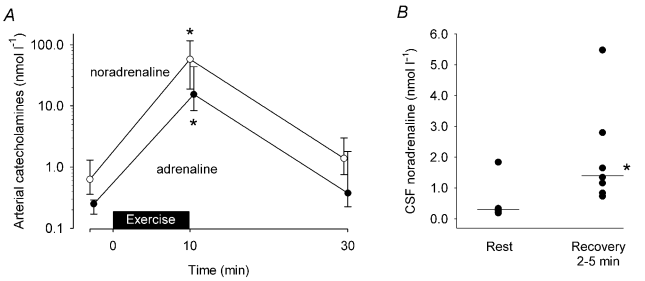

Figure 1. *A*, the median (25–75 percentile) arterial concentration of noradrenaline (NA) and adrenaline (A) at rest, at the point of exhaustion and 30 min into recovery ($n = 9$). *B*, CSF NA concentration immediately after exercise from the same subjects as in the left picture and at rest ($n = 6$); also given is the median. Rec, recovery. *, Different from rest, $P < 0.05$.

Exercise increased the arterial levels of NA and A (Fig. 1*A*), but there was no cerebral uptake. Yet, following exercise CSF NA was 1.4 (0.73 −5.5) nmol l^{-1} and higher than at rest, 0.3 (0.19 −1.84) nmol l^{-1} (Fig. 1*B*; $P < 0.05$), whereas A did not show. Arterial NH_4^+ increased with exercise and equally for the uptake of NH_4^+ from 1 (−12 − 5) to 17 (5 − 41) μmol l^{-1}. Conversely CSF NH_4^+ was reduced to 7 (0–10) *vs.* 11 (7 −16) μmol l^{-1} ($P < 0.05$). Plasma insulin was low while the brain took up a surplus of carbohydrate, but it increased to above resting levels after 30 min. Other arterial variables were elevated with exercise, but there was no cerebral uptake or changes in CSF concentrations.

The findings suggest that a maximal exercise bout is not associated with IL-6 or TNF-α accumulation, in or release from the brain. However, NA seems to play an intrinsic role for the cerebral response to exercise, and a cerebral uptake of NH_4^+ may be of value for amino acid metabolism within the brain. In contrast, blood borne catecholamines, cortisol, IGF-I are seemingly unimportant. Notably, during and immediately after exercise the brain appears to take up glucose independently of insulin.

Dalsgaard MK *et al.* (2002). *J Physiol* **540**, 681–9.

All procedures accord with current local guidelines and the Declaration of Helsinki

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Invasion of red blood cells by *Plasmodium falciparum* is strongly influenced by the volume or density of the target cells

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Invasion of human red blood cells (RBCs) by *P. falciparum* merozoites is a complex, multistage process. It has been known for some time that the condition of the targeted RBC may affect the efficiency of invasion and hence the severity of the disease. The work reported here arose from a casual observation which suggested that the volume of RBCs has a profound effect on parasite invasion.

To investigate this volume effect, invasion and growth parameters of *P. falciparum* parasites were compared in cultures sustained with normal-volume RBCs and with RBCs whose volume had been altered experimentally. The volume of target RBCs was modified isotonically, by transient activation of the RBC Ca^{2+}-sensitive K^+ channels (Gardos channels). Venous blood from healthy volunteers (blood group O) was drawn into heparinized syringes after informed, written consent. After washes, RBCs were suspended in isotonic Hepes-buffered salines containing 10 mM SCN^-, 50 μM Ca^{2+} and different K^+ concentrations, from 2 to 137 mM, and incubated at 37 °C. A uniform Ca^{2+} load was induced by addition of a high concentration of a Ca^{2+} ionophore (A23187). This activated the Gardos channels allowing rapid K^+ equilibration with cell shrinkage or swelling by net KCl loss or gain, respectively, depending on the set K^+ gradients. Addition to the suspension of EGTA in excess of Ca^{2+} extracted the cell Ca^{2+}. Subsequent washes with albumin removed the ionophore from the cells, thus restoring the normal low Ca^{2+} and K^+ permeabilities. *P. falciparum* parasites of the A4 clone were cultured in human RBCs and synchronized by standard methods. Schizont-infected RBCs were enriched to about 90 % parasitaemia, mixed with uninfected RBCs of different volumes to final parasitaemias of about 6 %, and cultured in standard conditions. Invasion and growth were followed by microscopic inspection of Giemsa stained thin smears after 6 h, and by ^{3}H-hypoxanthine incorporation after 24 h.

In one experiment, typical of five, ring-parasitaemias at 6 h in controls and in cells equilibrated at $[K^+]_o$ of 2, 45, 80 and 137 mM, were 10, 0, 4.8, 9.6 and 11.4 %, respectively. Invasion was similar in untreated controls and in K^+-equilibrated normal-volume RBCs indicating that brief exposures to ionophore and high internal Ca^{2+} had no effect on invasion. ^{3}H-hypoxanthine incorporation showed an identical trend in three experiments: maximal inhibition in profoundly dehydrated cells, and normal or elevated incorporation in swollen cells, with graded responses in between. Although the mechanism of this volume-effect is yet to be investigated, our results may help explain the selective advantage in malaria endemic regions of mutations which generate subpopulations of relatively dense RBCs in the circulation. The reduced infectivity of dehydrated RBCs would prevent the development of high parasitaemias and hence the incidence of severe malaria.

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Orthostatic tolerance and blood volume in high altitude dwellers

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Orthostatic tolerance (OT) is a measure of a person's ability to maintain consciousness and adequate blood pressure during gravitational stress. It is partly dependent on plasma volume (El-Sayed and Hainsworth, 1995) and on the magnitude of reflex vasoconstriction (Brown and Hainsworth, 2000). We postulated that it would also be influenced by red cell volume, and that chronically hypoxic high altitude dwellers with large red cell volumes, would also have high orthostatic tolerance.

We studied 22 male residents of Cerro de Pasco, Peru (4338m, PB 450 mmHg), including 11 with Chronic Mountain Sickness (CMS) characterised by haematocrits over 60 %. Plasma and blood volumes were determined by Evans blue dye dilution and peripheral haematocrit. Orthostatic tolerance was the time to presyncope in a test of head-up tilt (20 min), followed by tilt and lower body suction at −20, −40 and −60 mmHg for 10 min each. We recorded ECG, finger blood pressure (photo-plethysmography) and forearm blood velocity (Doppler). The study was approved by the local ethics committee. Data are presented as means ± S.E.M. Statistical significance was assessed using student *t* test, ANOVA and Pearson correlation.

Plasma volumes in normals and CMS (38.3±1.9 and 34.8±1.6ml kg^{-1}) were similar to those previously reported in low altitude dwellers (42.4±3.3ml kg^{-1}, El-Sayed and Hainsworth, 1995). Red cell volumes were higher in CMS than normals (71.7±7.3 and 45.3 ± 2.5ml kg^{-1}, $P < 0.01$) and both were higher than in lowlanders ($P < 0.05$). Blood volumes also were higher in CMS than in normals (106.5±8.3 and 83.6±4.0ml kg^{-1}, $P < 0.05$) and higher in both groups than in lowlanders ($P < 0.05$). Orthostatic tolerance was high in both groups. All tolerated the stress for

more than 40 min, compared with only 15 % of sea level dwellers (El-Bedawi and Hainsworth, 1994) and 9 of the 22 tolerated the entire procedure (50 min). Overall orthostatic tolerance was significantly correlated with red cell volume ($r = 0.54$; $P < 0.02$) and blood volume ($r = 0.48$; $P < 0.05$). The maximum change in forearm vascular resistance (pressure/velocity) was +110±20 and +88±14 % (NS) and similar to that reported in sea level dwellers (Brown and Hainsworth, 2000). All tests were repeated one day after descending to sea level. Oxygen saturation increased from 82.8±3.7 to 95.3±2.4 %, $P < 0.0001$. Plasma and red cell volumes, and orthostatic tolerance, however, were not significantly changed.

Although there are other differences between altitude and sea level dwellers, the high orthostatic tolerance seen in these subjects, which is not immediately altered by relief of hypoxia, is compatible with the view that red cell volume is a significant factor in orthostatic tolerance.

Brown CM & Hainsworth R (2000). *Clin Auton Res* **10**, 57–61.

El-Bedawi KM & Hainsworth R (1994). *Clin Auton Res* **4**, 41–47.

El-Sayed H & Hainsworth R (1995). *Clin Sci* **88**, 463–470.

All procedures accord with current local guidelines and the Declaration of Helsinki

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On-line conductivity monitoring demonstrates alterations of sodium flux: a randomised controlled trial of reduced dialysate sodium concentration

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Relatively low dialysate conductivity (D cond.) is desirable as it is associated with lower interdialytic weight gains, and improved BP control. Excess sodium removal can, however, lead to hemodynamic instability. We performed a randomised controlled trial of empirical reduction of dialysate conductivity.

This study was approved by the Local Regional ethics Committee, and all patients gave written informed consent. Sodium removal as ionic mass balance (IMB), as well as plasma conductivity, was measured by conductivity monitoring, and routine clinical measurements were used to assess the clinical impact. 28 patients were recruited, and randomised to either maintenance of D cond at 13.6 mS/cm (equivalent to 140 mmol/l of Na^+), or serial reduction of D cond in steps of 0.2 mS/cm. Reduction was guided by symptoms and BP. Of the 16 patients randomized to reduction of D cond., 6 achieved D cond 13.4 mS/cm, 6 achieved 13.2 mS/cm, and 4 achieved 13.0 mS/cm (13.0 mS/cm was pre-specified as the lowest acceptable D cond). No episodes of dysequilibrium occurred. Results are expressed as those achieved at minimum D cond once this was established, compared with baseline at 13.6 mS/cm (shown as mean ± S.E.M., significance calculated using paired *t* test).

Interdialytic weight gain was reduced from 2.34 ± 0.10 kg to 1.57 ± 0.11 kg $P < 0.0001$. Both pre and post dialysis BPs were significantly reduced (pre-dialysis systolic BP fell from 144 ± 3 mmHg to 137 ± 4 mmHg $P < 0.05$). The reduction in convective sodium removal due to reduced weight gains was matched by an increase in the amount of sodium removed by diffusion (91 ± 12 mmol cf. 158 ± 12 mmol $P < 0.0001$). Finally pre-dialysis plasma conductivity also fell, from 14.23 ± 0.04 mS/cm to 14.02 ± 0.05 mS/cm, suggesting a significant reduction in the overall sodium load, given that the target weight did not change.

In summary, we have demonstrated that reduction in D cond monitored by IMB is safe and practical, and leads to improvement in interdialytic weight gains, and BP control while avoiding excessive sodium removal.

All procedures accord with current local guidelines and the Declaration of Helsinki

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Hypercapnic acidosis increases tolerance to orthostatic stress in humans

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Baroreceptor and chemoreceptor reflexes exert considerable influence on autonomic cardiovascular regulation, although limited information on interactions between these reflexes exists for humans (Somers *et al.* 1991). During baroreceptor stimulation using a simulated orthostatic challenge to pre-syncope, the influence of hypercapnic acidosis has not been previously studied. Therefore, this study investigated the effect of hypercapnic acidosis on tolerance to orthostatic stress in humans.

With DeMontfort University ethical approval, nine subjects (5 males and 4 females; mean ± S.D., age 21.9 ± 0.9 years, height 172.4 ± 9.7 cm, mass 70.3 ± 7.1 kg) were exposed to lower body negative pressure (LBNP) until the onset of pre-syncope on two occasions, each separated by approximately one week. In a counterbalanced design, investigations were carried out while subjects were breathing either room air (RA), or normoxic air containing 5 % carbon dioxide (CO_2). While breathing the appropriate mixture, a period of 15 min supine rest was followed by LBNP at −20 mmHg for 3 min, and subsequent decreases in pressure of −10 mmHg every 3 min until pre-syncope, at which time the test was terminated. Measurement of cardiovascular variables and continuous expired gas analysis were carried out on both occasions.

For both mixtures, LBNP induced a decrease in forearm blood flow, estimated cardiac output and stroke volume (all $P < 0.05$, ANOVA with Fisher post hoc). Minute ventilation, end tidal CO_2, and estimated arterial P_{CO_2} were elevated when breathing CO_2 compared to RA during supine rest and LBNP ($P < 0.001$, ANOVA with Fisher post hoc). Compared to RA (using Student paired *t* test, values are mean ± S.D.), CO_2 increased orthostatic tolerance (191.9 ± 20.4 *vs.* 210.3 ± 20.9 mmHg min), peak heart rate (123.9 ± 23.7 *vs.* 131.9 ± 24.9 beats min^{-1}), and time to peak heart rate (16.1 ± 2.1 *vs.* 18.1 ± 1.8 min) during LBNP. At pre-syncope, estimated stroke volume was 37.3 ± 2.1 ml when breathing RA, and 29.0 ± 6.7 ml when breathing CO_2 ($P > 0.05$).

These results suggest that the possible protective element of pre-syncope was delayed during hypercapnic acidosis at the expense of further reductions in stroke volume. This delayed pre-syncope response may have been associated with increases in cerebral blood flow induced by the increased arterial P_{CO_2}.

Somers V *et al.* (1991). *J Clin Invest* **87**, 1953–1957.

All procedures accord with current local guidelines and the Declaration of Helsinki

C111

Exercise-induced expression of haem oxygenase-1 in human lymphocytes

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Haem oxygenase-1 (HO-1) is an anti-inflammatory and cyto-protective enzyme that is activated by various forms of oxidative stress in a variety of mammalian cells (Applegate *et al.* 1991). In view of the functional significance of the inducible response (Wagener *et al.* 2003), HO-1 gene expression is a potentially valuable parameter to exploit as a biomarker of oxidative stress. Given the importance of lymphocytes in immune function and various diseases, we examined whether there was an increase in HO-1 mRNA accumulation in lymphocytes following an acute bout of exercise.

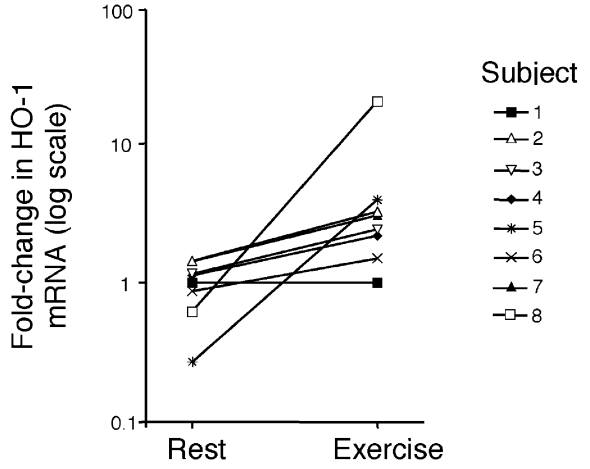

Figure 1. The peak exercise-induced fold-change in HO-1 mRNA from pre-exercise values is presented for each subject and expressed graphically against the fold-change from baseline at the corresponding time point in the rest trial. The peak fold-change in HO-1 mRNA following exercise was significantly greater than corresponding values in the rest trial ($P<0.05$). Data were analysed using the Wilcoxon signed-rank test.

Eight male subjects volunteered to take part in the present study, which had local Ethics Committee approval. Mean (± S.E.M.) age, height, body mass and maximal oxygen uptake were 21 ± 1 years, 179 ± 1 cm, 73.9 ± 2.4 kg and 64.4 ± 2.4 ml kg^{-1} min^{-1}, respectively. Subjects performed an exercise and a rest trial in a randomised order at least 10 days apart. In the exercise trial subjects ran on a level treadmill for 75 min at a speed corresponding to 70 % maximal oxygen uptake, and in the resting trial subjects sat calmly in the laboratory for an equivalent period of time. Lymphocytes were harvested from blood samples taken before and after each trial. Total RNA was isolated and used to determine the fold-change in HO-1 mRNA relative to baseline values using real time reverse transcription-polymerase chain reaction (LightCycler, Roche, Switzerland, UK). HO-1 was normalised to glyceraldehyde-3-phosphate dehydrogenase (GAPDH).

Six of the eight subjects showed an increase in HO-1 mRNA greater than two-fold after exercise (Fig. 1). However, there was considerable variation in terms of the magnitude of this response and the time of peak HO-1 mRNA accumulation. One subject showed a particularly pronounced response with a 20-fold increase above baseline in HO-1 mRNA 24 h post-exercise. HO-1 mRNA in the rest trial did not change over the period of investigation.

These results show that an acute bout of exercise leads to an increase in HO-1 mRNA accumulation in lymphocytes.

Applegate L *et al.* (1991). *Cancer Res* **51**, 974–978.

Wagener F *et al.* (2003). *Pharmacol Rev* **55**, 551–571.

All procedures accord with current local guidelines and the Declaration of Helsinki

C112

The effect of exercise on the expression and function of human monocyte toll-like receptors

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Exercise can have both a positive and negative effect on the immune system. Previous studies have shown that exercise can induce changes in many cytokines and signal molecules but the mechanism of action for these changes is not clear. Toll-like receptors (TLRs) play a crucial role in the detection of microbial pathogens by recognising products of microbial metabolism such as lipoproteins (TLRs 1, 2 and 6), zymosan (TLR2), lipopolysaccharide (LPS)(TLR4), and unmethylated CpG DNA motifs (TLR9) (Medzhitov, 2001). Following recognition of their specific ligand, TLRs expressed by antigen-presenting cells regulate the production of several cytokines including interleukin (IL)-6, IL-8 and tumour necrosis factor (TNF)-α, as well as the expression of accessory signal molecules CD80, CD86 and IL-12, that are required for the activation of naive T lymphocytes and the subsequent induction of the immune response. The effect of exercise or other forms of stress on TLRs is not well characterised.

The purpose of the present study was to examine the effects of exercise on the regulation of TLR expression and function. With local Ethics committee approval, venous blood samples were obtained from 11 healthy trained male cyclists (age 25 ± 1 years; body mass 74 ± 2 kg; maximal oxygen uptake, $\dot{V}_{O_2max}$ 4.7 ± 0.2 l min^{-1}; mean ± S.E.M.) at rest, immediately after, and following 2 h of resting recovery from 90 min cycling at 65 % $\dot{V}_{O_2max}$ in the heat (34°C). The expression of TLRs 1, 2, 4 and 9 on monocytes was assessed by flow cytometry. Monocyte intracellular cytokine (TNF-α and IL-6) production, the expression of CD80, CD86 and MHCII was also assessed following stimulation with LPS, zymosan and poly (I:C). Data were analysed using one-way ANOVA and post hoc Tukey tests where appropriate.

Table 1. Effects of exercise on TLR expression (MFI in arbitrary units) and LPS-stimulated up-regulation of CD80, CD86, MHCII and IL-6 in monocytes

	Rest	Post-exercise	2 h Post-exercise
TLR1 expression	5.0 ± 0.9	1.8 ± 0.5*	2.4 ± 0.5*
TLR2 expression	50 ± 4	27 ± 3*	23 ± 3*
TLR4 expression	6.9 ± 1.1	3.4 ± 0.7*	3.5 ± 0.7*
TLR9 expression	680 ± 99	737 ± 85	665 ± 79
CD80 expression	330 ± 35	305 ± 30	310 ± 35
CD86 expression	236 ± 21	202 ± 16*	187 ± 12*
MHCII expression	334 ± 50	255 ± 24*	234 ± 28*
IL-6 expression	146 ± 21	121 ± 16*	95 ± 11*

Values are means ± S.E.M. (n=11). *$P < 0.05$ *vs* Rest

Following exercise, monocyte expression of TLRs 1, 2 and 4 (but not TLR9) was substantially decreased ($P < 0.05$) with little or no

recovery by 2 h post-exercise (Table 1). Furthermore, the LPS-stimulated induction of CD86 and MHCII expression on monocytes was significantly lower in samples obtained following exercise compared with pre-exercise ($P < 0.05$). Similar results were obtained with zymosan and poly (I:C). LPS-stimulated monocyte IL-6 production was significantly reduced after exercise ($P < 0.05$).

These results indicate that monocyte TLR expression is reduced following an acute bout of prolonged exercise and that this is associated with decreased induction of co-stimulatory molecules and cytokines following stimulation with TLR ligands. These effects may represent a mechanism through which exercise stress impairs immune function and increases susceptibility to infection.

Medzhitov R (2001). *Nat Rev Immunol* **1**, 135–145.

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All procedures accord with current local guidelines and the Declaration of Helsinki

C113

Effects of carbohydrate supplementation during the first exercise bout on blood neutrophil responses to a second bout of prolonged cycling

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Prolonged exercise temporarily alters the number and functions of circulating neutrophils and the changes may last for several h after exercise. When two bouts of exercise are performed on the same day, the observed changes are even greater (Boyum *et al.* 2002). The aim of the present study was to compare the effect of carbohydrate supplementation during the first exercise bout (EX1) on neutrophil responses to a second bout (EX2) of prolonged cycling.

With the approval of Ethics Committee of Loughborough University, eight males (age 29.6 ± 1.5 years, body mass 73.1 ± 1.7 kg, $\dot{V}_{O_2max}$ 48.7 ± 2.1 ml kg^{-1} min^{-1}; means ± S.E.M.) performed two bouts of 90 min cycling (EX1 started at 09:00 and EX2 started at 13:30) at 60 % $\dot{V}_{O_2max}$ after an overnight fast on two occasions, separated by at least 4 days in a counterbalanced order. Subjects consumed 500 ml of a carbohydrate (10 % w/v glucose; CHO) or placebo (PLA) beverage at 5 min pre-exercise (500 ml), and 250 ml every 20 min during the first exercise bout. Water ingestion was allowed *ad libitum* after the first exercise bout. Blood samples were collected by venepuncture at pre-exercise and post-exercise for both bouts. Haematological analysis was performed using an automated cell counter. Plasma hormones concentrations were determined using ELISA kits. Phorbol myristate acetate (PMA)-induced respiratory burst was measured using a chemiluminescence (CL) assay (Knight Scientific Limited, Plymouth) and lipopolysaccharide (LPS)-stimulated neutrophil degranuation was measured as described by Robson *et al.* (1999). Results were analysed using a two-factor (trial × time) repeated measures ANOVA with post hoc Tukey tests and paired *t* tests applied where appropriate.

The ingestion of CHO during EX1 significantly attenuated the increase in circulating neutrophil number and the decrease in LPS-stimulated elastase release and PMA-induced CL per neutrophil in the recovery period after EX1 (Table 1). On PLA, the plasma glucose concentration was significantly lower ($P \leqslant 0.01$) and the adrenaline and cortisol concentrations were significantly higher ($P \leqslant 0.05$ and $P \leqslant 0.01$, respectively) at post-EX2 compared with CHO though neutrophil number and function were not significantly influenced.

Table 1. The plasma glucose, adrenaline and cortisol concentrations, and blood neutrophil responses following the two exercise bouts.

		Pre-EX1	Pre-EX2	Post-EX2
Neutrophils	CHO	1.80 (0.13)	3.46 (0.55)	5.57 (0.74)**
(10^9·L^{-1})	PLA	1.95 (0.14)	5.42 (0.66)**¶¶	6.38 (0.66)**
Elastase release	CHO	100 (0)	84 (6)	71 (9)*
(% of pre-EX1)	PLA	100 (0)	69 (6)* ¶¶	61 (7)**
CL	CHO	100 (0)	88 (4)	85 (3)*
(% of pre-EX1)	PLA	100 (0)	70 (5)** ¶	80 (7)**
Glucose	CHO	5.07 (0.08)	4.32 (0.12)*	4.21 (0.12)*
(mM)	PLA	5.01 (0.11)	4.67 (0.09)	3.28 (0.09)**¶¶
Adrenaline	CHO	378 (52)		735 (149)
(pM)	PLA	348 (55)		1290 (235)**¶
Cortisol	CHO	447 (55)	307 (93)	515 (102)
(nM)	PLA	397 (49)	282 (93)	812 (68)**¶¶

Values are mean (± SEM, n=8). Significantly different from pre-EX1 (* P<0.05, **P<0.01); significantly different from CHO (¶ P<0.05, ¶¶ P<0.01).

These findings suggest CHO feeding during a first exercise bout attenuates stress hormone responses to a subsequent bout of exercise but does not appear to markedly reduce subsequent exercise-induced suppression of neutrophil function.

Boyum A *et al.* (2002). *Eur J Appl Physiol* **88**, 20–28.
Robson PJ *et al.* (1999). *Int J Sports Med* **20**, 128–135.

All procedures accord with current local guidelines and the Declaration of Helsinki

C114

Thromboxane A_2-stimulated proliferation of cultured human colonic cancer cells

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Cyclooxygenase-2 (COX-2), which catalyses a key step in conversion of arachidonic acid to prostaglandin H_2, is overexpressed in human colorectal carcinoma tissue, and is associated with angiogenesis in the colorectal cancer (Williams *et al.* 1999). Thromboxane A_2 (TXA_2) is produced by thromboxane synthase (TXS), a downstream enzyme of COX-2, and is well known to induce platelet aggregation and vasoconstriction. TXA_2 has been reported to be associated with endothelial migration, angiogenesis and tumor metastasis (Nie *et al.* 2000). Recently, we found that TXS is overexpressed in human colorectal carcinoma tissues (Sakai *et al.* 2003). Here we found a novel function of TXA_2 in the colonic carcinoma cells.

The expression of TXS mRNA and the protein in human colonic carcinoma cell lines such as KM12-L4, HT-29, T-84, WiDr was examined by Northern blotting and Western blotting, respectively. To clarify the functional role of TXA_2 in colonic carcinoma cells, the TXS protein in the cells was disrupted by using specific antisense oligonucleotide (TXS-AS). Cell proliferation assay was performed by counting the number of the cell in a 12-well plate. In each well, 1×10^5 cells were seeded and cultured for two days. All data were statistically analysed using one-way ANOVA and Tukey's multiple comparison test.

We found that both the mRNA and protein of TXS were highly expressed in KM12-L4, HT-29, T-84 and WiDr cells. In KM12-L4 cells, the cell proliferation was significantly inhibited by the disruption of TXS protein by using TXS-AS. TXS inhibitors, such as Y-20811 (5 μM) and OKY-1581 (1 μM), also inhibited the proliferation of KM12-L4 and HT-29 cells. Furthermore, direct addition of 9, 11-epithio-11, 12-methano-TXA_2 (STA_2; 0.1 μM), a stable analogue of TXA_2, accelerated the proliferation of KM12-L4 and HT-29 cells. After the disruption of TXS protein in KM12-L4 cells, STA_2 still induced the cell proliferation.

Table 1: Involvement of thromboxane A_2 on the proliferation of cultured human colonic cancer cell lines

proliferation

	cell number (x 10^5 cells)	
	KM12-L4	HT-29
control	1.62 ± 0.03 (n = 4)	-
TXS-AS	1.34 ± 0.04 (n = 4)**	-
control	1.69 ± 0.06 (n = 6)	1.46 ± 0.03 (n = 5)
Y-20811	1.33 ± 0.07 (n = 6)**	1.29 ± 0.03 (n = 5)*
OKY-1581	1.38 ± 0.04 (n = 6)**	1.29 ± 0.04 (n = 5)*
control	1.56 ± 0.03 (n = 6)	1.44 ± 0.02 (n = 6)
STA_2	2.02 ± 0.05 (n = 6)**	1.69 ± 0.02 (n = 6)**
TXS-AS (control)	1.35 ± 0.02 (n = 6)	-
TXS-AS + STA_2	1.67 ± 0.03 (n = 6)**	-

All data are shown as means ± S.E.M. ** P< 0.01 *vs.* control, * P< 0.05 *vs.* control

These results indicate that TXA_2 stimulates the proliferation of human colonic carcinoma cells.

Nie D *et al.* (2000). *Biochem Biophys Res Commun* **267**, 245–251.
Sakai H *et al.* (2003). *J Physiol* **547.P**, C113.
Williams CS *et al.* (1999). *Oncogene* **18**, 7908–7916.

C116

The functional impact of central fatigue after exercise in man.

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Stimulation of the motor cortex using transcranial magnetic stimulation (TMS) and electromyographic (EMG) recordings have shown corticospinal excitability to be depressed following exercise (Brasil-Neto *et al.* 1994). When the exercise is exhaustive, depression can also be seen in motor evoked potentials (MEPs) of homonymous non-exercising muscles (Williams *et al.* 2003) and can therefore be attributed exclusively to central fatigue processes. We have now induced central fatigue in non-exercising muscles and measured its influence on force and speed of movement and reactions.

With local ethical approval and informed consent, six healthy male volunteers (aged 18–22 years) were seated with their arms relaxed on horizontal armrests and bilateral surface EMG recordings taken from the biceps brachii (BB) muscles. TMS was applied using a MagStim 200 stimulator connected to a 9-cm circular coil centred over the vertex. The stimulus intensity was set to 1.2 × threshold for evoking MEPs in relaxed BB muscles; MEP areas were measured bilaterally. Functional assessments were made bilaterally as follows: maximum voluntary contraction (MVC) force in elbow flexors, maximum hand grip (MHG) force, movement times (MTs) and simple reaction times (SRTs). Two assessment trials were completed before starting the exercise protocol and five more were made during the 35 min immediately following exercise. A 3.5kg weight was strapped to the wrist and exercise consisted of right-arm biceps curls, to a tone repeating at a frequency of 1 Hz, until exhaustion.

MEP areas in the exercised BB were depressed to 26.2 ± 6.6 % (mean ± S.E.M.) of the pre-exercise level (PEL) and MVC to 74 ± 8.6 % PEL in the nine min after exercise (P < 0.05; ANOVA on ranks). MEP areas in the non-exercised BB were depressed to 69.5 ± 8.8 % (PEL) but MVC showed a small insignificant (P > 0.05) rise (103.1 ± 5.2 % PEL) in the eleven min after exercise. MEPs remained depressed for up to 30 min (exercised 25.8 ± 3.8 % PEL; non-exercised 64.8 ± 8.2 % PEL); MVC recovered slightly in the exercised arm to 80.3 ± 4.6 % PEL and was unchanged in the non-exercised arm (102.8 ± 4.8 % PEL). Decrease in MEP area correlated with an increase in latency in the exercised (P < 0.05; r^2 = 0.13; linear regression) and non-exercised (r^2 = 0.13) arm but MEP area correlated with MVC only in the exercised BB (r^2 = 0.25). MHG showed drop to 96.5 ± 1.6 % PEL after ten min in the exercised arm and to 91.2 ± 2.6 % PEL in the non-exercised arm increasing to 88.0 ± 1.6 % PEL after 30 min. SRT decreased after 13 min in the non-exercised arm (95.4 ± 1.1 % PEL) and there was no change in MTs.We conclude that the central fatigue seen in the non-exercised arm has no effect on MVC in that arm. MHG was reduced in both arms after exercise but this could have been the result of peripheral fatigue induced by repeated testing procedures. The small improvement in SRT (faster reactions) seen after 13 min in the non-exercised arm may have been the result of practice. Although we saw depression of MEPs and increased latency in the non-exercised BB there appeared to be no measurable functional deficit in force or speed of movement or reaction.

Brasil-Neto JP *et al.* (1993). *Exp Brain Res* **93**, 181–184.
Williams KE *et al.* (2003). *J Physiol* **547P**, C143.

All procedures accord with current local guidelines and the Declaration of Helsinki

C117

Voluntary activation level and single fibre recruitment of human knee extensor muscle during lengthening contractions

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Maximal voluntary torque during lengthening contractions may be limited by a reduced neural drive, either by lower activation of all recruited fibres and/or by selective recruitment of type II fibres and concomitant de-recruitment of type I fibres.

Following ethic committee approval, ten subjects (6 males) signed informed consent forms. Voluntary activation levels during maximal lengthening, isometric and shortening contractions was investigated using superimposed electrical stimulation of the femoral nerve (triplet, 300 Hz). Recruitment of type I, IIA and IIAX fibre populations was assessed during these three modes of contraction (n = 5, 4 males) by analysis of needle muscle biopsies obtained at rest and immediately after exercise (10 contractions, 1 s on/1 s off). Single fibre fragments were isolated from freeze-dried samples and characterized using

mATPase stainings. Subsequently, remaining fragments of these fibres were measured for phosphocreatine (PCr) and creatine (Cr) content. A reduction in the ratio of PCr to Cr (PCr/Cr) was used as a measure of fibre activation (Beltman *et al.* 2003).

Voluntary activation (mean ± S.D) was significantly lower during lengthening (79 ± 8 %) than during isometric (93 ± 5 %) and shortening contractions (92 ± 3 %)($P < 0.05$; ANOVA for repeated measures). Maximal voluntary torque during lengthening (270 ± 55 Nm) was not significantly different from isometric contractions (252 ± 47 Nm, $P > 0.05$) but significantly higher compared with shortening contractions (199 ± 47 Nm, $P < 0.05$).At rest, PCr/Cr ratios (mean ± S.D) were 2.5 ± 0.6, 2.0 ± 0.7 and 2.0 ± 0.7, respectively for type I, IIA and IIAX fibres. After 10 lengthening, isometric or shortening contractions, the mean PCr/Cr ratios for grouped fibre populations were significantly different from rest and between contraction modes; 1.3 ± 0.2, 0.7 ± 0.3, 0.8 ± 0.6, respectively ($P < 0.05$, Kruskall Wallis followed by Mann-Whitney U-test). The calculated cumulative frequency distributions for the PCr/Cr ratio of each fibre type were significantly different from rest ($P < 0.05$, Kolmogorov-Smirnov two-sample test), suggesting activation of all fibre types. Compared to rest the shift in distributions was smallest for lengthening contractions ($P < 0.05$).

The present results show that there was a reduced voluntary drive during lengthening contractions. Changes in PCr/Cr ratios of single fibres suggest that this reduced drive is not caused by a selective recruitment of type II fibres but that all fibre populations were recruited at a lower rate.

Beltman JGM *et al.* (2003). *Acta Physiol Scand* (in press).

All procedures accord with current local guidelines and the Declaration of Helsinki

C118

Cerebral blood flow response to isocapnic hypoxia, during sleep and wakefulness

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Nocturnal hypoxia is a major pathological factor associated with cardio-respiratory diseases including obstructive sleep apnea and congestive heart failure. During wakefulness, a decrease in arterial oxygen tension results in a decrease in cerebral vascular tone and a consequent increase in cortical blood flow; however, the cerebral vascular response to hypoxia during sleep is unknown. We recently reported that cerebral vascular reactivity to CO_2 is markedly reduced during sleep (Meadows *et al.* 2003). From this we hypothesised that, in normal human subjects, isocapnic hypoxic cerebral vascular reactivity is decreased during stage III/IV, non-rapid eye movement (NREM) sleep compared to wakefulness.

In 13 healthy male individuals (mean age ± sd: 22 ± 4), left middle cerebral artery velocity (MCAV) was measured using transcranial Doppler ultrasound. In each subject, the cortical blood flow responses to four separate conditions (eucapnic euoxia, isocapnic euoxia, isocapnic hypoxia −5 % arterial oxygen saturation (S_{aO_2}), and isocapnic hypoxia −10 % S_{aO_2}), were tested during wakefulness and during the first 90-minute cycle of stage III/IV, NREM sleep. Isocapnic hypoxia was achieved by regulating the inspired fraction of oxygen, whilst maintaining the end-tidal partial pressure of carbon dioxide (P_{ETCO_2}) within ± 2 mmHg of a predetermined level, independent of changes in ventilation (Banzett *et al.* 2000). NREM sleep was determined using electroencephalograms and electro-oculograms.

During wakefulness, in response to isocapnic hypoxia (−5 and −10 % S_{aO_2}), the mean (± S.E.M.) MCAV increased by 8.8 ± 1.7 % and 12.9 ± 2.2 %, respectively ($P < 0.001$, ANOVA); during NREM sleep, the same levels of isocapnic hypoxia was associated with a −6.97 ± 1.6 % and −7.0 ± 1.6 % reduction in MCAV ($P < 0.001$). Mean arterial blood pressure was unaffected by isocapnic hypoxia ($P > 0.05$); R-R interval decreased similarly in response to isocapnic hypoxia during wakefulness −21.9 ± 10.4 %, $P < 0.001$) and sleep −20.5 ± 8.5 %, $P < 0.001$).

In summary, during wakefulness, in response to isocapnic hypoxia, cortical blood flow increased; in contrast, during sleep, in response to the same degree of isocapnic hypoxia, cortical blood flow decreased. The inability of the cerebral vasculature to react to hypoxia during sleep suggests a major state-dependent vulnerability associated with the control of the cerebral circulation and may contribute to the pathophysiologies of stroke and sleep apnea.

Banzett RB *et al.* (2000). *J Appl Physiol* **88**, 1597–1600.

Meadows GE *et al.* (2003). *J Appl Physiol* **94**, 2197–2202.

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C119

Molecular and morphological changes to the hypoxic human brain; focus on acute mountain sickness

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Acute mountain sickness (AMS) is a cerebral syndrome that presents at high-altitude and though preliminary neuroimaging data suggest a potential etiological role for extracellular vasogenic oedema (Hackett *et al*. 1998), the precise mechanisms responsible for disruption of blood-brain barrier function in hypoxia remain unresolved.

The present study therefore combined magnetic resonance imaging (MRI) with electron paramagnetic resonance (EPR) spectroscopy to directly examine, for the first time, the pathophysiological significance of free radical-mediated molecular damage to barrier function and associated neurological sequelae. Ethical approval was obtained from the Department of Medicine, University of Heidelberg. Fasting venous and cerebrospinal fluid (CSF) samples were obtained in the lateral decubitus position from twenty two healthy subjects aged 24 ± 2 (mean ± S.D.) years old after 16-18h passive exposure to normobaric hypoxia (12% O_2). Serum was analysed for the ascorbate free radical ($A^{\cdot -}$) or mixed *ex-vivo* with the diamagnetic spin trap, α-phenyl-*tert*-butylnitrone (PBN) prior to room temperature EPR spectroscopy (Bruker, EMX). Brain-specific proteins (neuron-specific enolase, S-100β), proinflammatory cytokines (TNF-α, IL-1β, IL-6) and vascular endothelial growth factor were also assayed via routine

techniques. Barrier function was examined using an established clinical technique (CSF/serum concentration quotient for albumin, IgG, IgA, IgM). Dual echo-, diffusion-weighted imaging [T_2-relaxation time/apparent diffusion coefficient (ADC)] and T_1-3D (volumetry) MRI sequences were determined in specific regions of interest in normoxia and after ~16 h of hypoxia. Clinical AMS was confirmed at 18 h if subjects presented with a Lake Louise self-assessment and clinical score of $\geqslant$5 points combined with an Environmental Symptoms Questionnaire Cerebral Score $\geqslant$0.7 points. Data were mathematically examined for distribution normality and selective differences investigated using an independent samples *t* or Mann Whitney *U* test where appropriate.

PBN spin-adducts recovered in serum and CSF were later identified via *in-vitro* experimentation and computer simulation experiments as lipid-derived alkoxyl/alkyl radicals [a^N = 13.6-14.2 Gauss (G) and a^H_β = 1.5-2.3 G] formed "downstream" subsequent to the oxidative catalysis of iron [Fe(III)-EDTA]. Eleven subjects diagnosed with AMS (50%) presented with a selective elevation in serum $A^{\cdot -}$ (AMS: 1638 ±274 *vs.* non-AMS: 1204 ±271 arbitrary units/$\sqrt{G}$, $P < 0.05$) and decrease in the CSF concentration of IL-6 (AMS: 9.26 ± 17.69 pg/ml *vs.* non-AMS: 17.19 ± 13.85 pg/ml, $P < 0.05$). Differences in absolute brain tissue volume (AMS: +0.7 ± 0.4 *vs.* non-AMS: +0.5 ± 0.4%) despite T_2-prolongation in the splenium of the corpus callosum, ADC, lumbar pressure (AMS: 11.8 ± 3.7 *vs.* non-AMS: 12.2 ± 5.1 cm/H_2O) and barrier function were unremarkable ($P > 0.05$).

In conclusion, these findings provide the first direct evidence for spin-trapped oxygen/carbon-centered free radicals in human CSF. Selective changes in brain morphology subsequent to free radical-mediated molecular damage to barrier function do not appear to be an initiating factor in AMS.

Hackett PH *et al.* (1998). *JAMA* **280**, 1920–1925.

All procedures accord with current local guidelines and the Declaration of Helsinki

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C120

Cardiac power output in ageing sedentary and endurance trained men

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The mammalian heart has a limited regenerative capacity (Anversa *et al.* 2002), so cardiomyocyte death, whether it occurs with normal ageing or ischaemic damage, results in a net loss of contractile cells. Furthermore, Olivetti *et al.* (1991) have reported a 33% loss of cardiomyocytes between 20 and 70 years of age. This, together with a decrease in peak cardiac output, and blunted inotropic and chronotopic responses to catecholamines etc are known to occur with increasing age (Lakatta 2000). Cardiac power output (CPO) represents the overall function of the heart, measuring both its pressure and flow generating capacities. When determined at rest CPO (CPO_{rest}) is around 1 watt (W) and like other indices of heart function fails to discriminate between age, fitness or heart failure (HF) (Fig. 1). It is only when the heart is maximally stimulated that true differences in CPO (CPO_{max}), and hence cardiac functional reserve (CR), become apparent (Fig. 1).

To test the hypothesis that healthy ageing is associated with a decrease in CPO_{max} and CR, and that endurance training improves CPO, six groups of male subjects were studied (Fig. 1). Ethical approval was given by the university and all subjects signed a consent form. All subjects were screened (questionnaire) to ensure that they were free from known cardiovascular diseases and medications. Cardiac output (CO) and mean arterial pressure (MAP) were measured non-invasively using the CO_2 rebreathe and auscultation methods respectively, at rest and at maximal exercise (Cooke *et al.* 1998) and CPO was calculated as $(MAP \times CO) \times 2.22 \times 10^{-3}$. CR was calculated from $CPO_{max} - CPO_{rest}$. A one-way ANOVA was used to measure statistical significances between groups, with significance defined as $P < 0.05$, using a Tukey's post hoc analysis.

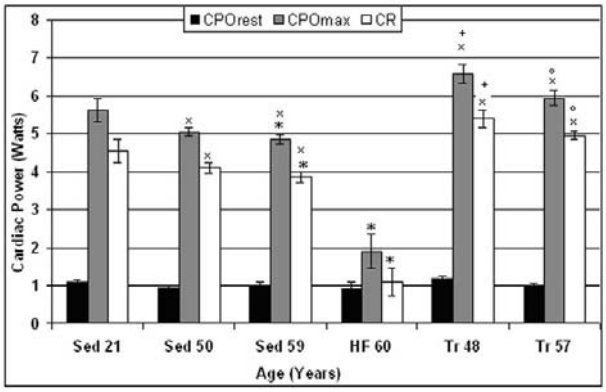

Figure 1. Cardiac Power with Age and Physical ActivityData are means ± S.E.M. and *n* = minimum of six in each group. Significant differences ($P < 0.05$) were found when compared with *Sed 21-yr-old, +Sed 50-yr old, °Sed 59-yr old, ×HF Sed 60-yr-old.

CPOmax and CR declined significantly ($P < 0.05$) by 16% and 18%, respectively in sedentary (Sed) subjects between the ages of 20 and 60 years. These changes were accentuated in patients suffering from NY Class III HF (Fig. 1). In contrast to these effects of ageing, the veteran athletes (Tr) demonstrated significantly ($P < 0.05$) higher CPOmax and CR values than their 'age-matched' sedentary counterparts at both 50 and 60 years of age (Fig. 1). Of interest is the finding that these trained individuals exhibited similar or better cardiac function than the Sed 20 yr-old subjects.

These results indicate that normal healthy ageing is associated with a reduction in overall cardiac function, represented by a decline in CPO_{max} and CR and that endurance training can ameliorate such changes in cardiac functional reserve.

Anversa P *et al.* (2002). *Journal of Molecular Cellular Cardiology* **34**, 91–105.

Cooke GA *et al.* (1998). *Heart* **79** , 289–294.

Lakatta E (2000). *Clinics in Geriatric Medicine* **16**, 419–443.

Olivetti G *et al.* (1991). *Circulation Research* **68**, 1560–1568.

We are grateful to the British Heart Foundation for their support of this research

All procedures accord with current local guidelines and the Declaration of Helsinki

C121

Serum S100b, a proposed marker of blood-brain barrier permeability, increases following prolonged exercise in a warm environment in man

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The blood-brain barrier (BBB) maintains a stable environment for the central nervous system (CNS) by regulating the influx and efflux of molecules. Metabolic changes during prolonged exercise may lead to increased BBB permeability (Chaouloff, 1997; Sharma *et al.* 1991). This may affect normal brain function and contribute to the development of central fatigue during exercise by altering the transport kinetics of neurotransmitter precursors or allowing the accumulation of unwanted substances in the CNS. The aim of this preliminary study was to examine changes in serum S100b, a proposed peripheral marker of BBB permeability (Kapural *et al.* 2002), following prolonged exercise in temperate and warm conditions.

With ethics committee approval, seven active males (mean ± S.D.; age 26 ± 5 years, $\dot{V}_{O_2,peak}$ 4.07 ± 0.22 l min^{-1}) completed experimental trials in temperate (Tmp) and hyperthermic (Hyp) conditions separated by at least 7 days. Subjects entered the laboratory in the morning following an overnight fast, and were seated in a comfortable environment (22–24 °C) for 15 min before sitting immersed to the neck in water at 35.0 ± 0.1 °C (Tmp) or 39.0 ± 0.1 °C (Hyp) for 30 min. Subjects then entered a room maintained at either 18.3 ± 1.8 °C (Tmp) or 35.0 ± 0.3 °C (Hyp) and cycled for 60 min at 60 % $\dot{V}_{O_2,peak}$. Serial venous blood samples were collected and analysed for serum S100b by microplate ELISA. Differences ($P < 0.05$) between trials were evaluated using ANOVA with repeated measures and Tukey's post-hoc and paired *t* tests as appropriate.

Serum S100b was elevated after exercise in the Hyp trial (+0.12 ± 0.10 μg l^{-1}; $P = 0.02$), but not after the Tmp trial ($P = 0.238$). Water immersion and exercise elevated core temperature by 2.1 ± 0.5 °C to 39.5 ± 0.3 °C at the end of exercise in the Hyp trial compared to a 0.9 ± 0.2 °C increase during the Tmp trial ($P < 0.001$). Heart rate ($P < 0.001$), as well as blood glucose ($P < 0.001$) and lactate ($P < 0.001$), was significantly higher during exercise in the warm environment. Ratings of perceived exertion ($P < 0.001$) and thermal comfort ($P < 0.001$) were markedly higher throughout the Hyp trial than the Tmp trial.

This study has demonstrated that serum S100b is elevated following prolonged exercise in a warm environment, suggesting that BBB permeability may be altered. Previous animal studies have observed a marked increase in BBB permeability following forced swimming exercise (Sharma *et al.* 1991). The development of hyperthermia, an upregulation in central serotonin synthesis and an increased production of cytokines have all been suggested as possible factors contributing to this response.

Chaouloff F (1997). *Med Sci Sports Exerc* **29**, 58–62.

Kapural M *et al.* (2002). *Brain Res* **940**, 102–104.

Sharma HS *et al.* (1991). *Neurosci Res* **10**, 211–21.

All procedures accord with current local guidelines and the Declaration of Helsinki

C122

Carotid baroreflex regulation of vascular resistance in high altitude Andean natives with and without chronic mountain sickness (CMS)

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Chronic mountain sickness (CMS), a maladaptation syndrome to chronic hypoxia, occurs in some Andean high altitude natives. Recently it has been reported that baroreflex cardiac control is impaired in CMS (Keyl *et al.* 2003). The experiments reported here were undertaken with the aim of determining whether carotid baroreflex control of vascular resistance is also altered in human subjects with CMS.

Nineteen Andean natives (10 with CMS and 9 control) were recruited in Cerro de Pasco, Peru (altitude 4338 m). The local ethics committee approved the study. Carotid baroreflex function was studied on the day following arrival in Lima (150 m). Baroreflex control of vascular resistance in the forearm was assessed from changes in finger blood pressure (photoplethysmography) divided by brachial flow velocity (Doppler ultrasound). A modified neck chamber and graded pressures of −40 to + 60 mmHg were used to change the stimulus to carotid baroreceptors. Individual stimulus-response curves were defined and sigmoid functions were applied. From the first derivative of the sigmoid curves, the maximal slopes (equivalent to peak gain) and the corresponding carotid pressures (equivalent to 'set point') were determined.

There were no significant differences between indicators of carotid baroreflex function at 150 m in the two subject groups. The maximal slope of the stimulus-response curve was −2.1 ± 0.6 units for CMS subjects and −2.4 ± 0.7 units for control subjects (mean ± S.E.M.; unpaired *t* test, $P > 0.05$). The corresponding carotid pressures were 82 ± 4 mmHg and 77 ± 6 mmHg for CMS and control subjects respectively ($P > 0.05$).

We conclude that CMS has no effect on carotid baroreflex regulation of vascular resistance in high altitude Andean natives tested at 150 m.

Keyl C *et al.* (2003). *J App Physiology* **94**, 213–219.

All procedures accord with current local guidelines and the Declaration of Helsinki

C124

When upright moderate internal jugular venous pressure remains zero during exercise in humans

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When upright cerebral blood flow may be supported by a siphon, i.e. that the hydrostatic challenge of raising arterial blood from heart-level to the brain is balanced by the equal gravitational force accelerating the venous outflow from the brain. On the

other hand, if the delicate veins collapse when they leave the skull and enter the neck as indicated by an ultrasound evaluation of the internal jugular vein (Cirovic *et al.* 2003), then the siphon may work only for that part of the circulation to the brain that is encompassed within the skull. Outside the brain the venous outflow pressure may approach zero and the value dependent only on how rigid the neck facia is. Given the siphon effect on cerebral blood flow, it is of no consequence whether mean arterial pressure (MAP) varies with the distance from the heart to the level of the brain. In contrast, MAP of a giraffe is elevated according to the length of its neck (Hargens *et al.* 1986) as would be expected in order to preserve perfusion pressure to the brain under the premises that a siphon does not contribute significantly to its flow.

Five male subjects (mean ± S.D.; age: 22.8 ± 22 years) participated in this study which was approved by the Ethics Committee of Copenhagen (KF 01-369/97). A retrograde catheter determined jugular venous pressure (JVP) at the level of the venous bulb while mean arterial pressure (MAP) and central venous pressure (CVP) were monitored. Measurements were taken when subjects were supine and sitting up. The determination was continued during submaximal exercise (heart rate of 130 beats min^{-1}), which might influence central venous pressure especially during semi-supine cycling. A repeated measureas one-way ANOVA was used to check difference in variables with the Tukey post hoc test employed when main effects were statistically significant at $P < 0.01$.

When supine MAP was 94.3 ± 1.7 mmHg (mean ± S.D.), CVP 7.0 ± 0.5 and JVP 11.1 ± 0.6 mmHg. When sitting MAP increased to 95.3 ± 2.0 mmHg, while CVP and JVP decreased to 0.8 ± 1.4 and −3.0 ± 1.0 mmHg, respectively ($P < 0.01$). During exercise MAP was 93.9 ± 1.9, CVP 0.5 ± 1.3 and JVP −3.26 ± 1.0 mmHg (values different from those when supine, $P < 0.01$, but not from those when upright at rest).

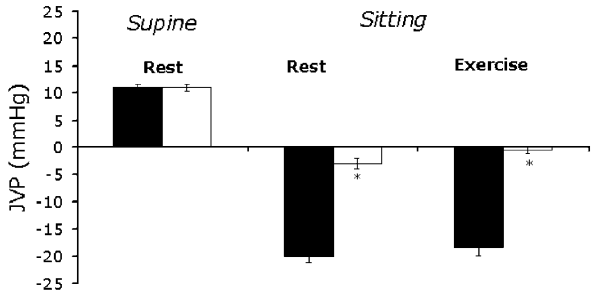

Figure 1. Jugular venous pressure during supine rest, during sitting rest and during sitting exercise. Black block, JVP estimated from CVP and distance to the jugular vein. White block, measured JVP. Values are mean ± S.D. *, significantly different from supine rest, $P < 0.01$.

These results indicate that in upright humans, both at rest and during exercise, jugular venous pressure is zero and negates a siphon effect on the outflow from the brain.

Cirovic S *et al.* (2003). *Aviat Space Envron Med* **74**, 125–131.

Hargens AR *et al.* (1987). *Nature* **329**, 59–60.

All procedures accord with current local guidelines and the Declaration of Helsinki

C125

Rate of torque development at the start of maximal voluntary and electrically-induced isometric contractions at different knee angles

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The effect of knee angle on maximal isometric torque rise was investigated with voluntary and electrically activated contractions of the knee extensors.

Seven healthy male subjects (19–40 years) with different training backgrounds, signed informed consent and the local ethics committee approved the study. Following a practice session, subjects were tested at 30 deg and 60 deg, and at 90 deg and 60 deg (random order) knee angles (full extension 0 deg) on separate days. Subjects were firmly secured in a chair with straps on hips and shoulders, and the lower leg was tightly strapped to a force transducer. Before each muscle contraction the upper leg was firmly strapped to the seat, this strap was released between contractions (3 min rest in between). Stimulation current was increased until force measured at the shin in response to burst femoral nerve stimulation (eight 100 μs pulses applied at 300 Hz) levelled off. Thereafter, maximal voluntary extension and flexion torque was obtained. This was followed by three to six voluntary attempts to increase knee extension torque from a fully relaxed state, without any pre-flexion, as fast (and hard) as possible. Finally, two maximal extensions with superimposed burst stimulation were performed to calculate the maximal torque generating capacity (MTGC) of the knee extensors. Surface EMG electrode pairs were placed over the vastus lateralis, rectus and biceps femoris muscles. Angle effects (means±S.D.) were tested for significance ($P < 0.05$) with repeated measures ANOVA followed by Bonferroni post-hoc tests.

MTGC at 30 deg, 60 deg and 90 deg knee angle was 188±45 Nm, 315±64 Nm and 245±35 Nm ($P < 0.05$) respectively. Torque time integral over the first 40 ms of the voluntary fast contractions (TTI40) was similar ($P = 0.62$) across knee angles. When expressed as a percentage of TTI40 obtained with burst stimulation (an indication of the muscles maximum potential), the fastest and slowest subjects used 83.3±3.2 % (mean across angles) and 10.5±3.1 % of their muscles' maximal potential, respectively. Furthermore, a positive linear relation was found ($r = 0.87$) between average (across extensor muscles and knee angles) rectified surface EMG (% MVC) of the knee extensors during the 40 ms before torque rise and fast voluntary TTI40.It was concluded that in contrast to maximal torque, maximal initial rate of voluntary isometric torque rise (absolute values) was independent of knee angle. In addition, there were substantial and consistent differences among subjects with respect to their ability for maximal voluntary activation of the knee extensors at the start of a fast isometric contraction.

All procedures accord with current local guidelines and the Declaration of Helsinki

C127

Does hormone replacement therapy affect steadiness of quadriceps force in post-menopausal women?

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With advancing age there is a progressive muscle atrophy that results in a loss of strength and power. Aged muscle has also been reported to be less able to produce steady force contractions than younger muscle (Tracy & Enoka, 2001). The mechanisms could include altered muscle control and may be linked to the increased susceptibility to falling and loss of performance of functional activities with increasing age. The decline in strength follows a different time course in men and women. In men there is a gradual decline from the fifth decade onwards, whereas in women there is a faster decline around the menopause (Samson *et al.* 2000). The latter is believed to be due to reduced oestrogen levels and the effect is lessened by hormone replacement therapy (HRT) (Skelton *et al.* 1999). This study aimed to investigate whether HRT can also affect the decline in steadiness of quadriceps force in women.

The quadriceps were tested for isometric strength. Steadiness of contraction force at 10, 25, 50 and 100 % of the maximum voluntary isometric force (MVC) using the force coefficient of variation (CoV). Three groups of subjects were studied: 1) 15 elderly women (68.1 ± 5.3 years (mean ± S.D.)) taking HRT, 2) 14 elderly women who had never taken HRT (70.5 ± 5.8 years), and 3) 16 young females (27.4 ± 5.6 years). Local ethical approval was obtained for the study, which was carried out in accordance with the Declaration of Helsinki. Results were tested for significance using ANOVA.

There was a tendency for all groups to be most steady during maximal, compared to sub-maximal contractions. There were no significant differences in steadiness between the three groups at any of the contraction levels (e.g. CoV at 50% MVC; 1.61 ± 0.13%, 1.58 ± 0.13 and 1.95 ± 0.15 for groups 1, 2 and 3 respectively). The young were stronger (297.5±13N) than the elderly group not on HRT (241.3 ± 12.7N) $P < 0.05$, but of similar strength to the group on HRT (255.4 ± 10.0).

These findings suggest that the steadiness of force generation in the quadriceps is unaffected by HRT and that the loss of this aspect of motor control is not a universal finding in older people.

Samson MM *et al.* (2000). *Age & Ageing* **29**, 235–242.

Skelton DA *et al.* (1999). *Clin Sci* **96**, 357–364.

Tracy BL & Enoka RM (2001). *J Appl Physiol* **92**, 1004–1012.

This work was supported by The Guy's and St.Thomas' Charitable Foundation.

All procedures accord with current local guidelines

C128

Intra-individual variability in heat strain responses to a standardised work-in-dry heat test in humans

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Little is known about how much of the inter-individual variability in heat strain responses to exercise-heat stress is due to intra-individual variability, although preliminary research suggests that it may be relatively high (Livingstone *et al.* 1992). As an understanding of all sources of variability is important for setting occupational heat stress limits, the aim of the present study was to quantify the intra-individual variability in heat strain to a standardised work-in-heat test.

Following local ethics committee approval, 7 men (mean (1 S.D.): age, 26.1 (4.9) years; body mass, 77.7 (12.0) kg; peak oxygen uptake, 51.8 (7.9) ml min^{-1} kg^{-1}) undertook 120 min of treadmill walking (speed, 1.53 m s^{-1}; grade, 5 %) in a hot-dry environment on five occasions (HD1–5). Dry-bulb temperature, globe temperature, relative humidity, and air speed were 35.33 (0.1) °C, 35.73 (0.1) °C, 19.93 (0.1) %, and 1.13 (0.1) m s^{-1} respectively. Subjects wore lightweight clothing and drank water *ad libitum*. The tests were undertaken at a rate of one per week to minimise the effects of partial heat adaptation (Barnett & Maughan 1993). Rectal temperature (T_{re}), mean skin temperature (T_{sk}) and heart rate (HR) were measured every min. Sweat loss (m_{sl}) was calculated from changes in nude body mass. Metabolic rate (*M*) was measured (Douglas bags) at 10 and 110 min. Data were analysed by a two-factor (Week and Time) ANOVA.

There was a Week-Time interaction in T_{re} ($P < 0.05$) (Fig. 1), although there were no significant effects in the other variables. The mean heat strain variables (across Weeks) at 120 min were: T_{re}, 38.3 (0.4) °C; T_{sk}, 36.0 (0.8) °C; HR, 146 (22) b min^{-1}; and m_{sl}, 1.0 (0.2) kg h^{-1}. Mean *M* was 295 (26) W m^{-2} at 110 min.

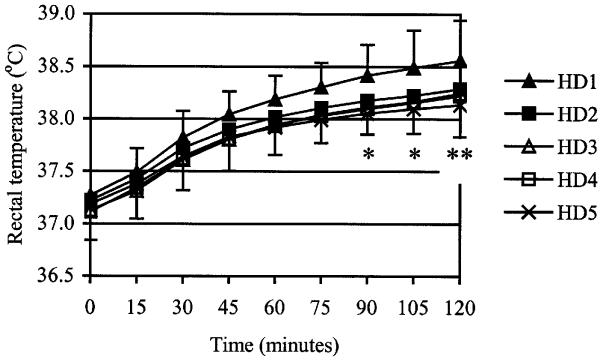

Figure 1. Rectal temperature during HD1–5 Differences (Newman-Keuls) between HD1 and HD5 are shown by * ($P < 0.05$) and ** ($P < 0.01$).

The mean intra-individual S.D. / coefficient of variation (across HD1–5) were: T_{re}, 0.2 °C / 0.5 %; T_{sk}, 0.3 °C / 0.8 %; HR, 5.9 b min^{-1} / 4.3 %; and m_{sl}, 0.1 kg h^{-1} / 6.3 %; and *M*, 7.5 W m^{-2} / 2.6 %. The intra-individual variability in heat strain is relatively large and likely to account for a significant proportion of the total variability during exercise-heat stress. If this variability in T_{re} is greater than the differences between heat stress limits for different values of *M* (typically 2–3°C) then the limits may be impracticable. This study has also established that separating work-in-heat tests by 7 days may be insufficient to negate the effects of heat adaptation.

Barnett A & Maughan RJ (1993). *Br J Sp Med* **27(1)**, 39–44.

Livingstone SD *et al.* (1992). *Proceedings of The Fifth Int Conf On Environmental Ergonomics*, 4–5.

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All procedures accord with current local guidelines and the Declaration of Helsinki

C129

Comparison of isometric and anisometric physiological tremor in three muscles in man

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There is evidence that instability in the control of a human muscle may occur under appropriate conditions of inertial and spring loading (Joyce & Rack, 1974; Matthews & Muir, 1980; Matthews, 1997). This is believed to depend on the stretch reflex and causes an oscillation in the 8–12 Hz range. However, the extent to which this mechanism contributes to normal and exaggerated physiological tremor is uncertain. Here we analyse the tremor in three muscles, known to have different stretch reflex characteristics: strong in biceps brachii, weak in first dorsal interosseous of the hand (FDI) and absent in anterior digastric.

The nine subects taking part (aged 22–73 years) had no history of neurological disorder, but four of them commented that their tremor was more noticeable than they thought normal (exaggerated physiological tremor). The study was approved by the local Ethical Committee and subjects gave written, informed consent. The surface electromyogram (EMG) recorded from each muscle was amplified (bandwidth 50 Hz – 3 kHz), rectified and filtered (0.5–60 Hz). Force of muscle contraction was recorded through a strain gauge via a rigid connection (isometric) or via a spring (anisometric: compliance 0.31 mm N^{-1}) and the signal also filtered as for the rectified EMG. Both signals were sampled at 500 Hz. Subjects were asked to maintain a known proportion of their maximum voluntary contraction (MVC) for 20 seconds for three periods separated by a min of rest. For high levels of force, the maintained period was reduced to 10 s and repeated 6 times to avoid fatigue. Recordings during contraction were analysed to compute auto-and cross-spectra.

For normal subjects, no peak in the 8–12 Hz range was observed for either the EMG or force spectra for any muscle in the isometric condition. In anisometric conditions, the spectra of biceps were both dominated by peaks in this region, particularly at 30–50 % MVC. In FDI, peaks were either absent or small, even when the finger was inertially loaded to bring its natural frequency to be similar to that for the forearm. In digastric, no peaks were observed under any conditions. These observations are consistent with the instability being due to the stretch reflex.

For subjects with exaggerated tremor, the clearest difference was the presence of 8–12 Hz oscillations in biceps and FDI in both isometric and anisometric conditions. Signs of instability were never seen in digastric. It appears that a cause of exaggerated physiological tremor may be an enhanced stretch reflex.

Joyce GC & Rack PMH (1974). *J Physiol* **240**, 375–396.

Matthews PBC (1997). *J Physiol* **489**, 249–275.

Matthews PBC & Muir RB (1980). *J Physiol* **302**, 427–441.

All procedures accord with current local guidelines and the Declaration of Helsinki

C130

Effect of body tilt angle on calf muscle performance in humans

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Tilting the body from a horizontal to a more upright position increases the time on a maximal graded cycle test by ~15 % (Koga *et al.* 1999). To further explore this effect, we tested the effect of body tilt angle on the strength and endurance of the plantar flexors.

Young human subjects were fixed to a tilt-table that could tilt them from the horizontal (0°) to upright (90°) position, and the application of force to a footplate through isometric action of the right calf muscle. In Experiment 1, six subjects completed five maximal voluntary efforts to determine the maximum force (Fmax). They then performed a graded test to the point of failure at three tilt angles (0°, 47° and 90°), during which the force increased by 100 N each three min from an initial force of 100 N. The force was applied in an intermittent manner (3 s contraction, 3 s relaxation). The 0° and 47° tests were repeated with the blood flow to the leg eliminated (i.e. ischaemia) in four of the six subjects. In Experiment 2, seven subjects performed four maximal voluntary efforts to determine Fmax and then a constant-force test (70 % Fmax; 2 s contraction, 4 s relaxation) to the point of failure. The constant-force tests were performed in the horizontal and three inclined positions (32°, 47° and 67°). The 0° and 67° tests were also performed under ischaemia in four of the seven subjects. These procedures were approved by the ethics committee of Trinity College Dublin. All results are shown as means ± standard deviations. Significant differences ($P < 0.05$) between body tilt angles were identified using a one-way repeated measures ANOVA and then located using a Tukey's HSD test.

In Experiment 1, tilt angle had no effect on Fmax: 1215 ± 181 N (0°), 1237 ± 146 N (47°) and 1201 ± 95 N (90°). Likewise, in Experiment 2 tilt angle had no effect on Fmax: 1392 ± 215 N (0°), 1424 ± 239 N (32°), 1448 ± 243 N (47°) and 1442 ± 239 N (67°). Time to failure during the graded test was significantly higher at 47° (25.9 ± 2.0 min) and 90° (25.1 ± 3.0 min) than 0° (22.2 ± 2.6 min). Under ischaemia there was no difference in time to failure during the graded test at 47° (7.1 ± 1.2 min) and 0° (7.1 ± 1.0 min). Time to failure during the constant-force test was also significantly higher at 32° (7.9 ± 3.2 min), 47° (8.9 ± 5.0 min) and 67° (9.3 ± 5.7 min) compared with 0° (4.6 ± 2.4 min). Under ischaemia there was no significant difference in time to failure during constant-force exercise between 67° (1.6 ± 0.5 min) and 0° (1.6 ± 0.7 min).

These results confirm that the endurance of a small muscle group is improved as the limb is tilted above the horizontal, that this effect occurs in the absence of an effect on strength, and that it depends on an intact peripheral circulation.

Koga S *et al.* (1999). *J Appl Physiol* **87**, 253–260.

All procedures accord with current local guidelines and the Declaration of Helsinki

C131

Signal evaluation of an indirect piezoelectric respiratory transducer in man

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Respiratory transducers requiring a mask or mouthpiece generate volumetric data that is notionally accurate, but they are unsuitable for prolonged use and also cause significant artefactual changes in ventilatory parameters (Perez & Tobin, 1985; Weissman *et al.* 1984). Accordingly a variety of indirect respiratory transducers has been developed (Sackner & Krieger, 1989; Sartene *et al.* 1990). One such is the Pneumotrace transducer (PT), based on a piezoelectric device incorporated into an elastic belt (UFI, 545 Main C-2, Morro Bay, CA 93442; UK suppliers: AD Instruments Ltd., Unit 56, Monument Business Park, Chalgrove, Oxfordshire OX44 7RW). There is little or nothing by way of published evaluation of this particular type of transducer.

In order to evaluate the PT's signal characteristics, a mechanical spring-loaded device was constructed, capable of imposing step extensions on the PT, of variable magnitude and from a range of starting lengths. The transducer showed a phasic response to this type of input, with exponential die-away of time constant 2.4 s. Peak output was linearly related to step magnitude for any fixed starting length of the transducer, but the proportionality constant decreased markedly with increasing starting length. The resulting experimentally-derived expression for the step response was differentiated to give an impulse response function, and Fourier transform of the latter yielded the system response in the frequency domain. This analysis revealed that, if presented with a sinusoidal input of constant amplitude, the magnitude and phase of the PT output would be, respectively, directly and inversely related to input frequency.

In vivo measurements were then made in human subjects ($n = 5$), using simultaneous recording from a pneumotachometer and PT transducer. The study had local ethical committee approval and the written consent of the participants. The subjects performed both spontaneous resting breathing as well as breaths having exaggerated and suppressed volumes. The results confirmed that the PT imposed a phase change on the signal (under 5% of average breath cycle time), although this was less than predicted by the sinusoidal modelling data. Integrated pneumotachometer output and PT peak height showed Pearson correlation coefficients of 0.85 to 0.97. Estimates of the error associated with prediction of individual tidal volumes from the PT data indicated confidence intervals of the order of 20–30%, in line with other non-invasive techniques (Sackner & Krieger, 1989).

In steadily-breathing subjects, the PT is capable of generating useful data for breath cycle time. Breath volume data are more approximate and, in addition, the device yields a less accurate record of ventilation when breathing pattern is unstable. This results both from its phasic responsiveness and also from the dependence of signal magnitude on initial transducer extension.

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All procedures accord with current local guidelines and the Declaration of Helsinki

C132

Effects of an increase in core body temperature with and without elevations in muscle temperature on repeated high-intensity exercise performance in man

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An elevated muscle temperature increases power production in man (Sargeant 1987), however elevating core body temperature has been shown to reduce force generation during prolonged maximal voluntary contractions (Nybo & Nielsen, 2001). The purpose of this study was to investigate the effect of an increase in core body temperature with and without elevations in muscle temperature on power output during repeated high-intensity cycle exercise.

Following local ethics committee approval nine healthy males (age 20 ± 1 years (mean ± S.D.), height 185 ± 6 cm, body mass 77.6 ± 12.2 kg) completed three experimental trials in randomised order, after habituation to the exercise test. Condition 1, subjects were immersed in hot water (42.9 ± 0.2 °C) up to the gluteal fold for 40 min and up to the sternal notch for the last 5 min (Hot Core+Legs). Condition 2, subjects were immersed in hot water up to the sternal notch until core body temperature (rectal) increased to 38.5 °C while the leg temperature was maintained at normal with ice packs and cold water (Hot Core). Condition 3, subjects stood in an empty tub for 45 min (Control). Ten min after each intervention subjects completed two 30-s sprints on a cycle ergometer (frictional load 0.075 kg kg^{-1} body mass) with 4 min recovery between sprints. Power output was corrected for flywheel acceleration. Muscle (vastus lateralis) temperature was measured at rest and 5 min before the first sprint (pre-SpI). Data were analysed using repeated measures ANOVA with Tukey *post-hoc* where appropriate. Statistical significance was accepted at $P < 0.05$.

Table 1. Performance variables for Sprint I & II for Control, Hot Core and Hot Core+Legs conditions

	Sprint I		
	Control	Hot Core	Hot Core+Legs
PPO (W)	1095 ± 173	1131 ± 180	1214 ± 178 *#
MPO (W)	645 ± 91	632 ± 83	668 ± 88 #
MinPO (W)	423 ± 79	378 ± 89 +	405 ± 110
MPC (rpm)	114 ± 8	112 ± 8	118 ± 11 #
F.I. (%)	60 ± 11	65 ± 11 +	66 ± 11 *
	Sprint II		
	Control	Hot Core	Hot Core+Legs
PPO (W)	1052 ± 145	1027 ± 163 ^	1008 ± 132 ^
MPO (W)	565 ± 89 ^	548 ± 85 ^	555 ± 98 ^
MinPO (W)	351 ± 101 ^	332 ± 111 ^	328 ± 112 ^
MPC (rpm)	101 ± 10 ^	98 ± 11 ^	100 ± 17 ^
F.I. (%)	67 ± 12 ^	67 ± 13	67 ± 13

+ Hot Core *vs.* Control, * Hot Core+Legs *vs.* Control

Hot Core+Legs *vs.* Hot Core, ^ Sprint II *vs.* Sprint I, $P < 0.05$

Prior to the first sprint (SpI) core (rectal) temperature was significantly greater in both hot trials than Control (Hot Core+Legs, 38.7 ± 0.2 °C; Hot Core, 38.6 ± 0.2 °C; Control, 37.4 ± 0.3 °C, $P < 0.05$). Muscle temperature was significantly greater ($P < 0.05$) pre-SpI in the Hot Core+Legs (39.1 ± 0.5 °C) compared to Control (35.6 ± 0.4 °C) and Hot Core

(35.5 ± 1.5 °C), $P < 0.05$. Mean power output (MPO) and pedal cadence (MPC) was ~5 % higher in Hot Core+Legs compared with Hot Core (SpI, $P < 0.05$, Table 1). Peak power output (PPO) was greater in Hot Core+Legs compared with both Control and Hot Core (SpI, $P < 0.05$). Minimum power output (MinPO) was greater in Control compared to Hot Core, which was reflected by a lower fatigue index (F.I.) compared to both hot conditions (SpI, $P < 0.05$). There were no differences in any power variables between conditions in the second sprint.

The results of this study show that an increase in core (rectal) temperature alone has no beneficial effect on repeated maximal exercise performance. The findings demonstrate that the beneficial effects of pre-warming on subsequent power output are more related to a higher muscle rather than core temperature. Any temperature-induced improvements in the first 30-s sprint did not persist during a subsequent sprint.

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Sargeant AJ (1987). *Eur J Appl Physiol* **56**, 693–698.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC73

The role of hypocapnia in the development of syncope during orthostatic stress following plasma volume expansion

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Hypotension is considered to be the cause of syncope during lower body negative pressure (LBNP). However, hypocapnia induced by hyperventilation has been observed in subjects preceding syncope, and may contribute to the onset of syncope by reducing cerebral blood flow during orthostatic stress (Morgan *et al.* 1997). Enhancing the 'respiratory pump' may partially attenuate the reduction in venous return that occurs during LBNP (Lipsitz *et al.* 1998; Novak *et al.* 1998), but the associated hyperventilation may actually exacerbate orthostatic intolerance. The aims of this study were to examine the effect of plasma volume expansion on tolerance to LBNP, and to consider the role of hypocapnia in the development of syncope.

Following approval by King's College London Ethics Committee, five subjects volunteered to participate in the study. Subjects performed two LBNP stress tests. LBNP was applied at 10 mmHg increments every 3 min until cessation at pre-syncope, or due to breathing difficulties. Subjects performed one test after ingesting 12 ml/kg body weight of electrolyte fluid (F trial), and one test following no fluid (NF trial). Measurements included change in plasma volume (cyanmethaemoglobin and microhaematocrit values from venous blood samples incorporated into equation by Dill & Costill, 1974), end-tidal PCO_2 (nasal catheter), blood pressure (Finapres), heart rate (ECG). Data presented as means ± S.E.M., and compared using paired *t* tests.

Following fluid ingestion, plasma volume increased ($P < 0.05$) by 3.25 %, and time to cessation of LBNP increased ($P < 0.05$) by 4.28 min (NF: 18.85 ± 1.28 min; F: 23.13 ± 1.81 min). End-tidal PCO_2 ($n = 4$) did not change from baseline values during mild LBNP up to −20 mmHg (NF: 42.13 ± 0.86; F: 42.23 ± 1.66 mmHg). However, as suction pressure increased to ⁻30 mmHg and above, end-tidal PCO_2 fell. Hypocapnia was greater during the NF trial, with a significantly lower ($P < 0.05$) end-tidal PCO_2 at suction of −50 mmHg (NF: 36.24 ± 1.67; F: 40.9 ± 1.55 mmHg). Mean end-tidal PCO_2 at time of LBNP cessation were similar between trials (NF: 31.70 ± 3.04; F: 33.27 ± 2.35 mmHg).

In conclusion, the smaller reduction in end-tidal PCO_2 during suction following plasma volume expansion was associated with increased tolerance to LBNP. Therefore, hypocapnia may be an important factor that contributes to orthostatic intolerance. However, plasma volume expansion may play an important role in reducing the hypocapnic contribution implicated in syncope during orthostatic stress by perhaps reducing the degree of hyperventilation that often leads to cerebral vasoconstriction.

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All procedures accord with current national and local guidelines and the Declaration of Helsinki

PC74

Haem oxygenase-1 induction *ex vivo* following hydrogen peroxide treatment of human lymphocytes and monocytes

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The induction of the antioxidant enzyme haem oxygenase-1 (HO-1) is a general response to oxidant stress in mammalian cells (Keyse and Tyrrell, 1989). Rothfuss *et al.* (2001) observed that an increase in HO-1 protein was associated with decreased oxidative DNA damage in lymphocytes exposed to hyperbaric oxygen. However, little is known about the time-course of HO-1 up-regulation in mononuclear cells. Therefore, the aim of the present study was to assess the HO-1 protein response over time in freshly harvested monocytes and lymphocytes exposed to oxidant stress in the form of hydrogen peroxide (H_2O_2).

After local ethical committee approval, seven male subjects (age 26 ± 2 years, height 1.77 ± 0.15 m, and body mass 80 ± 2 kg; mean ± S.E.M.) reported to the laboratory following an overnight fast. Subjects rested in a supine position for ten min prior to a venous blood sample (25ml) being taken. Subjects refrained from exercise, drinking alcohol and kept a record of their food and fluid intake for three days prior to blood sample collection. Mononuclear cells were isolated from peripheral blood and exposed to 50 μM H_2O_2 for 30 min at 37°C. Levels of HO-1 protein were analysed by flow cytometry (FACScan, Becton Dickinson, USA) using a monoclonal antibody (Stressgen, Canada) and fluorescein isothiocyanate conjugated secondary antibody (FITC) (Sigma, UK) at 4, 6, 24 and 48 h after treatment. HO-1 protein was expressed as the percentage change from the initial baseline value using the median fluorescence intensities of the treated and corresponding sham-treated controls. Data were analysed at each timepoint using a one-way ANOVA with *post-hoc* Tukey's test where appropriate. Statistical significance was accepted at $P < 0.05$.

There was a significant increase in HO-1 protein over time in both cell types ($P < 0.01$). The change in HO-1 protein was 287 ± 10 % and 183 ± 7 % at 48 h post treatment in lymphocytes and monocytes, respectively.

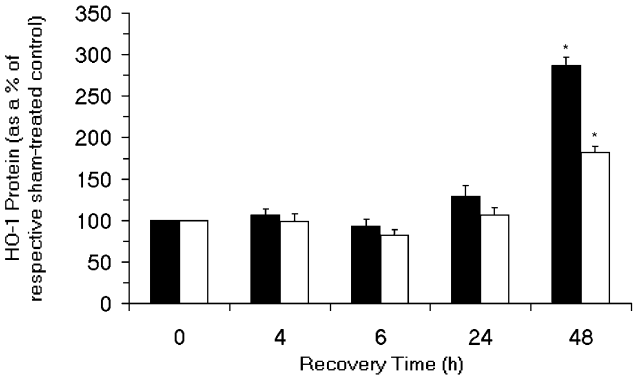

Figure 1. Time course of the HO-1 protein response to ex vivo H2O2 treatment in lymphocytes (solid bars) and monocytes (open bars) expressed as a percentage increase with respect to sham-treated controls. *$P < 0.01$ vs. 0,4, 6 and 24 h ($n = 7$; means ± S.E.M.)

Maximum levels of HO-1 protein were observed in every subject at 48 h post H_2O_2 treatment in both lymphocytes and monocytes. There was very little variability in the HO-1 response at this timepoint.

Keyse SM & Tyrrell RM (1989). *Proc Natl Acad Sci* **86**, 99–103.
Rothfuss A *et al* (2001). *Carcinogenesis* **22**, 1979–1985.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC75

The effect of intermittent exercise on complex motor skill performance in man

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Intermittent exercise attenuates performance of motor skills with a 'speed' criterion (McGregor *et al.* 1999) and imparts a greater thermoregulatory strain (Kraning & Gonzalez, 1991) compared to continuous exercise. Therefore, the aim of this study was to examine the effect of strenuous, intermittent exercise on performance in a complex motor skill task with an 'accuracy' criterion.

Five healthy male subjects (age 23.4 ± 5.6 years, height 1.80 ± 0.02 m, body mass 73.9 ± 4.6 kg, maximal $\dot{V}_{O_2}$ 62.3 ± 7.2 ml kg^{-1} min^{-1}; mean ± S.D.) completed a complex motor skill task before and immediately after an intermittent exercise challenge (Sherman *et al.* 2003). Subjects gave written, informed consent prior to the study, which had prior local ethics committee approval. Core body temperature (T_c) was monitored every 60 s using an ingested telemetry pill and changes in heart rate (HR) were measured every 15 s. Venous blood samples were obtained from a superficial forearm vein, at rest and after exercise, to determine concentration of whole blood glucose and lactate (La). Subjects consumed water *ad libitum* during each rest period. Body mass loss was corrected for volume of water ingested. Student's paired *t* tests were used to identify any differences, pre-*vs*. post-exercise. Level of significance was set at $P < 0.05$.

Table 1. The effect of intermittent exercise on accuracy of a complex motor skill task

	Rest	Post-exercise
Inner target accuracy (%)	29.0 ± 8.4	20.0 ± 10.5*
Outer target accuracy (%)	41.5 ± 11.0	37.0 ± 10.2
Gross error (%)	29.5 ± 16.9	43.0 ± 18.3

Values are mean ± S.D. ($n = 5$). Significantly different from pre-exercise: * $P < 0.05$.

Inner target accuracy was lower ($P < 0.05$) after intermittent exercise (Table 1), but T_c (pre, 37.3 ± 0.4 °C, post, 38.9 ± 0.6 °C, $P < 0.01$), HR (pre, 132 ± 14 beats min^{-1}, post, 169 ± 9 beats min^{-1}, $P < 0.01$) and La concentration (pre, 0.85 ± 0.19 mmol l^{-1}, post, 1.85 ± 0.73 mmol l^{-1}, $P < 0.05$) were increased. Body mass loss was 1.41 ± 0.54 kg and equivalent to 1.89 ± 0.62 % of pre-exercise body mass.This study provides preliminary evidence that increased thermoregulatory strain is temporally associated with decreased accuracy of a complex motor skill task.

Kraning KK & Gonzalez RR (1991). *J Appl Physiol* **71**, 2138–2145.
McGregor SJ *et al* (1999). *J Sports Sci* **17**, 895–903.
Sherman RA *et al* (2003). *Science and Racket Sports III* (in press).

All procedures accord with current local guidelines and the Declaration of Helsinki

PC76

An investigation into the impact of limiting the number of matches of competitive soccer on the fitness of elite youth team soccer players, aged 10 and 11 years old

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This study aimed to investigate the changes in physical fitness in young soccer players as result of participation in competitive soccer.

Season one (S1) was unrestricted in competitive soccer but during season two (S2), the subjects were only allowed to participate in 30 games. Subjects were recruited from a professional soccer academy (S1 $n = 12$; S2 $n = 14$). The subjects were aged between 10–11 years at the start of each season. Speed (sprint time over 10m), aerobic capacity from the multistage shuttle run (MSR) (Ramsbottom *et al.* 1988), and agility (measured by a run through cones) were monitored at regular intervals across S1 and S2, as well as anthropometric measurements. Somatotype was calculated using the Heath & Carter (1967) method. Linear regression coefficients (LRCs) for each measured parameter were calculated over each season and one-tailed *t* tests used to detect any significant changes. A Pearson correlation coefficient was calculated to test the relationships between parameters. Independent *t* tests were used to test whether the rate of change was different between the two seasons.

Height (S1 LRC = 0.048;S2 LRC = 0.125) and weight (S1 LRC = 0.137; S2 LRC = 0.187) increased significantly across both seasons ($P < 0.05$). Of the somatotype characteristics only ectomorphy decreased significantly across S2 (LRC = -0.032).

Examination of fitness parameters showed MSR and agility run performance were maintained with a significant improvement in 10 metre sprint time ($P < 0.05$) in S1 (LRC = -0.004). Across S2, 10 metre sprint performance was maintained, with agility run performance improving (LRC = -0.0092) and MSR performance decreasing significantly ($P < 0.05$) (LRC = -0.037). A comparison of the rate of change between S1 and S2 showed a significant decrease in MSR performance. There were no significant correlations between the anthropometric and fitness parameters.

MSR performance was maintained across S1 in accordance with the limited research in adult populations. Restricting the number of games would appear to result in a decrease in aerobic fitness across the season, possibly as a result of the removal of the physical stimulus from competitive soccer matches. This could be remedied by the introduction of sport specific aerobic training in practice sessions.

Ethical approval was obtained from the Chester College Ethics Committee.

Heath B & Carter JEL (1967). *American Journal of Anthropology* **27**, 57–74.

Ramsbottom RJ *et al.* (1988). *British Journal of Sports Medicine* **22(4)**, 141–144.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC77

Effect of sprint exercise on serum insulin-like growth factor-I (IGF-I) concentration and bioavailability

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Brief high intensity sub-maximal exercise elicits acute increases in serum (total) IGF-I independent of growth hormone (GH) release and has been suggested to alter the bioavailability of IGF-I through regulation of the IGF binding proteins (IGFBPs) (Schwarz *et al.* 1996). The responses of total IGF-I, free IGF-I and the IGFBPs to maximal sprint exercise have not previously been characterised. Therefore, we examined whether sprint exercise alters circulating IGF-I and IGF-I binding capacity as estimated by measurement of IGFBP-1 bound IGF-I.

Following ethics committee approval, five non-obese healthy men arrived at the laboratory after an overnight fast, completed a standardised warm-up and then a maximal 30 s sprint on a cycle ergometer. Venous blood samples were drawn pre-and 5 min post-sprint. Serum levels of total IGF-I, free IGF-I, IGFBP-1 and IGFBP-1 bound IGF-I (binary complex) were determined as previously described (Frystyk *et al.* 2002). Data were analysed using a Student's paired *t* test; significance taken at $P < 0.05$. Effect sizes (e.s.) were calculated for each variable; large effect when e.s.>0.8.

There were no significant changes in serum total IGF-I, free IGF-I, binary complex or IGFBP-I after sprint exercise (Table 1). IGFBP-I saturation increased following exercise ($P < 0.05$). Effect sizes for pre-vs. post-sprint were: total IGF-I, 0.90; free IGF-I, 0.01; binary complex, 0.37; IGFBP-I, 0.43; IGFBP-I saturation, 0.69. There was, therefore, a large effect for total IGF-I and a very small effect for free IGF-I.

	pre-	5 min post-	Δ
Total IGF-I ($\mu g\ l^{-1}$)	236 ± 16	274 ± 24	38
Free IGF-I ($\mu g\ l^{-1}$)	1.22 ± 0.19	1.21 ± 0.20	-0.01
Binary complex ($\mu g\ l^{-1}$)	15.0 ± 1.9	16.8 ± 3.4	1.8
IGFBP-I ($\mu g\ l^{-1}$)	26.8 ± 2.3	24.1 ± 3.2	-2.7
IGFBP-I saturation (%)	58.2 ± 8.8	74.2 ± 14.6*	16

Table 1. Serum total IGF-I, free IGF-I, binary complex and IGFBP-I and IGFBP-I saturation pre-and post-sprint (mean ± S.E.M.; $n = 5$). *Significant difference between pre- and post-sprint ($P < 0.05$).

This preliminary study indicates that a maximal 30 s sprint acutely increases IGFBP-I saturation, and tends to increase total IGF-I levels. However, free IGF-I remains unaltered.

Frystyk J *et al.* (2002). *J Clin Endocrinol Metab* **87**, 260–266.

Schwarz AJ *et al.* (1996). *J Clin Endocrinol Metab* **81**, 3492–3497.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC78

Plasma triacylglycerol concentrations following combined dietary and exercise intervention

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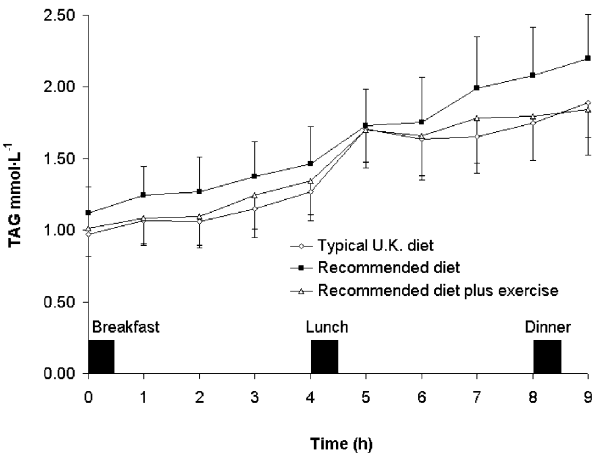
Although low-fat high-carbohydrate diets produce a desirable reduction in plasma cholesterol concentration they frequently lead to an undesirable increase in plasma triacylglycerol (TAG) concentration (Parks and Hellerstein 2000). Exercise has been shown to prevent a carbohydrate-induced hypertriacylglycerolaemia, at least in the short term (Koutsari *et al.* 2001). This suggests that the combination of a high-carbohydrate diet and frequent exercise may be an effective strategy for reducing cardiovascular disease risk. This study tested the hypothesis that adherence to physical activity targets (Pate *et al.* 1995) would offset the increase in plasma TAG concentration associated with dietary change in line with current recommendations (Krauss *et al.* 2000).



Six men and 8 women volunteered to participate in this study that was approved by Loughborough University's Ethical Advisory Committee. The age, height and body mass (mean ± S.E.M.) of the subjects were: 57.3 ± 1.3 yr, 1.70 ± 0.03 m

and 76.2 ± 6.0 kg. Subjects underwent 3, 4-day trials in a randomised, balanced design. The trials were: 1) typical UK diet (% energy: 40 % fat, 45 % carbohydrate, 15 % protein), 2) recommended diet (30 % fat, 55 % carbohydrate, 15 % protein), 3) recommended diet plus exercise (30 min of brisk treadmill walking daily). On day 4 of each trial blood samples were collected in the fasted state and for 9-h postprandially via a venous cannula. Breakfast, lunch and an early evening meal were consumed during this 9-h period at 0 h, 4 h and 8 h respectively. TAG concentrations were determined in plasma using an enzymatic assay (Humphreys *et al.* 1990), with correction for free glycerol. Findings were analysed with a repeated measures ANOVA using SPSS version 11.0 for Windows.

A significant ($P < 0.05$) trial × time interaction was obtained indicating a steeper increase in plasma TAG concentration over the observation day on the recommended diet compared with the typical UK diet. This effect was countered by exercise (Fig. 1). These data extend previous findings by showing the potential for even small changes in diet and exercise to influence TAG concentrations.

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Koutsari C *et al.* (2001). *Arterioscler Thromb Vasc Biol* **21**, 1520–1525.

Krauss RM *et al.* (2000). *Circulation* **102**, 2284–2299.

Parks EJ & Hellerstein MK (2000). *Am J Clin Nutr* **71**, 412–433.

Pate RR *et al.* (1995). *JAMA* **273**, 402–407.

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All procedures accord with current local guidelines and the Declaration of Helsinki

PC79

Regional variations in sweat composition in man

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Several studies have demonstrated regional variations in the solute content of sweat, but these studies have involved small subject numbers ($n = 6$, Lemon *et al.* 1986; $n = 10$, Patterson *et al.* 2000). The present study measured the electrolyte content of sweat from different anatomical sites in a larger subject population.

With Ethics Committee approval, 30 healthy young males drawn from two professional football clubs gave written consent to participate. Measurements were made during a 90-minute training session carried out as part of pre-season training. Regional sweat collections were made at the forearm, chest, back and thigh by application of absorbent gauze swabs which were covered with a non-porous adhesive film (Tegaderm dressing and pad, 3M, Loughborough). Before application the area was cleaned with distilled water and dried with a gauze swab. Body mass was measured before and after training and the mass of drinks consumed was also measured to estimate total sweat loss. Sweat volume in the patch was determined gravimetrically. Sodium and potassium concentration was measured by flame photometry and chloride concentration by coulometric titration. Statistical analysis was by one way ANOVA followed by the Tukey test as appropriate. Data are presented as mean ± S.D.

Mean whole body sweat rate was 1.43 ± 0.25l/h, with a wide range between individuals (1.03–1.89l/h). The sweat sodium (Na), potassium (K) and chloride (Cl) concentrations differed between the sample sites tested. The highest sweat Na concentration was measured at the chest (54 ± 21 mmol/l) and this was significantly greater than that at the arm and thigh (42 ± 16 and 35 ± 11 mmol/l; $P = 0.000$). Further, the thigh sweat Na concentration was lower than that on the back (49 ± 19 mmol/l). A very similar picture was found for sweat Cl concentration. The highest sweat Cl concentration was measured at the chest (53 ± 14 mmol/l): this was significantly greater than that at the arm and thigh (39 ± 11 and 32 ± 9 mmol/l; $P = 0.000$). Further, the thigh sweat Cl concentration was lower than that on the back (49 ± 14 mmol/l). However, for the sweat K concentrations, the results demonstrate the opposite effect. The highest sweat K concentration was measured at the thigh (6.7 ± 2.5 mmol/l) and this was significantly greater than that at the chest and back (5.4 ± 1.3 and 4.6 ± 1.2 mmol/l; $P = 0.000$). The arm sweat K concentration (6.4 ± 1.6 6 mmol/l) was higher than that on the back. The greatest sweat Na concentrations were therefore recorded from the trunk of the body while the greatest sweat K concentrations were recorded from the peripheral sites.

These results indicate a substantial regional variation in sweat electrolyte concentration. The K concentration cannot be explained by the effect of differences in sweating rate on ductal reabsorption. This is however likely to account for the differences in Na and Cl concentrations.

Lemon PWR *et al.* (1986). *J Appl Physiol* **61**, 1967–1971.

Patterson MJ *et al.* (2000). *Exp Physiol* **85**, 869–875.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC80

Lower expression of Na^+,K^+-ATPase α1-isoform in human colorectal cancer

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The catalytic subunit of Na^+,K^+-ATPase has at least four isoforms, α1, α2, α3 and α4, each derived from a different gene. The α-isoform exhibits a tissue-specific pattern of expression (Blanco & Mercer, 1998). α1-isoform is found in nearly every tissue and involved in a housekeeping function in all cells. α2-isoform is predominantly expressed in adipocytes, skeletal muscle, heart and brain. α3-isoform is abundant in brain. α4-isoform is expressed exclusively in testis. It has been reported that no significant change in the expression of α1-isoform was observed in human renal and lung carcinomas (Akopyanz *et al.* 1991; Rajasekaran *et al.* 1999).

Herein we investigated whether levels of protein expression of Na^+,K^+-ATPase α1-, α2- and α3-isoforms could be changed in human colorectal cancers. The specimens of colorectal adenocarcinomas and accompanying normal mucosa were obtained from surgical resection of patients in accordance with the recommendations of the Declaration of Helsinki and with ethics committee approval. Informed consents were obtained from all patients at Toyama Medical and Pharmaceutical University Hospital. Data are shown as means ± S.E.M. Difference between groups were analysed by one-way ANOVA. Comparison between the two groups was made with paired *t* test.

Western blotting was performed using specific antibodies against α1-, α2- or α3-isoform. Specificity of these antibodies was confirmed using the control antigen peptides. Interestingly, decrease in the expression of α1-isoform was observed in 12 of 12 cancers (100 %) compared with the normal mucosa. No

significant expression of α2-isoform was observed in any of the colorectal carcinomas and accompanying normal mucosa. On the other hand, increase in the expression of α3-isoform was observed in 9 of 12 cancers (75%) compared with the normal mucosa. Simultaneous change of expression of α1-isoform and α3-isoform was found in 6 of 12 cancers, and ouabain (5 μM)-sensitive ATPase activities of these cancer tissues and accompanying normal mucosa were 2.5 ± 0.6 and 3.2 ± 0.4 μmol Pi (mg protein)$^{-1}$ h^{-1}, respectively ($n = 9$).

These results suggest that changes in protein expression of α1- and α3-isoforms but not α2-isoform are associated with human colorectal cancers.

Akopyanz NS *et al.* (1991). *FEBS Lett* **289**, 8–10.
Blanco G & Mercer RW (1998). *Am J Physiol* **275**, F633–F650.
Rajasekaran SA *et al.* (1999). *J Urol* **162**, 574–580.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC81

The effects of exercise on biochemical markers of vascular inflammation and walking distance for patients with peripheral arterial occlusive disease (PAOD)

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Cramping pain and walking impairment results from muscular ischaemia in PAOD patients (Hiatt *et al.* 1990). Increases in C-reactive protein (CRP) and urinary microalbumin (quantified by an albumin creatinine ratio (ACR)) suggest endothelial damage from inflammation (Yudkin 1988), and may be involved in the progression of atherogenesis and PAOD (Kuller, *et al.* 1996). This study aimed to investigate the effect of a 12-week home exercise regimen (HER) on CRP, ACR, patient claudication walking distance (CWD) and maximum walking distances (MWD).

Ethical consent for all testing procedures was obtained by the South Cheshire Ethical Committee alongside approval from University College Chester's Ethical Board. Twelve PAOD patients (EG) and 12 age and sex match controls (CG) exercised for 1 h a day, five times per week for 12 weeks. Twelve age and sex matched healthy controls (CG) maintained normal activity. CWD was assessed as the distance (m) at which 'cramping' pain was first identified. MWD was recorded as the distance achieved upon cessation of the exercise test using a Gardner grade stage walking test. Venous CRP and urinary ACR were taken at T1 = baseline, pre HER, T2 = post treadmill test pre HER,T3 = baseline post HER and T4 = post treadmill test post HER.

CWD and MWD increased significantly in the EG (CWD 124m-258m) and (MWD 291m-543m) from pre-therapy distances. ACR levels in the EG increased from T1 (4 mg/mmol)-T2 (7.25 mg/mmol) ($P < 0.05$) and T3 (2.3 mg/mmol)-T4 (5.5 mg/mmol) ($P < 0.05$). ACR levels were significantly lower at post HER at T4 (5.5 mg/mmol) compared to pre-HER T2 (7.25 mg/mmol)($P < 0.05$), suggesting an attenuation of the acute phase response to persistent reperfusion injury. EG subjects' ACR levels were not significantly different to the CG at T3 and T4 indicating the possible normalisation of ACR levels post treatment. Analysis of CRP levels in the EG showed T4 (6 mg/l) being significantly higher than T2 (4 mg/l) levels indicating no lowering of any acute inflammatory response. T3 (EG 5 mg/l, CG 2 mg/l) and T4 (EG 6 mg/l, CG 2 mg/l) CRP levels were significantly higher in the EG than the CG). Results were analysed using Wilcoxon signed ranks test. Although the EG subjects increased both their CWD and MWD, it is not possible to suggest this is a result of a reduction in the acute phase response or an attenuation of inflammatory markers. Mechanisms into the benefits of improved exercise tolerance have yet to be fully elucidated. The continued use of exercise therapy may be an effective tool in the treatment of PAOD.

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Yudkin JS *et al.* (1988). *Lancet* **3**, 530–533.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC82

Thromboxane-induced Cl^- secretion in isolated human colorectum

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In isolated mucosa of the rat distal colon, we have found that endogenous thromboxane A_2 (TXA_2) released by anti-tumour drug irinotecan and 9, 11-epithio-11, 12-methano-thromboxane A_2 (STA_2), a stable TXA_2 analogue, induces Cl^- secretion (Sakai *et al.* 1997; 2002). But we do not yet know whether this function of TXA_2 is restricted only to rats or if it is also present in human. Herein we investigated the effect of STA_2 on the Cl^- secretion in isolated human colorectal mucosa.

In accordance with the recommendations of the Declaration of Helsinki, the specimens of the normal mucosa were obtained from surgical resection of patients having colorectal adenocarcinomas. Informed consents were obtained from all patients at Toyama Medical and Pharmaceutical University Hospital. Effects of the chemicals on the short-circuit current (*I*sc), the potential difference across the mucosa (Pd) and the tissue conductance (Gt) were examined in isolated human colorectal mucosa mounted on Ussing chamber. Data are shown as means ± S.E.M. Differences between groups were analysed by one-way ANOVA. Comparison between the two groups was made with paired *t* test.

When STA_2 (0.3 μM) was added to the serosal side, the *I*sc increased from 81 ± 11 to 170 ± 15 μA cm^{-2} ($n = 12$, $P < 0.01$), the Pd increased from 8.3 ± 0.9 to 13.0 ± 1.4 mV ($n = 10$, $P < 0.05$), and the Gt increased from 9.8 ± 0.4 to 14.0 ± 0.8 mS cm^{-2} ($n = 10$, $P < 0.01$). The effect of STA_2 was not apparently dependent on the location of colorectum (from cecum to rectum). The STA_2-induced effect was significantly inhibited by ONO-3708, a TXA_2 receptor antagonist (10 μM at the serosal side; $n = 4$, $P < 0.05$), NPPB, a Cl^- channel blocker (300 μM at the mucosal side; $n = 3$, $P < 0.05$), and furosemide (100 μM at the serosal side; $n = 4$, $P < 0.05$). In the low-Cl^- bathing solution, STA_2 increased the *I*sc only by 7 ± 4 μA cm^{-2} ($n = 4$). These results suggest that STA_2 induces Cl^- secretion via a TXA_2 receptor. The STA_2 (0.3 μM)-induced Cl^- secretion was inhibited by chromanol 293B (3 μM), a cAMP-dependent K^+ channel blocker. ($P < 0.01$; $n = 5$).

Our results suggest that the TXA_2 is an endogenous stimulant of the cAMP-mediated Cl^- secretion in human colorectal mucosa.

Sakai H *et al.* (1997). *J Physiol* **505**, 133–144.
Sakai H *et al.* (2002). *J Physiol* **543**, 261–271.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC83

Effect of heat acclimation on performance of intermittent, high intensity shuttle running in a hot environment

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It is well documented that heat acclimation of 6 or more sessions of at least 60 min duration prolongs the time to exhaustion during endurance walking, cycling and running in the heat. However, the effect of acclimation on prolonged high intensity intermittent running has not yet been investigated. High intensity intermittent exercise in the heat (35°C) has previously been demonstrated to provide a greater thermal strain than continuous exercise and may therefore be a more powerful stimulus for acclimation (Nevill *et al.* 1995). Furthermore, many acclimation studies have not included an equivalent training group in moderate conditions and/or a control group, which makes it difficult to determine the extent of the acclimation adaptations, compared with the training adaptations *per se.* The aim of the present study was to test the hypothesis that 4 high intensity intermittent running acclimation sessions will improve the performance of well-trained female games players during intermittent, high intensity shuttle running in a hot environment (30°C, 27% RH).

After Loughborough University ethical approval was obtained, seventeen well-trained female games players volunteered for the study. The impact of 4 short heat acclimation sessions (30–45 min of the Loughborough Intermittent Shuttle Test [LIST]; Nicholas *et al.* 2000) on high intensity intermittent running performance (LIST), in terms of distance run, was examined. Three groups were used, an acclimation group (30°C; $n = 6$), a moderate training group (18°C; $n = 6$) and a control group ($n = 5$) who did not complete any training between the main trials. The 4 acclimation or moderate training sessions were completed in a 10 day period prior to the post-acclimation trial. The acclimation and training sessions were interspersed with one or two rest days between each session within the 10 days. Data were analysed using a two-or three-way ANOVA with repeated measures over trial or trial and time. Data are presented as mean ± S.E.M.

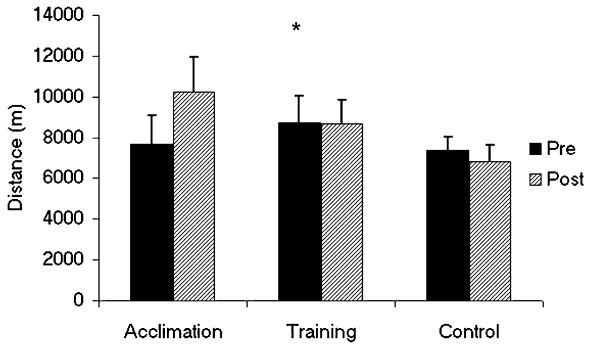

Figure 1. Distance completed during the main trials by the acclimation, training and control groups; *, interaction group × trial $P < 0.05$.

Exercise capacity was increased by 33 % in the acclimation group (interaction group × trial $P < 0.05$), but was unchanged in the moderate and control groups (Fig. 1). The acclimation group had a lower rectal temperature (interaction group × trial × time $P < 0.01$) and an increase in thermal comfort after acclimation (interaction group × trial $P < 0.01$). There was no difference in serum progesterone, aldosterone or cortisol concentrations following acclimation or between groups. There were no changes in estimated resting plasma volume or estimated sweat rate following acclimation. Therefore a lowering of deep body temperature and concomitant rise in thermal comfort may be responsible for the performance improvement.

Nevill ME *et al.* (1995). *J Physiol* **483P**, 124–125.
Nicholas CW *et al.* (2000). *J Sports Sci* **18**, 97–104.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC85

Prior sprint exercise does not speed pulmonary oxygen uptake kinetics during subsequent supra-maximal exercise in humans

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There is controversy concerning the factor(s) that limit the rate at which pulmonary oxygen uptake ($\dot{V}_{O_2}$) rises following the onset of exercise, with some groups favouring an intra-muscular metabolic inertia (e.g. Grassi *et al.* 1998) and others favouring an O_2 availability limitation (e.g. Hughson *et al.* 2001). The metabolic acidosis caused by the performance of prior heavy exercise results in enhanced vasodilatation and blood flow during subsequent exercise, and thus the prior exercise model represents a good test of whether muscle perfusion limits $\dot{V}_{O_2}$ kinetics following the onset of exercise. We therefore hypothesised that prior multiple-sprint exercise would reduce the time constant for the phase II $\dot{V}_{O_2}$ kinetics during subsequent supra-maximal intensity exercise (where there is evidence that O_2 availability may be compromised; Grassi *et al.* 2000).

Seven healthy males (age 20–33 yrs) gave written informed consent to participate in this study which was approved by the Manchester Metropolitan University Ethics Committee. On two separate occasions, the subjects performed square wave transitions from unloaded cycling to a work rate equivalent to ~105 % $\dot{V}_{O_2}$ peak following no prior exercise (control, C) and 12 min after the last bout of repeated sprint exercise (3 × 30 s maximal cycle ergometer sprints separated by 5 min rest, PSE). Pulmonary gas exchange was measured breath-by-breath and $\dot{V}_{O_2}$ kinetics were determined from the averaged response to each condition. The data were analysed using paired *t* tests and are expressed as the mean ± S.E.M. Statistical significance was accepted at $P < 0.05$.

Following the sprint exercise bouts, both pre-test heart rate (C: 98 ± 4 *vs.* PSE: 127 ± 4 b min^{-1}; $P < 0.01$) and pre-test blood [lactate] (C: 1.3 ± 0.1 *vs.* PSE: 7.7 ± 0.3 mM; $P < 0.01$) were significantly elevated. Near infra red spectroscopy also indicated a marked elevation in muscle oxygenation in PSE compared to C. However, despite indirect evidence that muscle blood flow and O_2 availability were enhanced, the phase II time constant was not significantly affected by prior sprint exercise (C: 33.8 ± 2.1 *vs.* PSE: 33.2 ± 2.9 s). The asymptotic gain of the fundamental $\dot{V}_{O_2}$ response (change in $\dot{V}_{O_2}$/change in work rate) (C: 8.1 ± 0.4 *vs.* PSE: 9.0 ± 0.3 ml min^{-1}.W^{-1}; $P < 0.05$), and the end-exercise $\dot{V}_{O_2}$

(C: 3.28 ± 0.15 *vs.* PSE: 3.53 ± 0.18 L min^{-1}; $P < 0.01$) were significantly elevated following sprint exercise. The performance of prior sprint exercise does not influence the time constant for the rise in $\dot{V}_{O_2}$ following the onset of subsequent supra-maximal exercise but does result in an increased asymptotic gain of the fundamental $\dot{V}_{O_2}$ response.

These data are consistent with the interpretation that the time constant for the rise in $\dot{V}_{O_2}$ following the onset of exercise is not limited by O_2 availability even in the transition to supra-maximal work rates (cf. Bangsbo *et al.* 2000). The cause of the higher initial 'target amplitude' for $\dot{V}_{O_2}$ following sprint exercise is unclear but may indicate that the gain of the fundamental $\dot{V}_{O_2}$ response is itself sensitive to O_2 availability or or to changes in muscle fibre recruitment.

Bangsbo J *et al.* (2000). *Am J Physiol* **279**, R899–906.

Grassi B *et al.* (1998). *J Appl Physiol* **85**, 1394–1403.

Grassi B *et al.* (2000). *J Appl Physiol* **89**, 1293–1301.

Hughson R *et al.* (2001). *Exerc Sport Sci Rev* **29**, 129–133.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC87

The force-velocity relation of human multi-joint movement: a study with force clamp analysis

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The force-velocity relation of muscle plays an important role in determining the performance of human movements. Studies on single-joint movements such as elbow flexion have shown that the relation between joint torque and angular velocity is described well with a classical, hyperbolic function (Hill equation). However, it remains unclear whether multi-joint movements, which are more closely related to our daily activities, are also characterized in a similar manner. The present study aimed: (1) to develop a dynamometer for human knee-hip extension movement, which can control precisely force and displacement, and (2) to determine the force-velocity relation of knee-hip extension movement in an isotonic condition.

Forty-one subjects (age 32.0 ± 15.1; height 160.8 ± 8.0; mass 54.1 ± 8.0, means ± S.D.) participated in the study. Written informed consent was obtained from all of the participants. This study was approved by the Ethical Committee for Human Experiments, University of Tokyo. The dynamometer consisted of a vertically placed tri-axial force plate, a servomotor and a computer-assisted control unit. Seated subjects kicked either bilaterally or unilaterally the force plate, the horizontal position of which was servo-controlled so that the measured force is matched with a force command at a time resolution of 2 ms (force clamp). The force command was made from the relation between maximal isometric force and foot position within the range between 70 and 90 % of "leg length" (longitudinal distance between sole of foot and hip joint), so that the same force relative to isometric force was consistently applied regardless of foot position (isotonic in terms of relative force). Electromyographic (EMG) recordings were made by using surface electrodes from seven major muscles: vastus medialis, vastus lateralis, rectus femoris, biceps femoris, semitendinosus, gluteus maximum, and gastrocnemius. EMG data were analysed using a one-way ANOVA with the Tukey post hoc test.

The force-velocity relation obtained was described by a linear function ($r^2 = 0.996$) more appropriately than a hyperbola within a range of force between ~0.1 to ~0.8 F_0 (F_0, maximal isometric force). The maximal force extrapolated from the linear regressions (Fext) coincided with F_0 (F_0/Fext = 0.982 ± 0.132). Also, the velocity at zero force (Vext) was obtained from the extrapolation. Compared to the bilateral movements, unilateral movements gave rise to the smaller Fext but the same Vext, suggesting that Vext is independent of force and therefore represents the proper unloaded velocity. The mean integrated electromyogram (mEMG) in each muscle was unchanged with force in the isotonic condition, whereas it was significantly smaller at F_0 than in the isotonic condition ($P < 0.05$) in knee extensor muscles. This suggests that some inhibitory mechanisms operate in knee extensor muscles when they generate large force (Westing *et al.* 1991). Such an inhibition may have an effect of moving the force-velocity relation downward as the force is large, thereby making the relation linear.

Westing SH *et al.* (1991). *Eur J Appl Physiol* **62**, 104–108.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC88

The effects of prolonged exercise in a hot environment on lipopolysaccharide (LPS)-stimulated monocyte TNF-α release in trained male cyclists

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Monocyte function has been reported to fall temporarily after prolonged strenuous exercise (Baum *et al.* 1997). Increases in plasma cortisol and prolactin concentration after prolonged strenuous exercise may be partly responsible for exercise-induced alterations in monocyte function (Woods, 2000). Prolonged exercise in hot conditions has been shown to elevate plasma cortisol (Mitchell *et al.* 2002), prolactin (Bridge *et al.* 2003) and the circulating monocyte count (Severs *et al.* 1996) to a greater extent than prolonged exercise in cool conditions. We therefore wished to examine the influence of prolonged strenuous exercise in hot and humid conditions on monocyte function (lipopolysaccharide (LPS)-stimulated TNF-α release) in endurance trained males.

With local ethics committee approval, thirteen well trained male cyclists (age 28 ± 2 years, body mass 75.4 ± 1.4 kg, $\dot{V}_{O_2,max}$ 61.2 ± 2.3 ml kg^{-1} min^{-1}, mean ± S.E.M.) volunteered to participate in the study. On two occasions, in random order and separated by one week, subjects reported to the laboratory at 1230 h following a 4 h fast. Subjects cycled for 2 h on a stationary ergometer at 55 % peak power output (194 ± 4 W) in an environmental chamber on one occasion at a temperature and humidity of 20.4 ± 0.1°C and 60 ± 1 % (CONTROL) and on another occasion 30.3 ± 0.1°C and 76 ± 1 % (HOT). Venous blood samples were collected at pre, post and 2 h post-exercise. Plasma cortisol, prolactin and TNF-α concentrations were determined using ELISA. Blood monocyte counts were performed using a Beckman Coulter counter. Monocyte function was assessed by measuring the plasma TNF-α concentration in unstimulated samples and after treatment of a 1 ml aliquot of whole blood with 50 μl LPS stimulant (10 mg ml^{-1}) for 1 h at 37°C. Results were analysed using a two-factor (trial × time) repeated measures ANOVA with post hoc

Tukey's HSD. Statistical significance was accepted at $P < 0.05$.

Final rectal temperatures were 38.1 ± 0.1°C (CONTROL) and 38.7 ± 0.1°C (HOT: $P < 0.01$). Plasma cortisol concentration increased post-exercise ($P < 0.01$) and was significantly higher on the HOT trial at post and 2 h post-exercise ($P < 0.01$). Plasma prolactin concentration increased post-exercise ($P < 0.01$) and was significantly higher on the HOT trial at post-exercise ($P < 0.01$). Circulating monocyte counts increased post-exercise ($P < 0.01$) and were significantly higher on the HOT trial at post and 2 h post-exercise (CONTROL: pre-exercise 0.4 ± 0.0, post-exercise 0.8 ± 0.1; HOT: pre-exercise 0.4 ± 0.0, post-exercise 0.9 ± 0.1 × 10^9 cells l^{-1}, $P < 0.01$). Plasma TNF-α concentration increased post-exercise ($P < 0.01$). LPS-stimulated TNF-α release increased 2 h post-exercise ($P < 0.01$). LPS-stimulated TNF-α release per monocyte decreased post-exercise (CONTROL: pre-exercise 3.4 ± 0.4, post-exercise 2.5 ± 0.4; HOT: pre-exercise 4.1 ± 0.7, post-exercise 1.7 ± 0.2 fg. $Cell^{-1}$, $P < 0.01$). The post-exercise decrease in LPS-stimulated TNF-α release per monocyte tended to be greater on the HOT trial but this did not reach statistical significance (interaction: $P = 0.06$ ANOVA).

These data suggest that prolonged strenuous exercise results in a decrease in LPS-stimulated monocyte TNF-α release (on a per cell basis) and that performing the exercise in hot and humid conditions does not significantly alter this response.

Baum M *et al.* (1997). *Eur J Appl Physiol* **76**, 165–169.
Bridge MW *et al.* (2003). *Exp Physiol* **88**, 627–635.
Mitchell JB *et al.* (2002). *Med Sci Sports Exerc* **34**, 1941–1950.
Severs Y *et al.* (1996). *Eur J Appl Physiol* **74**, 234–245.
Woods JA (2000). *Int J Sports Med* **21**, S24–30.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC89

Effects of inspiratory muscle training on critical power in trained cyclists

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Although inspiratory muscle training (IMT) improves cycling time-trial (TT) performance (Romer *et al.* 2002), mechanisms explaining such improvements remain unknown. Time-trial performance in competitive cyclists is strongly related to critical power (CP) (Smith *et al.* 1999). We questioned whether an increase in CP was a mechanism by which time-trial performance was improved in trained cyclists following a 6-wk IMT regimen.

Following local ethics committee approval and written informed consent 12 subjects were equally divided into either a pressure-threshold IMT group or a sham hypoxic placebo group. The IMT group performed 30 dynamic inspiratory efforts twice daily using a pressure-threshold device (POWERbreathe®), with the initial load set at 50 % baseline maximal inspiratory pressure (MIP). Placebo subjects used a sham hypoxic trainer 5 d wk^{-1} for 15-min (Sonetti *et al.* 2001). The power-time relationship, i.e., CP and anaerobic work capacity (AWC), was calculated using the power-1/time mathematical model (Whipp *et al.* 1982) following the completion of 3 fixed work-rate exhaustive trials. Prescribed powers were chosen to induce exercise intolerance within 3–10, 10–20, and 20–30 min.

An increase in MIP from (mean ± S.D.) 157.3 ± 12.3 to 179.1 ± 22.6 cmH_2O (+13.7 ± 9.2 %) was observed following IMT ($P < 0.05$, repeated measures ANOVA). No change was observed following placebo (pre *vs.* post: 171.2 ± 19.6 *vs.* 165 ± 15.9 cmH_2O). There was no change in CP (271 ± 66 *vs.* 269 ± 65 W) or AWC (25.2 ± 6.3 *vs.* 29.9 ± 9.7 kJ) following IMT (Fig. 1). Placebo also had no effect on CP (238 ± 25 *vs.* 236 ± 16 W) or AWC (34.5 ± 15.3 *vs.* 38.9 ± 17.4 kJ). Following IMT there was a non-significant reduction in min ventilation ($\dot{V}_E$) and an equivalent $\dot{V}_E$ was characterised by a higher tidal volume, an increase in mean inspiratory flow rate and a decrease in mean expiratory flow rate. However, these changes were not always accompanied by improvements in cycling endurance.

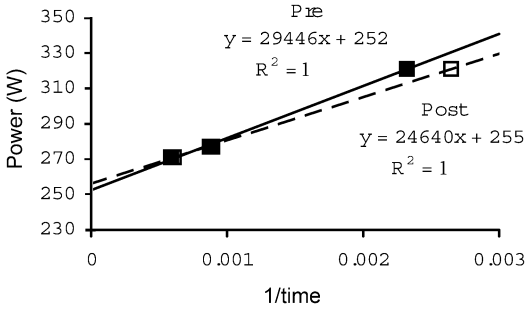

Figure 1. Effect of IMT on CP (y-intercept) and AWC (slope) in a representative subject. Pre-IMT (continuous line), post-IMT (dashed line).

These results suggest an increase in CP as an unlikely mechanism by which TT performance is improved in trained cyclists following IMT. We suggest other mechanisms are operative, such as placebo and test familiarisation effects, attenuated perceptual responses, an increase in the mechanical efficiency of ventilation, favourable changes in acid-base balance, and/or attenuated inspiratory muscle fatigue.

Romer LM *et al.* (2002). *Med Sci Sports Exerc* **34**, 785–792.
Smith JC *et al.* (1999). *Int J Sports Med* **20**, 374–378.
Sonetti DA *et al.* (2001). *Respir Physiol* **127**, 185–199.
Whipp BJ *et al.* (1982). *Fed Proc* **41**, 1591.

All procedures accord with current national and local guidelines

PC90

Paralysed human muscle expresses all three isoforms of IGF-I, which remain sensitive to increased mechanical activity

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Spinal cord injury and subsequent paralysis results in a loss of muscle mass, an infiltration of connective tissue and a shift towards fast myosin isoforms in the affected muscles. In contrast to disuse, muscle will adapt to increased use and overload through mechanisms regulated by locally expressed growth factors. In this regard three transcripts of the IGF-I gene have been shown to be expressed and upregulated in young and aged human skeletal muscle (Hameed *et al.* 2003*a,b*); IGF-IEa, IGF-IEb and IGF-IEc (also known as MGF). In the present study, undertaken with local ethics committee approval, we have

investigated whether paralysed tibialis anterior muscle firstly, expresses these growth factors and secondly, if so, to determine whether they remain sensitive to a severe exercise challenge imposed by chronic electrical stimulation training.

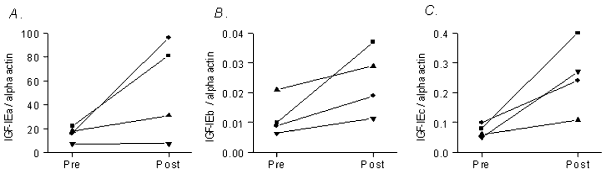

Figure 1. mRNA levels of IGF-I splice variants, normalised to α actin, before and after 4 weeks of electrical stimulation training. *A.* IGF-IEa, *B.* IGF-Eb and *C.* MGF.

Four spinal cord injured men (mean age 39 ± 9 S.D. years) completed 4 weeks of low frequency electrical stimulation training of the tibialis anterior muscle (2–6 h per day; at 10 Hz, 5 s on 5 s off under isometric loading conditions, see Harridge *et al.* 2002). Following local anaesthesia with 1 % lidocaine, biopsies of the tibialis anterior muscle were taken before and following 4 weeks of training. The mRNA levels of the three IGF-I splice variants were determined using real time quantitative PCR (Hameed *et al.* 2003a). Measurement of α actin mRNA levels showed no significant change as a result of the electrical stimulation training(13.1 ± 6.5 v 10.2 ± 7.0 ng mRNA 10^{-7} / μg RNA). The IGF-I mRNA data were normalised to these values to remove variability caused by connective tissue RNA contributing to total RNA.

All isoforms were expressed in the paralysed muscle and increased as a result of the electrical stimulation training protocol in each individual (Fig. 1). Although, statistically this only reached significance for MGF ($P < 0.05$, paired t test).

The data show that paralysed muscles can increase the expression of all three isoforms of IGF-I as a result of an exercise challenge. This is encouraging as it is becoming clear that the isoforms have important, but differing physiological roles both in muscle (such as stimulating protein synthesis and triggering the activation of satellite cells) and nerve repair.

Hameed *et al.* (2003*a*). *J Physiol* **547**, 247–254.

Hameed *et al.* (2003*b*). *J Physiol* **547.P**, C118.

Harridge *et al.* (2002). *Muscle & Nerve* **25**, 685–694.

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All procedures accord with current local guidelines and the Declaration of Helsinki

PC91

Intensity effects of supine exercise and recovery on vagal activity in man

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An imbalance of autonomic activity to the heart is a risk factor for adverse cardiovascular (CV) events that are potentially fatal. It is established that exercise training can alter autonomic activity thus be a treatment for those with CV disease. However, the alteration of the autonomic control of the CV system after an exercise bout has not been extensively researched. The effects of 3 intensities of supine exercise on cardiac autonomic control during supine recovery were studied using simple non-invasive time domain measures.

Following University of Essex ethics committee approval, in agreement with the Declaration of Helsinki, fourteen moderately fit subjects, (7 female), mean age 21.4 ± 3.9 years completed 3 intensities of supine cycling (Low Work (LW), 2 mM Blood Lactate (La); Moderate Work (MW), 3 mM La; and High Work (HW), 4 mM La) on 3 separate occasions. The intensities were calculated from La data obtained in an initial fitness test. Non-invasive ECG and blood pressure (BP) tracings were recorded continuously for 5 min of paced breathing, before (baseline) and post exercise (P) at P5, P15, P30, P45 and P65 min. The subjects remained in the supine position throughout all procedures. Heart rate variability was reported as the standard deviation of successive differences (SDSD) which gives an indication of vagal activity (Task Force, 1996).

A two-way ANOVA revealed that there were both time and intensity effects ($P \leqslant 0.05$) for R-R interval and SDSD. Post-hoc t tests with Bonferroni correction, revealed that R-R interval decreased significantly from baseline at P5 for all intensities −93.2 ms ± 91.6–179.8 ms ± 98.1, −237.3 ms ± 166.7 LW, MW and HW respectively, mean ± SD) and also at P15 for MW and HW intensities ($p \leqslant 0.01$). With LW R-R interval increased above baseline at P30 and the increase was graded with time. At all intensities SDSD decreased significantly at P5, at MW for P15, and at HW for P15 and P30 ($p \leqslant 0.01$), remaining below baseline for all time points except for P65 for MW where vagal activity rose above baseline in 9/14 subjects. When compared to HW this did not reach significance ($P = 0.062$). Diastolic BP was below baseline for all readings. Clearly there is a change in autonomic activity following an exercise bout. High intensity reveals a reduced vagal activity for up to 30 min during recovery. However, moderate exercise results in a small post-exercise increase in vagal activity after 1 h potentially providing some cardioprotection. This study may go some way to alluding to the cardiac benefits following moderate intensity exercise.

Task Force European Society of Cardiology & Nort (1996). *Circulation* **93**, 1043–1065.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC92

The effects of carbohydrate and vitamin C ingestion on plasma cortisol and blood neutrophil responses to prolonged cycling

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Increased susceptibility to infection may arise after endurance exercise owing to an exercise-induced suppression of immune system function. Carbohydrate (CHO) ingestion during exercise attenuates this immunosuppression (Gleeson *et al.* 2000). Supplementation with a high dose of vitamin C (VC) for a period of 3 weeks may also be beneficial, by increasing plasma VC concentration and so reducing the exercise-induced release of VC and cortisol from the adrenals (Peters *et al.* 2001) and/or by improving antioxidant defence. The aim of the present study was to investigate the effect of CHO, with or without VC, consumed shortly before and during exercise on the plasma cortisol and blood neutrophil functional responses.

With local ethics committee approval, 7 healthy men (age 26 ± 2 years, body mass 71.7 ± 2.4 kg, $\dot{V}_{O_2max}$ 55.2 ± 1.7 ml kg^{-1} min^{-1}; means ± S.E.M.) participated in the study. After determination of $\dot{V}_{O_2max}$ and completing a habituation ride they completed three

main trials (2.5 h cycling at 60 % $\dot{V}_{O_2max}$). In a single blind, counterbalanced-crossover design they consumed either a placebo (P), 6 % w/v CHO (C) or 6 % w/v CHO with 0.15 % w/v VC (V) drink in the following volumes: 5 ml kg^{-1} body mass 1 h before and 2.5 ml kg^{-1} body mass every 15 min during exercise. Venous blood samples were taken before the first drink (Pre-1), immediately pre-and post-exercise (Pre-0 and Post-0, respectively), and 1 h post-exercise (Post-1). Haematological analysis was performed using an automated cell counter (Beckman Coulter, UK). Plasma cortisol concentration was determined using an ELISA kit. The neutrophil degranulation response (elastase release) to lipopolysaccharide was determined on a per cell basis according to Robson *et al.* (1999). Results were analysed using a 2-factor repeated measures ANOVA with Tukey post hoc test used where appropriate.

The results (Table 1) support previous findings which suggest that CHO beverage consumption before and during prolonged exercise attenuates the post-exercise changes in immune cell function, probably by maintaining blood glucose and attenuating adrenal cortisol release (Gleeson *et al.* 2000). However, it appears that the addition of VC to CHO provides no additional benefit. In fact there is a trend for post-exercise (Post-0 and Post-1) neutrophil degranulation to be lower with V compared to C, which is supported by high estimates of effect size (0.429 and 0.447, respectively).

Table 1: Changes in plasma cortisol and blood neutrophil function.

		Pre-1	Pre-0	Post-0	Post-1
Cortisol	**P**	445 (62)	361 (41)	947 (122)**	783 (138)
(nM)	**C**	493 (61)	424 (74)*	562 (99)¶	385 (70)¶
	V	490 (57)	355 (46)**	503 (95)¶¶	383 (65)¶¶
Elastase release (% of Pre-1)	**P**	100 (0)	100 (9)	38 (9)**	37 (8)**
	C	100 (0)	100 (8)	92 (18)¶¶	90 (18)¶¶
	V	100 (0)	96 (8)	69 (9)*¶	60 (11)*

Values are means (± S.E.M., n = 7).
Significantly different from Pre-**1** (*$P < 0.05$; ** $P < 0.01$),
Significantly different from P (¶ $P < 0.05$; ¶¶ $P < 0.01$),
There were no significant differences between C and V.

Gleeson M *et al.* (2000). *Int J Sports Med* **21**, S44–S50.
Peters EM *et al.* (2001). *Int J Sports Med* **22**, 120–126.
Robson PJ *et al.* (1999). *Int J Sports Med* **20**, 128–135.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC93

Atrial natriuretic peptide and central venous pressure during rowing and running

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Rowing involves significant contribution from both the upper and lower body and is performed in a sitting position, while in running the body position is more erect and the legs are predominantly used. Active muscles propel blood towards the heart, while posture attenuates the central blood volume (van Lieshout *et al.* 2001). Thus, a lower heart rate (HR) and a larger oxygen pulse, as an index of the stroke volume of the heart, during rowing than during running could indicate that the central blood volume is the largest (Yoshiga & Higuchi 2002). We hypothesized that central venous pressure (CVP) and plasma atrial natriuretic peptide (ANP; Perko *et al.* 1994) would be higher during rowing than during running.

Six men and three women (age, mean ± S.D. 26 ± 3 years) participated in this study as approved by the Ethics Committee of Copenhagen (KF 01-186/2). The subjects performed two randomly ordered exercise trials of running and rowing. Comparison across exercise mode and intensity were evaluated by multiple analyses of variance with Newman & Cruise post-hoc validation. A P value of < 0.05 was considered statistically signficant.

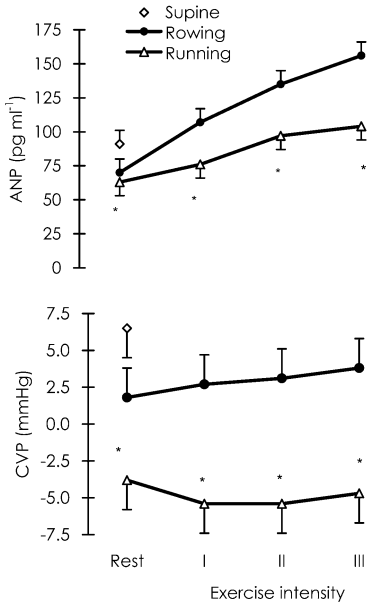

Figure 1. Atrial natriuretic peptide and central venous pressure during rowing and running. Values are means ± S.D. *, Different from rowing, $P < 0.05$.

Each exercise trial included three intensities aiming at a HR of 120, 140, and 160 beat min^{-1}, respectively. Thus, the average HR was similar between rowing and running (mean ± S.D. 138 ± 17 *vs.* 139 ± 18 beat min^{-1}). Yet, there was a larger average oxygen uptake and min ventilation during rowing than during running (2.5 ± 0.9 *vs.* 2.3 ± 0.8 l min^{-1} and 54 ± 23 *vs.* 49 ± 17 l min^{-1}, $P < 0.05$). MAP was similar between rowing and running (97 ± 1 vs. 92 ± 1 mmHg). There were enhanced CVP (2.6 ± 0.2 vs. −5.2 ± 0.3 mmHg, $P < 0.001$) and ANP (131 ± 23 vs. 106 ± 26 pg ml^{-1}, $P < 0.01$) during rowing compared with running. .

In conclusion, there was an enhanced central blood volume during rowing, as assessed by both plasma ANP and CVP, compared with running. These results support the suggestion that, during exercise, heart rate is influenced by the central blood volume.

Perko G *et al.* (1994). *Acta Physiol Scand* **150**, 494–454.
van Lieshout JJ *et al.* (2001). *Stroke* **32**, 1546–1551.
Yoshiga CC & Higuchi M (2002). *Eur J Appl Physiol* **87**, 97–100.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC94

Human bone, but not muscle, collagen synthesis is acutely responsive to feeding

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Collagen is the most abundant protein in the human body, fulfilling a wide range of mechanical and structural roles. Despite our considerable knowledge of its biochemistry and cell biology, very little is known about the physiology of collagen. Using a novel method to directly measure collagen synthesis using stable isotope tracers (Babraj *et al* 2002), we have previously demonstrated that collagen in human bone, tendon, ligament and muscle are synthesized at rates which are much faster than hitherto expected (Babraj *et al* 2003) and that muscle collagen (mostly endomysium) shows remarkable acute anabolic responses to strenuous exercise with a pattern and extent similar to that of human myofibrillar protein. We therefore hypothesized that as myofibrillar protein is also acutely stimulated by feeding protein or amino acids, both muscle and bone collagen synthesis might also be.

We studied four groups each of 4 healthy young men, (all 29 ± 3.5 y, BMI 24.3 ± 2.1, kg/m^2 mean ± S.E.M., with no significant differences between the groups). The studies had the approval of the Tayside Ethics Committee. In two groups of subjects we measured muscle myofibrillar protein synthesis and collagen synthesis and in two groups we measured bone collagen synthesis in the post absorptive and fed states. Subjects in the muscle group were given an infusion of octreotide and insulin throughout to maintain insulin (~10 mIU/l) and glucose (~4–5 mM) at the post-absorptive values and either ingested 20 g of essential amino acids or remained post-absorptive. Subjects in the bone group were either given a venous infusion of glucose, lipid and amino acids (equivalent to 1 × basal metabolic rate, calculated using Harris-Benedict equation) or remained post-absorptive. Quadriceps muscle and iliac crest were biopsied using the conchotome and bone needle techniques applied under local anaesthesia (1 % lignocaine). Bone collagen synthesis was measured, using established methods (Babraj *et al.* 2002) as the incorporation over 2 h of [1-^{13}C]proline applied as a flooding dose. Muscle collagen synthesis was measured as the incorporation over 3 h of [1-^{13}C]keto-isocaproic acid applied as a primed constant infusion.

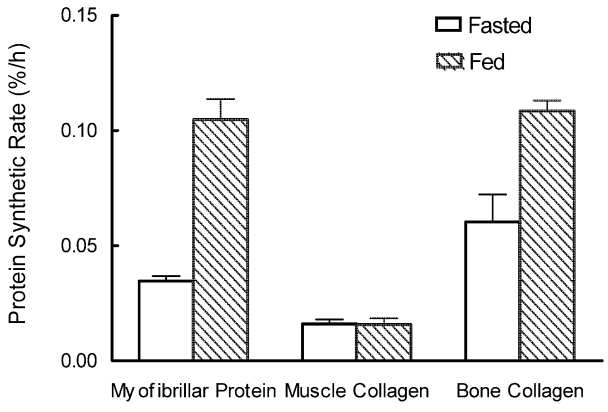

Figure 1. Responses of myofibrillar protein, endomysial collagen and bone collagen synthesis to feeding.

Although myofibrillar protein responded as expected, rising from 0.035 ± 0.002 to 0.105 ± 0.009 %/h within 3 h of feeding, muscle collagen synthesis was unchanged at 0.016 ± 0.002 %/h. In bone, however, the synthetic rate of the hot water extractable fraction (which contains 90 % of bone collagen) rose from 0.060 ± 0.012 to 0.108 ± 0.004 %/h on feeding.

The results suggest that nutritional modulation of bone collagen synthesis is rapid and marked, features which are likely to be of importance in the growth and maintenance of bone mass and bone quality.

Babraj J *et al.* (2002). *Biochem Soc Trans* **30**, 61–65.

Babraj *et al.* (2003). *J Physiol* **547.P**, PC 14.

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All procedures accord with current local guidelines and the Declaration of Helsinki

PC95

Prolonged ascorbic acid supplementation attenuates post-exercise lipid peroxidation but has no effect on delayed onset muscle soreness following downhill running in man

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Exercise involving lengthening muscle actions results in delayed onset muscle soreness (DOMS) which may be attributable to oxidative stress (OxS). Although exercise causes oxidant stress any link between OxS and DOMS remains equivocal. Ascorbic acid (AA) is a dietary antioxidant and a powerful inhibitor of lipid peroxidation. Given the antioxidant properties of AA, the aim of this study was to investigate the effects of AA supplementation on DOMS, muscle function and lipid peroxidation following downhill running.

In a double blind placebo controlled study, twenty male subjects were randomly assigned to two groups. Group A received 1 g of ascorbate 2 h pre and for 14-days post exercise, whilst group Pl received a placebo. Exercise involved running downhill on a motorised treadmill for 30-minutes at 60 % $\dot{V}_{O_2max}$. Venous blood samples were drawn pre and 2 h post-supplement, immediately post-exercise, and then 24, 96, and 308 h post-exercise for the analysis of creatine kinase (CK) using enzymatic methods, plasma ascorbate and malonaldehyde (MDA) using HPLC. DOMS was assessed using a visual analogue scale, whilst muscle function was assessed using an isokinetic dynamometer. Statistical analysis was carried out using a 2-way ANOVA.

		Pre	+24 h	+96 h	+308 h
DOMS	**Pl**	0.0 (0.0)	33.5* (4.5)	8.1* (3.7)	0.0 (1.6)
	A	0.0 (0.0)	29.8* (6.0)	12.8* (5.2)	0.6 (2.8)
MDA	**Pl**	0.86 (0.0)	0.92 (0.1)	1.13* (0.1)	0.89 (0.0)
	A	0.82 (0.0)	0.90 (0.1)	0.91$^{\$}$ (0.1)	0.93 (0.1)
CK	**Pl**	113 (37.3)	899* (36.2)	976* (69.3)	118 (56.3)
	A	99 (21.3)	830* (29.3)	281$^{\$}$ (92.8)	96 (17.4)
Leg torque	**Pl**	0.0 (0.0)	-24.3* (4.5)	-5.6 (3.7)	4.1 (1.6)
	A	0.0 (0.0)	-24.7* (6.0)	-7.0 (5.2)	-10.4$^{\$}$ (2.8)

Table 1. Mean ratings of DOMS (arbitrary units), MDA

(μmol l^{-1}), CK (U l^{-1}) and concentric leg torque at 1.04 rad sec^{-1} (% change before and after the run) (* indicates significant difference from pre-exercise, $ indicates significant difference from placebo).

AA concentrations were elevated in A compared to Pl ($P < 0.05$). Downhill running resulted in a significant increase in DOMS ($P < 0.05$) although there was no difference between the groups ($P > 0.05$). Muscle function was impaired post exercise in A and Pl ($P < 0.05$). Both groups exhibited an increase in plasma CK 24 h post-exercise ($P < 0.05$) whilst Pl exhibited a second peak 96 h post-exercise ($P < 0.05$). There was no increase in MDA immediately post-exercise in both A and Pl, although Pl exhibited a delayed increase 96 h post-exercise ($P < 0.05$).

Our data suggest that ascorbate supplementation attenuates MDA production and secondary increases in CK activity following downhill running although it does not reduce DOMS and may affect the recovery of muscle function.

The authors would like to acknowledge Lee Siemesko for assistance with the data collection and Dr Anne McArdle from Liverpool University for useful scientific discussions.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC96

Metabolic responses to duathlon performance following pre-exercise meals

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Carbohydrate intake prior to exercise has been shown to be beneficial, although recently the intake of lipids has also proved advantageous. The effect of high carbohydrate (CHO) and high fat (LCHO) isoenergetic pre-exercise meals on the metabolic responses to duathlon performance were investigated.

Nine male subjects of mean age 28.6±5.9 yrs, body mass 76.8 ± 8.5 kg and height 1.80 ± 0.7m performed simulated self-paced duathlon time trials (5km run, 30km cycle, 5km run) following three pre-exercise dietary conditions. Subjects fasted overnight and were randomly given isoenergetic meals of either a high CHO (214.8g) or low CHO (50.0 g) or fasted (F) at breakfast 3–4 h prior to exercise. Respiratory gas analysis took place at the start and end of each run stage and at the start, mid point and end of the cycle stage to determine CHO and fat oxidation rates. Blood samples were taken prior to and after each exercise stage to determine metabolite responses. All data were analysed using a general linear model ANOVA with repeated measures and means were compared using the Bonferroni confidence interval.

Table 1.Concentration of metabolites

		Pre-ex.	Transition 1	Transition 2	Post-ex.
NEFA	F	0.37±0.2	0.89±0.39	1.05±0.35	1.62±0.14
(mmol l^{-1})	CHO	0.13±0.20	0.42±0.41	0.81±0.35	1.20±0.23
	LCHO	0.46±0.18	1.05±0.22	1.37±0.40	1.83±0.43
Glycerol	F	62.1±11.9	231.4±58.4	273.8±105.4	403.8±120.4
(μmol l^{-1})	CHO	44.2±18.3	132.3±53.9	205.2±43.6	302.3±76.2
	LCHO	96.1±31.6	266.6±57.9	325.0±108.0	409.6±114.9
Glucose	F	5.0±0.4	7.3±1.2	4.8±0.5	4.96±0.70
(mmol l^{-1})	CHO	4.6±0.5	6.2±1.6	4.7±0.4	4.90±0.21
	LCHO	5.1±0.2	6.7±1.2	5.0±0.7	4.99±0.51

Duathlon performance (6.56 ± 0.39 m s^{-1}, 6.36 ± 0.45 m s^{-1}, 6.54 ± 0.44 m s^{-1} for the F, CHO and LCHO respectively) was not significantly different as a result of the pre-exercise meal strategies ($P > 0.05$). CHO oxidation rate was significantly higher and fat oxidation rate was significantly lower in CHO compared to LCHO and F ($P < 0.05$). Non-esterified fatty acid (NEFA) and glycerol were significantly lower in CHO ($P < 0.05$), though no differences were observed for glucose ($P > 0.05$). Table 1 shows the metabolic differences between the conditions.

These data emphasise the availability and oxidation of energy substrates is altered during duathlon by the manipulation of pre-exercise macronutrient intake, although no effects on overall performance are observed.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC97

Cortical excitability after voluntary and electrically stimulated muscle activity of the wrist extensors of normal subjects

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The adult central nervous system has been shown to be capable of significant plasticity. Electrical stimulation of the somatosensory pathway has increased cortical excitability in both normal subjects (Ridding & Rothwell, 1999; Ridding *et al.* 2000; McKay *et al.* 2002; Khaslavskaia *et al.* 2002) and patients with brain lesions (Fraser *et al.* 2002). However, a number of stimulation patterns have been used and the relationship between these and the extent of cortical change is unknown. There have been no studies of the differential effects of voluntary and different patterns of electrically stimulated muscle activity on cortical excitability. We investigated this in normal subjects.

With local ethical approval the wrist extensor muscles of 5 healthy subjects (22–57 years, 3 females) were activated for 60 min on 5 occasions at least one week apart using different patterns of activity. Passive stimulation was performed at 5 Hz, 300μs (Fraser *et al.* 2002); 25 Hz, 300 μs (Powell *et al.* 1999) and 10 Hz train of 1 ms pulses for 500 ms s^{-1} (Ridding *et al.* 2000). EMG triggered stimulation was performed at 25 Hz, 300 μs (Cauraugh *et al.* 2000) and voluntary exercise for 5 s every 25 s. Motor evoked potentials (MEPs) of the same muscles were elicited by transcranial magnetic stimulation (1.2 times threshold) at 10 min intervals before, during and for 20 min after muscle activity. The amplitude of 10 averaged MEPs was measured. The mean MEP amplitude before stimulation was 1.35 mV (S.D. 0.71 mV). None of the interventions caused a significant change in the MEP amplitude at any time and there were no significant differences between them.

These results indicate that a single 60 min episode of voluntary or stimulated activity of the wrist extensors has no effect on cortical excitability in normal adult brains regardless of the stimulation pattern. This is in contrast to the reported increase in cortical excitability in the pharyngeal muscles after only 10 min (Fraser *et al.* 2002). It remains to be seen whether brains recovering from neurological insults respond differently.

Cauraugh J *et al.* (2000). *Stroke* **31**, 1360–1364.

Fraser C *et al.* (2002). *Neuron* **34**, 831–40.

Khaslavskaias S *et al.* (2002). *Exptl Brain Res* **145**, 309–315.

McKay DR *et al.* (2002). *Exptl Brain Res* **143**, 342–349.

Powell J *et al.* (1999). *Stroke* **30**, 1384–1389.

Ridding MC *et al.* (2000). *Exptl Brain Res* **131** , 135–143.

Ridding MC & Rothwell JC (1999). *Exptl Brain Res* **126**, 536–544

Isaac Sorinola is sponsored by the Association of Commonwealth Universities PhD studentship

All procedures accord with current local guidelines and the Declaration of Helsinki

PC98

Hydration and energy provision during soccer-specific exercise

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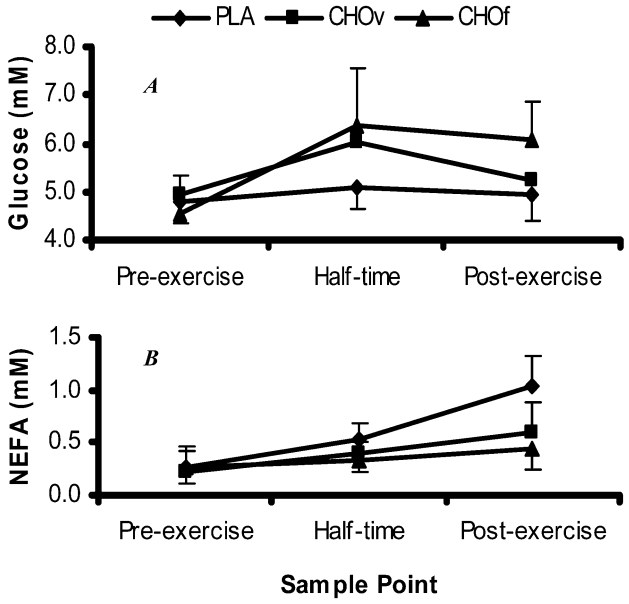
During soccer play there is a net depletion of muscle glycogen and players may lose 2–3 L of sweat. Therefore there are opportunities for enhancing performance during a game by adopting refuelling and rehydration regimes. The present aim was to manipulate the provision of sports drinks during soccer-specific exercise and to investigate the effect on metabolic responses and on components of performance.

Twelve male soccer players of mean (± S.D.) age 24.5 (± 3) y; height 1.77 (± 0.1) m; body mass 74.5 (± 7) kg; $\dot{V}_{O_2max}$ 59.37 (± 7) ml kg^{-1} min^{-1} performed a soccer-specific protocol, incorporating 3 s-s sprints on a non-motorised treadmill (Drust *et al.* 2000) after providing written informed consent. On two occasions either 7 ml kg^{-1} BM of carbohydrate-electrolyte (CHOv) or placebo (PLA) solution was ingested before and at half-time (532 ± 38 ml; total 1065 ± 76 ml). On a third occasion the same volume of carbohydrate-electrolyte solution was consumed (CHOf) but in smaller volumes at 0, 15, 30, half-time, 60, 75 min (178 ± 13 ml). Blood samples were collected at rest, half-time and full-time and analysed for glucose and Non-esterified Free Fatty Acids (NEFA). Respiratory analyses were undertaken throughout to determine the rate of carbohydrate oxidation, as was 3-s sprint power. Trials were performed in a double-blind counter-balanced manner. Repeated measures ANOVAs were used with significance at $P < 0.05$.



Plasma glucose (Fig. 1A) and carbohydrate oxidation (Table 1) were higher ($P < 0.05$) during CHOf compared with PLA. The concentration of NEFA (Fig. 1B) was reduced ($P < 0.05$) with CHOv and CHOf compared with PLA.

Table 1: Carbohydrate oxidation (g·min^{-1}).

	15 min	30 min	45 min	60 min	75 min	90 min
PLA	1.91±0.3	1.82±0.4	1.69±0.5	1.59±0.4	1.69±0.3	1.64±0.4
CHOv	2.08±0.4	1.91±0.3	1.92±0.5	1.96±0.5	2.01±0.5	1.92±0.6
CHOf	2.09±0.4	2.05±0.4	1.98±0.4	2.10±0.6	2.23±0.6	1.97±0.4

Mean sprint power was not affected ($P > 0.05$) by the experimental treatments (PLA: 1080.42 ± 241 W; CHOv: 1103.67 ± 228 W; CHOf: 1090.59 ± 136 W).Ingesting carbohydrate-electrolyte solution significantly affected plasma metabolites and increased carbohydrate oxidation but failed to impact on performance of short sprints during soccer-specific exercise. Furthermore, the timing and volume of ingestion did not significantly affect metabolism or sprint power.

Drust B *et al.* (2000). *Eur J Appl Physiol* **81**, 11–17.

This study was sponsored by GSK.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC99

Effect of wearing protective clothing and self contained breathing apparatus on heart rate, temperature and oxygen consumption.

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Fire fighters must possess the ability to respond to both extrinsic stress and stress from wearing protective clothing (PC) and self-contained breathing apparatus (SCBA) (White *et al*, 1991, Richardson & Capra, 2001). The effects of wearing PC+SCBA (20.42 ± 1.5 kg mean ± S.D.) on heart rate (HR), temperature responses and oxygen cost in six subjects (age 20.3 ± 0.8 years, weight 77.7 ± 7.0 kg and height 180.3 ± 4.3 cm) were observed. Ethical approval was obtained from the University College Chester Ethics Committee and the Health and Safety Officer from Greater Manchester Fire Service.

Table 1 –Mean HR (bpm) responses to the CST whilst dressed in GK, WGK and PC+SCBA

CST Level	Gym Kit	Gym Kit and Weighted Rucksack	PC+SCBA
	HR (bpm)	HR (bpm)	HR (bpm)
1 (Low)	106 (±4.1)	123 (±7.6)	126 (±7.8)
5 (High)	163 (±3.7)	185 (±6.6)	188 (±5.6)

There were significant increases in HR when carrying out the Chester Step Test (CST) (Sykes, 1995) wearing gym kit (GK), gym kit and weighted rucksack (WGK) (weighted to PC+SCBA equivalent) and PC+SCBA (thermoneutral conditions) (Table 1). Data was analysed using a one way ANOVA with post hoc Tukey analysis. Significant increases ($P < 0.05$) at CST level 5 were observed between GK and WGK for HR (Δ 23.3 ± 5.8bpm) and GK and SCBA+PC for HR (Δ 25.2 ± 5.2bpm) and for O_2 cost (Δ 6.1 ± 3.8 ml O_2 kg^{-1} min^{-1}). Thus, cardiovascular responses are elicited both from the workload and weight of the PC+SCBA (Table 1). Skin temperature significantly increased ($P < 0.05$) between GK and PC+SCBA (Δ 3.1 ± 1.3°C) and also WGK and PC+SCBA (Δ 3.5 ± 1.7°C). This may suggest therefore that the

PC further increases the stress already elicited by the weight of the PC+SCBA. The results suggest that being able to dissipate heat from the PC may be imperative to reduce the stress placed on fire fighter training instructors during training exercises.

Sykes K (1995). *J Occ Health* Jan, 20–22.
Richardson JE & Capra MF (2001). *J Occ Environ Med* **43**, 1064–1072.
White MK *et al.* (1991). *Ergonomics* **34**, 445–457.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC102

Changes in human saliva and serum immunoglobulins during the Flora 1000-mile Challenge

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An increased incidence of upper respiratory tract infections is often reported by individuals undertaking prolonged strenuous exercise or periods of heavy training (Gleeson, 2000; Mackinnon, 2000). It has been suggested that falls in salivary immunoglobulin A (IgA) may be a causal factor but it is not generally known if changes in mucosal secretory IgA reflect similar changes in circulating levels of IgA. The present study reports changes in salivary and serum immunoglobulins in individuals who completed the Flora 1000-mile Challenge. This was a repeat of the feat of endurance completed by Captain Robert Barclay in 1809 in which he covered, on foot, one mile every h for 1000 consecutive hours. This was, therefore, a challenge of endurance combined with frequently interrupted sleep. He successfully completed this feat (and won a bet of £16, 000) by covering 2 miles at a time – one mile at the end of one h and one mile at the start of the next. This allowed him the maximum rest period of anything up to 105 min depending on how fast he walked or ran. In the present study 6 individuals attempted to repeat this feat; 5 completed it and 4 agreed to provide saliva samples and blood samples at 0, 250, 500, 750 and 910 miles.

Following approval by the East London and the City Research Ethics Committee, two healthy endurance-trained men (aged 25 and 56 years, body mass 81 and 82 kg, respectively) and two healthy endurance-trained women (aged 31 and 39 years, body mass both 50 kg) completed 1000 miles in 1000 h by adopting the same strategy as Barclay. The event began at 4 p.m. on 2nd March 2003 and ended 42 days later. The course was the same as that used for the 2003 London Marathon. Unstimulated saliva samples were obtained at rest before the Challenge and at rest (between exercise bouts) after completing 250, 500, 750 and 910 miles. Venous blood samples were obtained at 0, 500 and 910 miles. Saliva was analysed for IgA, amylase and total protein (Walsh *et al.* 1999) and serum samples were analysed for IgA, IgG, IgM, albumin, total protein and creatine kinase (CK). Various haematological measures including a differential leukocyte count and lymphocyte subsets were also recorded.

At baseline (0 miles), saliva IgA concentration was 141 (128–160) mg l^{-1} (median and range). Saliva IgA concentration declined progressively during the 1000-mile challenge and was 84 (53–137), 75 (67–99), 47 (35–89) and 59 (39–81) mg l^{-1} after completion of 250, 500, 750 and 910 miles, respectively. Saliva amylase activity and total protein also declined during the challenge (Table 1). In contrast, serum immunoglobulins (IgA, IgG and IgM), albumin and total protein concentrations remained relatively stable (Table 1). Leukocyte and lymphocyte subsets were virtually unchanged at 910 miles compared with baseline. Substantial elevations in circulating T (CD3+) cells, B (CD19+) cells and NK (CD3-CD56+) cells observed in 3 of the subjects at the 500-mile point.

Table 1. Salivary, serum and whole blood variables at 0 and 910 miles of the Flora 1000-mile Challenge (values are median and range, n=4)

	0 miles	910 miles
Saliva IgA (mg.l^{-1})	141 (128-160)	59 (39-81)
Saliva Amylase (U.l^{-1})	1460 (284-1846)	597 (215-1172)
Saliva Total Protein (mg.l^{-1})	1047 (923-1130)	596 (327-936)
Serum IgA (g.l^{-1})	2.2 (1.1-4.3)	1.9 (1.2-3.4)
Serum IgG (g.l^{-1})	9.6 (7.0-10.7)	9.0 (7.9-10.0)
Serum IgM (g.l^{-1})	0.83 (0.44-1.08)	0.75 (0.41-0.95)
Serum Albumin (g.l^{-1})	43 (42-50)	45 (43-46)
Serum Total Protein (g.l^{-1})	71 (68-73)	70 (68-71)
Serum CK (U.l^{-1})	160 (84-192)	806 (530-1014)

These results indicate that salivary IgA concentration was decreased after 7 weeks of the challenge. In contrast, serum IgA concentration remained relatively stable.

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PC104

The human coronary baroreflex during isolated coronary perfusion, ventricular fibrillation and cardio-pulmonary bypass

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In addition to the classical carotid and aortic baroreceptors, experiments in animals have demonstrated the existence of baroreceptors in the coronary arteries. Coronary baroreceptors have an effect on systemic vascular resistance (SVR) similar in magnitude to carotid baroreceptors (Drinkhill *et al.* 2001, Wright *et al.* 2000; Bennetts *et al.* 2002). Whilst the importance of carotid baroreceptors in blood pressure control is widely recognised in man the contribution of the coronary baroreceptors is unknown. The aim of this study was to determine whether evidence of the existence of coronary baroreceptor reflex could be obtained in man during cardiac surgery. All procedures were carried out with the approval of local ethical committee.

A human model was designed using cardiopulmonary bypass and modified use of a single pass blood cardioplegia device following aortic cross-clamping and ventricular fibrillation during open heart surgery for mitral valve replacement. Excluded from the study were those patients with coronary artery disease, peripheral vascular disease, diabetes and aortic valve incompetence. The selected patients all had normal coronary anatomy as determined angiographically. Anaesthesia involved temazopam premedication, alfentanil, etomidate and pancuronium induction and propofol and isoflurane maintenance. The proximal ascending aorta and coronary arteries were perfused through the cardioplegia device. The

systemic circulation perfused at constant blood flow from the heart-lung machine. Blood temperature was 32°C for the first six patients and 37°C for the next four. Changes in mean systemic blood pressure provided a measure of SVR. Coronary sinus blood samples were measured for troponin T, lactate and oxygen saturation to identify any myocardial ischaemia. Patients were randomly selected to have their coronary pressure held high (80 mmHg) for 90 seconds then reduced to low pressure (50 mmHg) for a further 90 seconds (H-L) or receive the reverse order of coronary pressure change (L-H). SVR during 30 second segments of coronary perfusion were calculated and compared with the SVR immediately before the change in coronary pressure.

Ten patients were initially recruited to the study, three were subsequently excluded due to technical problems or blood pressure instability during the experimental procedure. Increasing the coronary sinus pressure from 50 mmHg to 80 mmHg in 3 patients resulted in a decrease in vascular resistance in each patient (−10.1% mean, range −5.9 to −18.7). In four patients coronary pressure was decreased from 80 mmHg to 50mmHg. SVR increased in three patients (4.8% mean, range 1.4 to 7.9) but decreased in one −15.8 %). The values of troponin T, lactate and oxygen saturation were not different during the two states of coronary pressure. These preliminary results would suggest that coronary baroreceptors exist in man and result in reflex vascular responses similar to those previously reported in animal models.

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PC105

Striking differences in motor system oscillatory activity of a deafferented subject compared with healthy controls

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Neurons within the sensorimotor cortex of monkeys and humans are known to have synchronous oscillatory activity in the 15–30 Hz range. Furthermore, this oscillatory activity is known to influence descending motor commands to the contralateral hand muscles (Conway *et al.* 1995; Baker *et al.* 1997). Such coherence in the 15–30 Hz range between sensorimotor cortex and contralateral muscle EMG and between different hand muscles exhibits task-dependent modulations; the coupling is abolished during finger movements and strongest during steady hold periods just following movement (Kilner *et al.* 2000). Previously we have speculated that levels of coherence might reflect important changes in sensorimotor state encompassing alterations in both grip force and digit position, and hypothesised that the level of coherence in the 15–30 Hz range is modulated by sensory afferent inputs from the hand (Kilner *et al.* 2000). The current study tested this hypothesis.

We measured the activity of four hand muscles in ten healthy human subjects and in a somatosencory deafferented patient, GL, whilst they performed a precision grip task. All subjects gave informed consent and the study had local ethical committee approval. The coupling between the muscles as a function of the task was subsequently estimated using coherence analysis.

The study demonstrated a significant difference ($P < 0.05$ Crawford modified t test) in the degree of muscle-muscle coherence during the second hold period: healthy subjects all showed robust 15–30 Hz coherence, while this was at a very low level in the deafferented subject. We have previously argued that oscillatory synchrony may characterise a low-level control system which engages and then maintains the particular level of activity in the large number of synergistic muscles that are needed to exert efficient grip between the digits. Such a control system would be highly sensitive to changes in finger position signalled by cutaneous, joint and muscle afferents.

The present results suggest that this oscillatory control system cannot exist in the complete absence of somatosensory afferent inputs. These afferent inputs may normally serve to modulate or even be involved in the generation of 15–30 Hz oscillations in the motor system.

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All procedures accord with current local guidelines and the Declaration of Helsinki

PC106

In vivo human torque-angle relation and its determinants: changes following strength training in old age

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Ageing-induced reductions in maximal muscle strength diminish the reserve capacity available to perform tasks of daily living. Strength training in old age is an effective method for increasing maximal strength (Frontera, *et al.* 1988, Harridge, *et al.* 1999), however, little is known regarding adaptations in the torque-angle relation and its determinants. Therefore, the present *in vivo* experiments aimed to investigate training-induced changes in the torque-angle relation and study the determinants of any alteration.

After receiving ethics committee approval, nine older adults were randomly assigned to a training group (age 74.3 ± 3.5 years; mean ± S.D.) and nine to a non-training control group (age 67.1 ± 2 years). Strength training consisted of 2 series of 10 repetitions at 80 % of the 5-repetition maximum for leg-extension and leg-press exercises, performed 3 times per week for 14 consecutive weeks. Maximal isometric knee extension torque was assessed across the knee joint angle range from 90 to 0 deg (0 deg = full extension). The architecture of the vastus lateralis (VL) muscle (fascicle length and pennation angle) was assessed *in vivo* using ultrasonography. The VL muscle fascicle force was estimated by using a geometrical muscle model and taking into account measurements of torque, agonist-antagonist muscle activation, the tendon moment arm length and muscle architecture. Data were analysed using factorial analysis of variance. Values presented are means ± S.D.

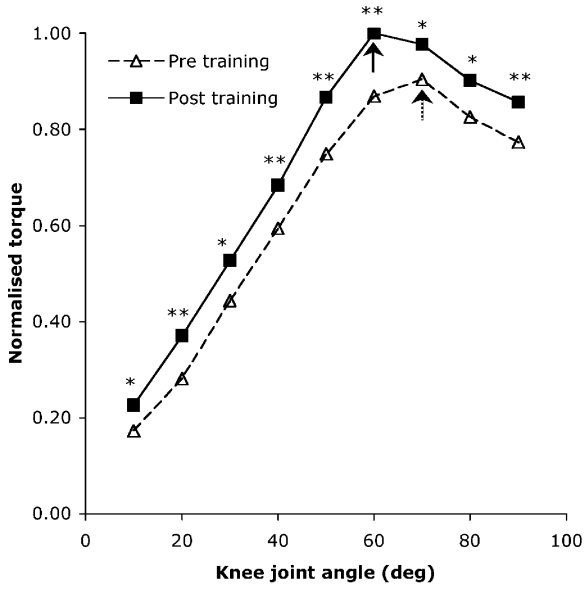

Figure 1. The knee extensor torque-angle relation, normalized to the maximal torque in both pre and post training conditions. Dashed and solid arrows indicate the optimal angle pre and post training, respectively. * and ** Significantly ($P < 0.05$ and $P < 0.01$, respectively) increased torque post training. Values are means ($n = 9$)

Training altered the torque-angle relation (1) displacing it by 9–31 % towards higher torque values and (2) shifting the optimal angle by 10 deg in the direction of full knee extension from 70 deg (121.4 ± 61 Nm) before training, to 60 deg (134.2 ± 57.2 Nm; $P < 0.05$) after training (Fig. 1). Training altered the fascicle force-length relation (1) displacing it by 11–35 % towards higher force values and (2) shifting the optimal fascicle length by 9.5 mm towards longer fascicle lengths from 83.7 ± 8 mm (847.9 ± 365.3 N) before training, to 93.2 ± 12.5 mm (939.3 ± 347.8 N; $P < 0.01$) after training. Despite the change in the fascicle force-length relation, the estimated sarcomere length range in the VL muscle remained the same before and after training, indicating training-induced increases in tendon stiffness (Reeves, *et al.* 2003). The upward displacement of the torque-angle relation was explained mainly by increased activation capacity of agonist muscles, whilst the shift in the optimal angle was due to the increased length of fascicles and increased tendon stiffness. The present findings have implications for muscle function and the assessment of training-induced adaptations in elderly humans.

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All procedures accord with current local guidelines and the Declaration of Helsinki

PC107

An electrophysiological predictor of human error

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This is the first demonstration that imminent absent-minded slips of action can be predicted on the basis of an electrophysiological signal.

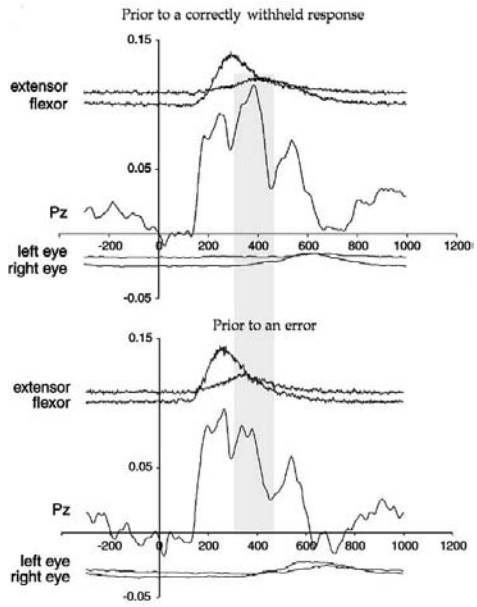

Figure 1. Group averaged ERP in trials prior to an error and prior to a correct withold. Averaged traces of rectified EMG from first dorsal interosseous muscle (flexor), extensor indicis muscle (extensor), EEG at Pz (Pz) and EOG from left and right eyes in 25 subjects. Units for all traces are volts divided by gains given in abstract. For each subject averages were taken of those Go trials immediately prior to a target No-Go trial.. In upper panel, Go trials prior to the subjects making a subsequent slip of action are averaged (Pre-False Press) whilst for lower panel, Go trials prior to subjects making a correct withholding of response to the subsequent No-Go trial are averaged (pre-correct). The P300/P200 ratio is larger in upper panel than in lower panel.

25 normal adult subjects (age 20 – 47;13 women) performed repeated trials of a computerised Go-No Go task with local ethical committee approval. Each subject completed 10 blocks of the SART task. Each block comprised 225 presentations of single digits (1–9) at a regular rate of one digit every 1.15 seconds on a computer screen. The order of the digits was randomised. On each trial, the digit was presented for 250 msec followed by a masking pattern for 900 msec. Subjects were asked to press a computer mouse key as quickly as possible for every number presented with the exception of 3, to which no response should be made. Subjects were given an opportunity to rest from the task between blocks. EEG recordings were made from Fz, Cz, Pz using silver/silver chloride electrodes (Grass). Movement of the eyes was monitored using Electro-oculogram (EOG) electrodes (F7 and F8). All signals were referenced to A2. EMG in the responding hand was monitored using bipolar silver/silver chloride electrodes from an index finger flexor (first dorsal

interosseous muscle) and an index finger extensor (extensor indicis). EEG was amplified 20, 000 fold, EMG 1000 fold, and EOG 2000 fold using AC coupled amplifiers (Biopac Systems Inc., Santa Barbara). Filtering was 10 Hz-5 KHz, 1–35 Hz and 0.05 Hz-100 Hz for EMG, EEG and EOG respectively. Full wave rectification of the EMG was performed digitally. The type of visual stimulus (Go or No Go), its time of presentation and the time of key press were logged digitally with the biological signals. This enabled every trial to be indexed and identified according to type of stimulus and type of response (correct press, false press or correct withhold). All data was digitised at 500 Hz, archived and averaged off-line using a purpose-written averaging program. Statistical analysis was performed with SPSS v8.0.

A highly significant reduction in the normalised P300 component of the visually evoked EEG potential was observed on the trial prior to the trial on which an action error was made (mean 0.92, S.D. 0.33) compared to the trial preceding a correctly withheld response(mean 1.28, S.D. 0.48; paired $t = 3.63$, $P < 0.001$, $df = 24$ see also Figure).

Furthermore, the general propensity of a subject to make errors was strongly predicted by the mean amplitude of subjects' P300/P200 component ratio throughout their task performance ($r = 0.46$, $P < 0.05$). The ability to predict impending action errors with passive scalp recording has potential applications for neurological rehabilitation and industry.

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Human EMGs have a 600 Hz component (I wave periodicity) during articulation

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A high frequency excitatory network in motor cortex generates multiple corticospinal I waves at ~ 600 Hz to a transient cortical stimulus; does the 600 Hz rhythm reflect a possible clock function present even during voluntary activity (Amassian and Stewart, 2003)? Neither individual corticospinal nor α motoneurons discharge at I wave periodicity during voluntary activity; however, a collective of discharging motor system neurons responding stochastically at the basic clock period might reveal I wave periodicity.

With Institutional Review Board approval and informed consent from four subjects (41–78 yrs of age), this 'clock' hypothesis was tested as follows: surface EMGs were recorded over sternohyoid-thyroid muscles by two silver balls, 1 mm in diameter and 10–15 mm interpolar distance; the overlying skin was cleaned with acetone and then covered with a thin layer of electrode paste. Activity by several motor units was elicited by silently articulating (without expiration) plosive consonants (e.g. D, T) and analysing the initial 100–200 ms period of activity. The EMG was half-wave rectified and converted to standard pulses; the onset of activity triggered the accumulation of a post-stimulus histogram (Fig. 1A). A dead time of 4.0 ms was introduced before the standard pulse (0.1 or 0.2 ms duration); thus intervals less than the dead time could appear only in the summed histogram.

First, the iterative expectation density (ED) function was computed on the summed histogram to identify any I wave periodicity. Iteration required that given 2 or more spike pulses in a bin, each was in turn translated to the origin before translating the contents of the next bin. Secondly, the cross-channel ED function was computed between a pulse train with a fixed period of, e.g. 1.4 or 1.5 ms and the EMG derived pulses. Pulse trains were started at the EMG onset. Cross-channel EDs were periodic for each subject at a particular pulse train period and much less periodic at neighboring periods. Figure 1B illustrates a cross-channel ED that was optimal with a pulse train period of 1.4 ms. In other recordings, the optimal period was 1.5 or 1.6 ms. In the 'best-fit' EDs for all subjects, the mean number of EMG pulses per bin outnumbered the in-phase increment.

Summarizing, during articulation, the discharges of at least some EMG units are related to I wave like periodicity.

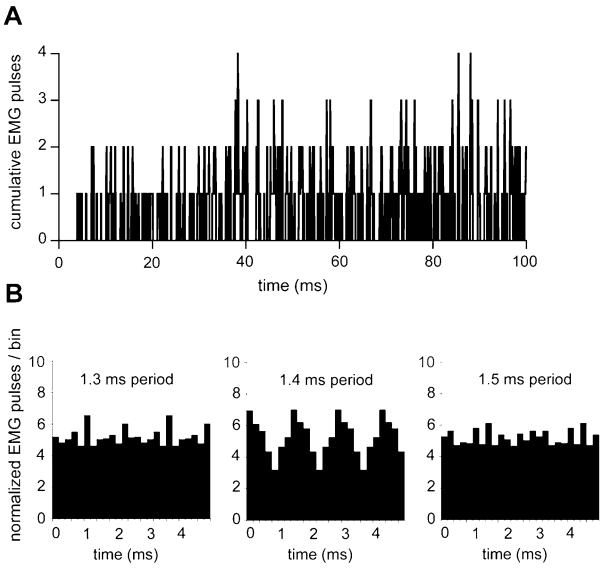

Figure 1. *A*, post-stimulus histogram of EMG pulses to 45 silently articulated 'T.' *B*, cross-channel EDs compared between three pulse trains of period 1.3, 1.4, and 1.5 ms, respectively and EMG spikes of *A*.

Amassian VE & Stewart M (2003). *Suppl Clin Neurophysiol* **56**, 119–142.

All procedures accord with current local guidelines and the Declaration of Helsinki

PC109

Apparent bilateral asymmetry in temperature of children of five and under using infrared ear thermometry

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Electronic 'tympanic' infrared thermometers have gained wide acceptance within the medical community. Although previous paediatric studies have shown a good correlation between rectal and 'tympanic' thermometers (Terndrup and Milewski 1991; Chamberlain *et al.* 1991), there are concerns being raised with respect to their reliability. Here we report data from a study into the relationship between the left and right ear canal temperatures in children between nine months and five years old inclusive.

Medical ethical approval was obtained, and temperatures were only taken bilaterally if the child's parent or guardian gave their verbal consent. The temperature of the ear canal was taken using an infrared thermometer (Genius 5: TycoHealth, N Ireland) following a standardised hospital protocol. The choice of the side

of the first ear to be assessed was alternated between consecutive patients. Basic demographic data were also recorded. The data was analysed using SPSS.

A good correlation was found between the left and right ear canal temperature recordings ($n = 17$; $R^2 = 0.7708$). The average temperature was 36.9°C with a range from 36.0° to 37.7°C. Comparison of first and second recorded temperature irrespective of side also showed a reasonable correlation ($R^2 = 0.6502$). However, it appeared that the first recorded temperature tended to be consistently lower than the second in this age group.

Even though this was a small normative study, this is the first report that has considered that some of the variability noticed in use of this assessment methodology might derive from a mixture of the technology and the physiology. It would appear that in young children, there is a reaction to the first reading that causes the second reading to be generally smaller. As such it would appear that reading from one ear only could lead to errors in the monitoring of temperature in this vulnerable group.

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All procedures accord with current local guidelines and the Declaration of Helsinki