

1 Title:
2 Physiological and psychological responses in Fire Instructors to heat exposures.

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4 Running Title:
5 Fire Instructor Health

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1 Abstract

2 *Aim:* Fire Service Instructors (FSI) are exposed to many repeated periods of high
3 environmental temperatures when training firefighters. Such repeated exposures will
4 impose significant strains on the function of instructors. We aimed to measure the
5 effects of a training programme including repeated exposures to heat, termed “Wears”
6 in the fire service, on the physiological, psychological some immunological markers of
7 Fire Service Instructors. *Methods:* Six FSI and six physiologically matched controls
8 completed blood and cardiovascular tests pre and post a 4wk heat instruction training
9 block, controls completed the tests only. FSI were given a 7wk period of no heat
10 exposure prior to starting the training. Physiological and perceptual measures were
11 taken pre and post the first and last Wear of the 4wk training protocol. *Results:* There
12 were acute effects of a Wear on core temperature and physiological strain index, as well
13 as measures of fatigue. The acute exposure to heat during a Wear led to a consistent
14 decrease in CRP (-10 to -40%), increased IL6 concentrations (33 to 45%) as well as
15 increased RPE and TSS. Over the training programme significantly lower quantities of
16 white cells, particularly neutrophils, leucocytes and monocytes were found in the FSI
17 group. Between the start and the end of the 4 week training programme the FSI showed
18 a significantly greater physiological strain index (PSI) to the Wears, which nearly
19 doubled from 2.5 to 4.7 ($p<0.05$). *Conclusion:* Physiological and psychological
20 measures indicate that FSI may be experiencing symptoms and changes to their health
21 consistent with an overtraining type condition.

22
23 Key words:

24 Immune function; Inflammation, Fire service; Heat exposure; Overtraining.
25

1	Abbreviations:	
2	Δ	Change
3	ANOVA	Analysis of variance
4	BP	Blood pressure
5	CO	Carbon monoxide
6	CON	Control
7	C-RP	C-reactive protein
8	Crt	Cortisol
9	FEV ₁	Forced expiratory volume at 1 sec
10	FSI	Fire service instructors
11	FVC	Forced vital capacity
12	Hb	Haemoglobin
13	Hct	Haematocrit
14	HR	Heart rate
15	IgG	Immunoglobulin G
16	IL-6	Interleukin-6
17	IQR	Interquartile range
18	MFSI-SF	Multidimensional Fatigue Symptom Inventory-Short
19	Form	
20	NBM	Nude body mass
21	PEF	Peak expiratory flow
22	PSI	Physiological strain index
23	RPE	Rating of perceived exertion

1	SCBA	Self-contained breathing apparatus
2	SD	Standard deviation
3	T_{re}	Rectal temperature
4	TSS	Thermal sensation scale
5	U_{col}	Urine colour
6	U_{osm}	Urine osmolality
7	U_{sg}	Urine specific gravity
8	$\dot{V}O_{2max}$	Maximal oxygen uptake
9	WBC	White blood cells
10	wk	Week

1 Introduction:

2 Training new recruits for the fire service entails exposure of the trainees and their
3 trainers to uncompensable heat stress (Montain et al., 1994), on successive days
4 throughout an intense training programme, which can often last 4-6 weeks before
5 receiving a break. Simulated firefighting exercises occur in uncompensable heat stress
6 conditions, with reported environmental temperatures ranging 67-190°C (Eglin et al
7 2004; Eglin 2007). The physiological strain experienced during such exposures is
8 indicated by increased rectal temperatures (38.0-39.0°C), near maximal heart rates
9 (119-189 b.min⁻¹), exaggerated levels of sweat loss (0.5-2.0 L.hr⁻¹) and high rates of
10 energy expenditure (oxygen consumption 2.3-3.55 L.min⁻¹) (Eglin, 2007). Fire service
11 instructors (FSI) are further affected by reduced heat dissipation from limited vapour
12 permeability and restricted bodily movement while wearing protective clothing (Selkirk
13 and McLellan 2004). Diminished work capacity and decrements in heart function have
14 been measured in firefighters as a result of working in the heat during firefighting
15 situations (Fernhall et al 2012). Investigations by Williams et al. (1996) on
16 cardiovascular responses in FSI showed near maximal heart rate values which were
17 higher than their trainee firefighters. More recently, Eglin et al. (2004) reported that FSI
18 experienced severe physiological strain, arising from the strenuous duties and
19 prolonged duration of heat stress and this was shown to compromise their ability to
20 perform instructional tasks. A variety of perceptions of psychological function in such
21 studies may reflect differences in external heat loads and work duties (Eglin et al 2004)
22 as well as the personnel investigated. When measured FSI report increased anxiety,
23 mostly through increased feelings of apprehension and nervousness (Eglin 2007),
24 which may impede mental performance, cognitive function (Smith et al 1996) and
25 result in injury through inappropriate decision making, which worsens with repeated or
26 prolonged exposures (Smith et al 1996, 1997).

27 The effects of simulated acute heat exposures may reduce immune function, in the short
28 term, (Sheppard et al 1986; Smith et al 2005) and it is likely to have longer term
29 consequences, especially if there is insufficient time for recovery between bouts of heat
30 exposure. As well as this firefighting personnel appear to be at greater risk of
31 respiratory (Sheppard et al 1986) and cardiovascular diseases (Fernhall et al 2012),
32 although nothing is known about the incidence in FSI. Measurement of immune
33 function and markers of systemic inflammation, following acute heat exposure, have

1 shown alterations indicative of infection or inflammatory process (Smith et al 2005;
2 Huang et al 2010). Elevated concentrations of cortisol and interleukin-6 (IL-6) support
3 the suggestion that fire service personal may be at a greater risk of health complications
4 due to repeatedly undertaking demanding thermal exposures when training and
5 throughout their career (Sheppard et al 1986; Fernhall et al 2012).

6 FSI are likely to experience larger number of heat exposures, combined with physical
7 exertion, compared to standard fire service personnel and are therefore likely to be at
8 greater risk of the consequences of accumulated heat stresses. However, repeated
9 exposure to heat and exercise may be beneficial in some circumstances. Given suitable
10 preparation and safeguard exercise in the heat on repeated occasions may lead to
11 improved performance, a process termed acclimation (Gibson et al 2015). The aim of
12 the study was to see whether the FSI were affected by the training programme and to
13 quantify the physiological strain, perceptual stress and markers of inflammation and
14 immune function in a cross-section of FSI and provide data on their changes during
15 their typical working routines. A further aim was to measure FSI experiences of the
16 repeated heat exposures and their effects on fatigue perception. The study examined the
17 effects of a rest period before return to training. In particular we were interested in the
18 acute response to a single bout of severe heat exposure, known by the Fire Service as a
19 'Wear', and the response to a 4wk fire instruction course. It was hypothesised there
20 would be significant differences in acute and chronic responses to the heat exposure for
21 FSI and that FSI would show physiological, immunological and psychological
22 characteristics different to a non-heat exposed group of matched individuals. We also
23 hypothesised that due to the process of heat acclimation the FSI would experience
24 lower stress due to the heat exposure at the end of the training period.

25

1 Methods:

2 *Participants*

3 Six male FSI from the Fire Service College (Moreton-in-Marsh, England, UK) and six
4 non-firefighter control (CON) participants volunteered after providing written informed
5 consent to participate (Table 1). The study was approved by the Institution Ethics
6 Committee and conducted in accordance with the Declaration of Helsinki (revised
7 2008). FSI had a range of service history (2-10 years), while CON participants had not
8 been exposed to ambient temperatures >25°C in the previous 4 months to any measures
9 taken.

10

11 *Experimental Design*

12

13 Initial physiological, perceptual, inflammatory and immunological measures, were
14 taken at the end of a previous instruction course, the control group were measured at
15 the same time of the year. The measures were repeated after 7wks of no-heat exposure
16 for the FSI, all volunteers maintained normal exercise routines. This was followed by a
17 4wk breathing apparatus fire instruction course that included fifteen Wears per
18 instructor. At the end of the course FSI were re-measured. The control group were re-
19 measured at a similar time of the year to the FSI to avoid any seasonal influences on the
20 variables measured and the activities in Wear 1 and Wear 2 were matched as closely as
21 possible. (Figure 1). For measuring the acute response to heat, Wear 1 and Wear 2 were
22 used and matched for working conditions as much as was possible, in terms of exposure
23 time and thermal environment.

24

25 *Experimental Procedures*

26 *Fire Instruction Drills*

27 The fire instruction drills included fire behaviour, fire attack and, search and rescue.
28 FSI wore their fire protective clothing during the Wears; including jacket (Bellyclare
29 Special Products Ltd.), trousers (Bellyclare Special Products Ltd.), boots (9005 GA,
30 Jolly Scarpe, USA), flash hood (MSA Gallet, Bellshill UK), helmet (FISF, MSA Gallet,
31 Bellshill, UK), gloves (Firemaster 3, Southcombe Brothers Ltd, Somerset, UK)
32 personal undergarment and self-contained breathing apparatus (SCBA), weighing

1 ~21kg in total. Wears lasted 37 ± 19 mins (range 15-120 mins), temperature in-Wear
2 averaged $174.0 \pm 83.9^\circ\text{C}$ and were typically conducted 1 or 2 times per day over 4
3 consecutive days, followed by 3 days without exposure. On one occasion a FSI had
4 three Wears in 1 day. Work rotas determined the role of the FSI during the Wear and
5 therefore the duration and thermal stress experienced. Although FSI do not undertake
6 firefighting duties, they continually assess and monitor trainees, set and stoke fires,
7 carry dummy casualties and climb stairs. FSI roles are intermittently changed between
8 instructors, as some are situated within direct heat exposure, while others observe from
9 a distance.

10 *Physiological Measures*

11 Heart rate (HR) was measured continuously using telemetric monitors (Accurex+, Polar
12 Electro, Oy, Kempele, Finland). Rectal temperature (T_{re}) was measured pre and post
13 Wears using a single use probe (449H, Henleys Medical, Hertfordshire, UK), placed
14 10cm past the anal sphincter, and displayed on logging monitors (YSI, 4600 series,
15 Hampshire, UK). Physiological strain index (PSI) was calculated using the equation of
16 Moran et al. (1998):

$$\text{PSI} = 5 (T_{ret} - T_{re0}) \cdot (39.5 - T_{re0})^{-1} + 5 (HR_t - HR_0) \cdot (180 - HR_0)^{-1}$$

17 Where T_{re0} and HR_0 are initial resting values, and T_{ret} and HR_t were measured post-
18 wear.

19 Blood pressure (BP), lung function (forced vital capacity [FVC], forced expiratory
20 volume at 1 sec [FEV₁], FEV₁/FVC ratio (%) and peak expiratory flow [PEF]) and
21 carbon monoxide (CO) were measured using automatic monitors (Boso, Bosch-Sohn,
22 Medicus, Germany) hand-held spirometers (Micro Medical Ltd., Cranleigh Ltd., UK)
23 and a piCO⁺ Smokerlyzer® (Bedfont, Scientific Ltd., Maidstone, Kent, UK),
24 respectively.

25 Hydration status was assessed from urine samples (colour [U_{col}], osmolality [U_{osm}] and
26 specific gravity [U_{sg}]) using a colour chart, Pocket Pal-Osmo (Vitech Scientific, Ltd)
27 and refractometer (Atago Co., Tokyo, Japan). Criteria for adequate hydration were set
28 at $<700\text{mOsm.kg}^{-1}$ and <1.020 (Sawka et al 2007). Nude body mass (NBM) was
29 recorded (Detecto Scale Company, USA) pre and post Wears to estimate non-urine
30 fluid loss, calculated as; pre NBM – post NBM / Wear time (mins).

1 *Cardiovascular Function*

2 Cardiorespiratory fitness was assessed pre and post the 4wk course using an
3 incremental $\dot{V}O_{2\max}$ test, conducted at the Fire Service College and within the
4 Institution laboratory. Tests occurred on Technogym (Technogym UK Ltd., Bracknell,
5 Berkshire) and Woodway (ELG2, GmbH) treadmills set at 1% incline (Jones and
6 Doust, 1996). The running test started with 5 min warm up at 5 km.hr⁻¹, which
7 increased to 8 km.hr⁻¹ and then subsequently by 0.8 km.min⁻¹ until exhaustion (Aoyagi
8 et al., 1994). HR and rating of perceived exertion (RPE) (Borg 1982) were recorded
9 during the final 15s of every one minute stage. Pulmonary gas measurements were
10 recorded continuously using a Cortex Sport Metalyser (Cranleigh Ltd., UK), calibrated
11 at the start of each test, for ambient pressure (Barometer), for volume using a 3 litre
12 syringe (Hans Rudolph, series 4900, Kansas City, USA) and gas composition, using
13 standard gases (17.1% oxygen and 5.0% carbon dioxide).

14 *Blood Measures*

15 Blood samples were taken from an antecubital vein from the participant's non-
16 dominant arm, using a 20ml syringe and 21G hypodermic needle, while participants
17 were sitting upright and resting. To assess the impact of longer term changes to the
18 variables we measured, blood samples were taken pre and post the 7wk no-heat
19 exposure period. For determining the acute responses blood was taken immediately
20 before and as soon as possible after Wear 1 and Wear 2 of the 4wk training course.
21 Plasma was obtained by centrifugation (2500g for 15 min in a refrigerated centrifuge)
22 and stored frozen (-85C) for analysis when all samples had been collected.

23 Whole blood samples were assessed for blood cell count using automated flow
24 cytometry with electrical impedance and light detectors (full blood count analyser,
25 Sysmex, Europe). Values for white blood cells (WBC) and white blood cell content
26 (neutrophils, eosinophils, basophils, lymphocytes and monocytes) were analysed.
27 While haemoglobin (Hb) and haematocrit (Hct) estimated change in plasma volume
28 (Δ PV) (Dill and Costill, 1974).

29 Plasma was thawed before being analysed for C-reactive protein (C-RP) (Sigma
30 Aldrich, UK), cortisol (Enzo Life Sciences, UK), IL-6 (Sigma Aldrich, UK) and
31 Immunoglobulin G (IgG) (Enzo Life Sciences, UK) using ELISA.

1 *Perceptual Measures*

2 Perception of fatigue was measured pre and post Wears using the Multidimensional
3 Fatigue Symptom Inventory-Short Form (MFSI-SF, (Stein et al 2004)). The
4 introductory statement was modified for FSI to relate to that particular present moment.
5 FSI rated themselves against 30 items on a scale of 0 representing 'not at all' to 4
6 meaning 'extremely'. These items load equally onto fatigue subscales (General,
7 Physical, Emotional, Mental, Vigour) and an overall Total Fatigue Scale (for details on
8 scoring see Stein et al 2004). Thermal sensation (TSS, (Toner et al 1986)), RPE and
9 Affect (feeling, (Rejeski and Kenney 1987)), were measured using visual scales. FSI
10 were familiarised to the scales during their first visit.

11 *Data analysis*

12 Acute effects

13 Differences between pre and post Wears and differences between Wear 1 and Wear 2 in
14 the physiological measures were analysed using dependent-samples t-tests. TSS and
15 RPE was analysed using 2-way repeated measures ANOVAs with Wear and Test as the
16 within-subject variables. Significant effects indicated by ANOVA were followed up
17 using Bonferroni corrected pairwise comparisons. The MFSI-SF was analysed using
18 Wilcoxon signed rank test on each of the scales of the questionnaire to compare
19 differences pre-to post Wear and between the Wears. Relationships between the
20 perceptual measures and physiological and measures of immune function were
21 examined using Pearson's product moment correlation coefficient. Blood measures
22 comparing the five test points were analysed using 1-way repeated measures ANOVAs
23 (prior to 7wk no-heat exposure, pre Wear 1, post Wear 1, pre Wear 2 and post Wear 2).
24 Blood measures comparing the effects of the FSI with the control group over the 4wk
25 fire instruction course were analysed using 2-way mixed design ANOVA with Group
26 and Test (prior to 7wk no-heat exposure, pre-4wk training course and post-4wk training
27 course). Data is presented as mean \pm standard deviation (SD), and statistical
28 significance was accepted when $p < 0.05$. Data were analysed using SPSS (version 20.0).

1 Results:

2 There were no significant differences in age, physical or performance characteristics
3 between the FSI and CON group (Table 1). Nor were there differences in baseline
4 resting measures of BP (systolic [p=0.55] and diastolic [p=0.49]), NBM [p=0.37], CO
5 [p=0.07] and resting HR [p=0.62]) between groups (Table 1).

6 *Responses to Single Wear - Acute Effects*

7 *Physiological Measures*

8 There were significant increases in T_{re} as a result of undertaking Wear 1 (p=0.04) and
9 Wear 2 (p=0.01), but no difference in ΔT_{re} between Wears (p=0.85, Table 2). HR was
10 not significantly different prior to both Wears, although significant increases did occur
11 as a result of Wear 1 (p=0.05) and Wear 2 (p=0.01). HR was also higher at the end of
12 Wear 2 and displayed a significantly greater rise than Wear 1 (p=0.01). However, peak
13 and mean HR were not significantly different over the two measured Wears. Large
14 fluid loss was observed during each Wear, although did not significantly differ between
15 Wear 1 and Wear 2 (p=0.58, Table 2). There were no significant differences in the pre
16 to post-Wear comparison for hydration status, respiratory function or blood pressure.

17 *Perceptual Measures*

18 RPE and TSS scores significantly increased during Wear 1 (p=0.01 and p=0.03,
19 respectively) and Wear 2 (p=0.01 and p=0.02, respectively). Although, no differences
20 in RPE or TSS between Wears transpired (p=0.14 and p=0.54, respectively).

21 *Responses to Repeated Wears –longer term effects*

22 *Physiological Measures*

23 Over the 4wk training course there was a significant reduction in the FVC for the FSI
24 group (p=0.001) and FEV₁ (p=0.06). No differences were observed for the CON group.
25 Cardiovascular fitness, as measured by $\dot{V}O_{2max}$, did not significantly change in either
26 group (FSI p=0.26 and CON p=0.48). However, FSI $\dot{V}O_{2max}$ showed a 7% decline at
27 the end of the 4wk training course.

28 *Perceptual Measures*

1 No differences ($p>0.05$) in MFSI-SF scales were found pre to post Wear 1. There were
2 significant ($Z=2.21$, $p=0.03$, $r=0.64$) differences pre to post Wear 2 in the General
3 scale, from 2.5 (interquartile range [IQR] =6.75) to 6 (IQR=15). No other scale
4 measures showed any differences ($p>0.05$). There was a trend for the Total Fatigue
5 scale to increase pre (5, IQR=11.25) to post (11.5, IQR=30) Wear 2 ($Z=1.83$, $p=0.07$,
6 $r=0.53$). No differences were found between pre Wear 1 and pre Wear 2 ($p>0.05$),
7 suggesting no increase in the perception of fatigue is associated with 4wks instruction.
8 A positive correlation was found between the ΔT_{re} and the change in general fatigue for
9 Wear 2 ($r(4)=0.89$, $p=0.18$), $y=0.044x + 0.41$, but was not found in Wear 1 ($r(4)=0.31$,
10 $p=0.56$), $y=0.030x + 0.6405$, nor between changes in general fatigue and other
11 physiological measures ($p>0.05$).

12 *Blood Measures*

13 WBC numbers and neutrophils increased during Wear 1 (+19% and +32.3%) and Wear
14 2 (+19% and +26.7%), respectively. A significant interaction ($p=0.04$) occurred, where
15 FSI total WBC numbers were highest at the first measurement ($p=0.00$), decreased after
16 7wks no-heat exposure, ($p=0.57$) and plateaued over the 4wk instruction course
17 ($p=0.041$), compared to no change in CON. FSI neutrophil counts were highest at the
18 first measurement, decreased after 7wks of no-heat exposure and remained steady over
19 the 4wk fire instruction course. FSI neutrophil numbers significantly increased pre to
20 post Wear 1 ($p=0.01$), but not Wear 2 ($p=0.82$). No interactions for basophil ($p=0.96$)
21 or eosinophil ($p=0.35$) concentrations occurred (Table 3). IgG measures showed no
22 change ($p=0.12$) between groups. There was a trend for lower concentrations of IgG in
23 FSI towards the end of the experimental period, displaying a significant ($p=0.04$)
24 reduction from the pre 7wk no-heat exposure period to post Wear 2. C-RP measures
25 also showed no difference ($p=0.22$) between or within FSI and CON. However,
26 elevated levels ($p>0.05$) of C-RP were found within FSI prior to the 7wks no-heat
27 exposure, pre Wear 1 and pre Wear 2, when compared to CON. C-RP concentrations
28 also declined ($p>0.05$) pre to post Wear 1 and Wear 2. IL-6 significantly differed in FSI
29 ($p=0.00$), but not CON. Resting IL-6 concentrations were elevated in FSI pre 7wk no-
30 heat exposure period, which then decreased after no-heat exposure and then
31 subsequently increased over the 4wk course. A significant interaction ($p=0.001$) was
32 observed between CON and FSI groups, where larger concentrations of IL-6 were
33 observed in FSI prior to the 7wk no-heat exposure period ($p=0.01$) and post 4wks

1 instruction ($p=0.00$). Cortisol levels increased pre to post Wear 1 ($p=0.01$) after 7wks
2 no-heat exposure in FSI, however, no interactions occurred between groups over time
3 (Table 3).

4

Discussion:

The aim of the study was to quantify important physiological, psychological, inflammatory and immunological measures in FSI during periods of no-heat exposure, acute heat exposure and following a 4wk fire instruction course. Large decrements in FVC and $\dot{V}O_{2\max}$ were observed in FSI after the 4wk course, while leukocytes and inflammatory responses significantly changed during periods of no-heat exposure, a single Wear and following the 4wk fire instruction course. These significant changes during the 4wk fire instruction course indicate an overtraining response. This is possibly contributed by repeated exposures in uncompensable conditions, increased physiological strain and insufficient recovery between wears.

Single Heat Exposure

Physiological measures

After 7wks of no-heat exposure, FSI experienced low physiological strain (2.5 ± 1.1) during Wear 1. However, after the 4wk training course, significantly higher end of Wear HR and moderate levels of physiological strain (4.7 ± 0.7) were displayed during Wear 2. Similar to data reported by Eglin and Tipton, (2005), post Wear T_{re} reached $\sim 38.1^{\circ}\text{C}$ in both Wears, with mean and maximal HR similar to those measured by Eglin et al. (2004) of 109 ± 22 and $138 \pm 26 \text{ b}\cdot\text{min}^{-1}$, respectively. These results are slightly lower than those found in other fire service studies (Romet and Frim 1987; Bennett et al 1995; Smith et al 1997; Ilmarinen and Koivistoinen 1999; Smith et al 2001a), where differences may be explained by the duration of heat exposure, workload intensity and choice of trainee participants as opposed to FSI in this study. Moreover, as FSI undertake different roles during wears, which predetermines their level of physical activity, duration and severity of thermal exposure experienced, it is difficult to perceive the Wear as a homogenous event and response reflecting the experience of the whole group. Therefore, longer or more intense thermal exposures would be expected to induce a greater strain, through larger rates of metabolic heat production and higher T_{re} (Eglin and Tipton 2005). Varied thermoregulatory responses would also be expected within FSI whilst working at similar relative intensities due to individual biophysical characteristics (Cramer and Jay 2014).

Hydration

On average the FSI group arrived in a hydrated state to both Wears, however, three arrived to Wear 1, and two arrived to Wear 2 in a hypohydrated state. Similar neglect in volunteers was observed by Eglin et al. (2004) and may have occurred at other times during the 4wk course. Arriving dehydrated might be expected to impair the instructor's performance, thermoregulation and cognitive function (González-Alonso et al 1997). Subsequent reductions in plasma volume and excessive sweating would have compromised blood flow supply to working tissues, placing a significant cardiovascular strain on the FSI (Cheuvront and Haymes 2001; Smith et al 2001). Both Wears induced mild levels of dehydration (~1% body mass), which are unlikely to affect thermoregulation or performance measures greatly (Cheuvront and Haymes 2001). Nevertheless, the magnitude of dehydration and its effects will be related to initial hydration status. The added burden of wearing protective clothing, which increases thermal strain by placing a greater metabolic cost and lowers heat loss opportunities for the individual, is likely to compound the problems of hypohydration. Further, reduced heat dissipation and sweat evaporation from the limited vapour permeability, continually increases body heat storage (Selkirk and McLellan 2004). This will be further affected by greater respiratory demands (Bygrave et al 2004) from the SCBA, causing greater rates of oxygen consumption and metabolic heat production, thus increasing the rate of body temperature and the risks for heat related illnesses.

Repeated Heat Exposure

Respiratory

Lung function decrements, to a similar extent to those measured here over a 4wk training course, have been observed in other studies, Peters et al. (1974) observed a worsening of lung function with increased number of fire exposures. Lung function impairments of a similar scale to this study, and respiratory symptoms have been identified after acute (Large et al 1990; Greven et al 2012) and repeated heat exposures in fire service personnel (Mustajbegovic et al 2001; Ribeiro et al 2009). Such symptoms and impairments may be associated with acute neutrophilic airway conditions, long term systemic inflammatory responses and lung tissue damage, temporary and chronic (Large et al 1990; Greven et al 2012). Possible causes include the adverse effects of daily smoke inhalation upon the respiratory system, as displayed by increased CO measures in FSI as they prepare, recover and change within buildings surrounding the Wears. This may have led to the decline in chronic lung function and

cardiorespiratory fitness (Large et al 1990; Mustajbegovic et al 2001; Ribeiro et al 2009; Greven et al 2012). Multiple air pollution exposures increase the possibility of lung dysfunctions and exacerbate respiratory symptoms, the higher concentrations of asphyxiating irritant toxic gases permit inhaled allergens to affect the immune system, when airway mucosa is damaged and mucociliary clearance is lowered (D'Amato et al 2005). Further studies are warranted to investigate the air pollutants and subsequent health risks while FSI prepare and recover in buildings surrounding Wears.

Cardiovascular

The FSI with the largest lung function decline (FVC -15% and FEV₁ -6%) also incurred the greatest reduction in $\dot{V}O_{2max}$ (-18%). Even small alterations in $\dot{V}O_{2max}$ (average FSI -7%), indicate physiological changes that would lead to impaired function. The variance in decline of cardiovascular function in FSI, such that some are highly affected, but not all, warrants further investigation with a larger cohort. It remains to be seen whether cardiovascular fitness has any influence over long-term health, but it is likely to contribute to how heat tolerant an individual may be (Epstein 1990). Other plausible explanations suggest motivational and physical fatigue post 4wks fire instruction as seen within psychological assessments, not necessarily due to underlying cardiovascular changes. Decrements in FSI fitness of the sizes measured here may be of concern, as fitness affects thermoregulation and performance abilities. Where those who are less trained will take longer to complete tasks under heat stress, work at higher percentages of their $\dot{V}O_{2max}$ and therefore, reach exhaustion earlier or experience heat related illnesses sooner (Sothmann et al 1990).

Perceptual measures

Consistent with other studies (Smith and Petruzzello 1998), RPE significantly increased during both Wears. There was a significant increase in the FSI perceptions of general fatigue post Wear 2, but not after Wear 1, suggesting the 4wk instruction course moderated this exposure effect and was the result of repeated exposures. This result did not appear to be dependent upon an overall increase in perceptions of fatigue during the 4wk course, because no differences in pre Wear scores transpired. No other significant effects were found for other scales measured by the MFSI-SF.

Perceptions of general and total fatigue were significantly correlated with T_{re} . In the case of total fatigue this relationship was dependent during acute heat exposure after the repeated 4wk course. The change in perceptions of general fatigue after Wear 2 was significantly correlated with the ΔT_{re} , however, no significant correlations were found in the corresponding comparison from Wear 1. Whilst the exact relationship is unclear, frequent repeated exposures to hot environments appear to lead to FSI to become more sensitive to their T_{re} . Furthermore, repeated heat exposure seems to result in sensory association of T_{re} with feelings of fatigue, rather than other physiological or psychological variables. This study is the first to show that repeated heat exposures change the way perceptions of fatigue relate to T_{re} . Whilst the exact relationship is unclear, frequent repeated exposures to hot environments may lead to sensory feedback from T_{re} to appraise one's general feelings of fatigue. Future research should be directed towards this sensory association hypothesis to determine if specific sensory adaptation occurs as a direct result from repeated heat exposure or as a consequence of general experience in the task domain.

Blood variables

Initial blood samples measured after a previous instruction course showed that total white cell counts in FSI neared the higher end of the normal clinical range and were higher than the CON group, suggesting some chronic effects of heat exposure on the immune system and its regulation (Bain et al 2011). During the no-heat exposure period, FSI white cell counts reduced and approached lower normal clinical values, suggestive of lower immune system stimulation during this period of recovery away from repeated heat exposure. These reductions were identified as lowering of neutrophils, lymphocytes and basophil cell numbers, possibly indicating that the rest period allows the instructors to recuperate and return their immune system towards more normal values. The lower concentrations of leukocytes, indicated by WBC, neutrophil and lymphocyte counts may demonstrate the FSI were in a condition where their immune system was either suppressed or under little challenge. Leukocytosis increased 19% over the 4wk instruction course, whereas others have shown increased leukocytes (38% and 85%) along with monocyte, lymphocyte and neutrophil increases from intense firefighting training (Smith et al 2004) and exercise (14%) alone (Jimenez et al 2008). The leukocytosis may be due to increased blood flow and plasma catecholamine concentration, but unlikely to be due to ΔPV (Gabriel et al 1992). The

FSI and CON group displayed IgG levels close to the recognised upper normal levels for most of the samples taken, with little evidence of any florid immune response. The FSI small IgG reduction during the final stages of the study may indicate a lowered immune response, i.e. minor immune challenge, or immune suppression and lower basal production of immunoglobulins. Without a broader set of analyses this is difficult to discriminate the major likely cause, however, combined with the cortisol data, which suggests normative values, the likelihood of immunosuppression is small. Significant declines in resting IL-6 over the no-heat exposure period occurred in FSI, suggesting a reduction in inflammation or other factors that regulate IL-6 release or removal processes. FSI values of IL-6 were at a level expected when exposed to continual exercise and heat, leading to significant increments over the 4wk course, as opposed to consistent CON measures who refrained from heat exposure and prolonged exercise. This chronic elevation of IL-6 may be indicative of a long-term inflammatory response, similar to the effects seen in overtraining syndrome (Jürimäe et al 2011). Cortisol levels in both groups were towards the lower end of normal range, however, no indications arose of hyper or hypo secretion (Marshall et al 2012). CRP in FSI were commonly elevated to values at, or above the upper limit for a normal population (Marshall et al 2012), indicative of an ongoing inflammatory condition. Interestingly, CRP concentrations declined in all FSI pre to post Wears. The raised T_{re} is likely to stimulate or promote the synthesis of CRP, therefore, the decreased concentration during a heat exposure is most likely the result of greater increases in the removal or breakdown of CRP (Thongtang et al 2013).

Study Limitations

The study evaluated a small sample of FSI, although integrating such testing into workloads is challenging. Further, it is impossible to control heat exposure and specific instructor roles throughout the 4wk course. Nevertheless, this design demonstrates an ecologically valid reflection of FSI working practices.

Conclusions:

This inter-disciplinary study demonstrated that fire instruction may induce physical fatigue, alters physiological responses, changes inflammation and immunological measures and perceptions of stress and psychological fatigue. This is the first study to effectively quantify acute and chronic responses of FSI during their normal working

practice, using an applied and innovative design. A single Wear exposure was shown to induce significant increments in physiological strain and acute inflammation, although FSI are able to recover from and do not perceive them as fatiguing. However, repeatedly working under heat stress with an associated cumulative physiological strain and perceptions of general fatigue during wears, demonstrates a similar response to an overtraining type of syndrome. Time away from heat based instruction appears to help recover the measures back to normal ranges considered healthy. Future research is required to investigate acute post-Wear recovery, cooling modalities and decontamination procedures. Similarly, further investigations are warranted concerning working rotas to reduce the length of heat based cycles, and to allow recovery periods to reduce chronic inflammation and immunosuppression for FSI long-term physical and mental welfare.

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Conflict of interest:

The authors declare that they have no conflicts of interest.

Biography:

Dr Peter Watt

Dr Peter Watt has published over 90 peer reviewed articles, 90 abstracts and currently acts as a reviewer for multiple international journals. Dr Watt's research focuses on the application of stable isotope methods to measure metabolic and physiological changes occurring in humans during exercise, with application to health related problems, e.g. diabetes, obesity. Dr Watt has supervised 11 Ph.D and 1 M.Phil completions, is currently supervising a further 5 PhD students and is now an Associate Professor.



Mr Ashley Willmott

Ash completed his B.Sc. (Hons.) undergraduate degree in Sport and Exercise Science in 2012 at the University of Brighton. He continued his academic studies at the University by starting an M.Phil. In October 2012, which is examining the effects of short and long term heat acclimation protocols on the interplay between heat acclimation state, training status and inflammatory markers in hot and humid conditions. Ash is currently working within the Sport and Exercise Science Consultancy Unit (SESCU), undertaking physiology support to athletes and is an active member of British Association of Sport and Exercise Science (BASES).



Dr Neil S. Maxwell

Neil joined the University of Brighton as a lecturer in sport and exercise science in 1997, where he lectures undergraduate and postgraduate students, predominantly in the areas of exercise and environmental physiology and research methods. Neil is research active, an approved higher degrees supervisor with M.Phil./Ph.D. completions and a bank of existing postgraduate research students. He has published extensively in the international, scientific literature in areas allied to thermal and hypoxic stress and how the body tolerates each, particularly during exercise. He is now Reader, assistant head of school (research) and head of the Centre for Sport and Exercise Science and Medicine, while also leading the Environmental Extremes Laboratory.



Dr Nicholas Smeeton

B.Sc. (Hons) Psychology and Physiology (University of Reading); M.Sc. Sports Psychology (Liverpool John Moores university); PhD Anticipation Skill in Tennis (Liverpool John Moores university). Nick lectures in a range of topics including sport and exercise psychology, motor behaviour and research methods. His research work focuses on the training of anticipation and decision-making skills in academy football players and young elite tennis players. He has worked in professional sport and delivered performance enhancement to national and international level athletes.



Dr Alan Richardson

Dr Alan Richardson's research focuses on the physiological changes and human tolerance to hypoxia and severe heat exposure. Dr Richardson worked as a research exercise physiologist on the Centre for Aviation, Space and Extreme Environment Medicine Xtreme Everest Project in 2007 and 2009, carrying out cardiopulmonary exercise testing in trekkers ascending to Everest Base Camp. Since then Dr Richardson has lead a research consultancy project with the National Fire Service and supervises a PhD student investigating the immune function and inflammatory responses to repeated fire exposures in Fire Service instructors. Dr Richardson is continuing to investigate thermal interventions and post heat exposure cooling strategies, while also course leader for the Sport and Exercise Science degree and leads the Physiology for Sport and Exercise, Expedition Physiology and Dissertation modules.



References:

- Aoyagi, Y., McLellan, T., Shephard, R. (1994) .Effects of training and acclimation on heat tolerance in exercising men wearing protective clothing. *Eur.J.Appl.Physiol.Occup.Physiol.* 68, 234–245.
- Bain BJ, Bates I, Laffan MA, Lewis SM (2011) *Dacie and Lewis Practical Haematology: Expert Consult: Online and Print.* 668.
- Bennett BL, Hagan RD, Banta G, Williams F (1995) Physiological responses during shipboard firefighting. *Aviat Space Environ Med* 66:225–31.
- Borg GA (1982) Psychophysical bases of perceived exertion. *Med Sci Sports Exerc* 14:377–81.
- Bygrave S, Legg S, Myers S, Llewellyn M (2004) Effect of backpack fit on lung function. *Ergonomics* 47:324–9.
- Chevront SN, Haymes EM (2001) Thermoregulation and marathon running: biological and environmental influences. *Sports Med* 31:743–62.
- Cramer MN, Jay O (2014) Selecting the correct exercise intensity for unbiased comparisons of thermoregulatory responses between groups of different mass and surface area. *J Appl Physiol* 116:1123–32. doi: 10.1152/jappphysiol.01312.2013
- D’Amato G, Liccardi G, D’Amato M, Holgate S (2005) Environmental risk factors and allergic bronchial asthma. *Clin Exp Allergy* 35:1113–24. doi: 10.1111/j.1365-2222.2005.02328.x
- Dill, D., Costill, D., 1974. Calculation of percentage changes in volumes of blood, plasma, and red cells in dehydration. *J. Appl. Physiol.* 37, 247–248.
- Eglin C (2007) Physiological responses to fire-fighting: Thermal and metabolic considerations. *J Human-Environmental Syst* 10:7–18.
- Eglin CM, Coles S, Tipton MJ (2004) Physiological responses of fire-fighter instructors during training exercises. *Ergonomics* 47:483–494. doi: 10.1080/0014013031000107568
- Eglin CM, Tipton MJ (2005) Can firefighter instructors perform a simulated rescue after a live fire training exercise? *Eur J Appl Physiol* 95:327–34. doi: 10.1007/s00421-005-0015-6
- Epstein Y (1990) Heat intolerance: predisposing factor or residual injury? *Med Sci Sports Exerc* 22:29–35.
- Fernhall B, Fahs C a, Horn G, et al (2012) Acute effects of firefighting on cardiac performance. *Eur J Appl Physiol* 112:735–41. doi: 10.1007/s00421-011-2033-x

- Gabriel H, Schwarz L, Born P, Kindermann W (1992) Differential mobilization of leucocyte and lymphocyte subpopulations into the circulation during endurance exercise. *Eur J Appl Physiol Occup Physiol* 65:529–34.
- Gibson, O. R., Mee, J. A., Tuttle, J. A., Taylor, L., Watt, P. W., & Maxwell, N. S. (2015). Isothermic and fixed intensity heat acclimation methods induce similar heat adaptation following short and long-term timescales. *Journal of thermal biology*, 49, 55-65.
- González-Alonso J, Mora-Rodríguez R, Below PR, Coyle EF (1997) Dehydration markedly impairs cardiovascular function in hyperthermic endurance athletes during exercise. *J Appl Physiol* 82:1229–36.
- Greven FE, Krop EJ, Spithoven JJ, et al (2012) Acute respiratory effects in firefighters. *Am J Ind Med* 55:54–62. doi: 10.1002/ajim.21012
- Huang C-J, Webb HE, Garten RS, et al (2010) Stress hormones and immunological responses to a dual challenge in professional firefighters. *Int J Psychophysiol* 75:312–8. doi: 10.1016/j.ijpsycho.2009.12.013
- Ilmarinen R, Koivistoinen K (1999) Heart rate and thermal responses in prolonged job-related fire-fighting drills. *Environ Ergon* VIII ... 99–102.
- Jimenez C, Mathieu J, Peinnequin A, et al (2008) Immune function during and after 60 min of moderate exercise wearing protective clothing. *Aviat Space Environ Med* 79:570–6.
- Jones, A., Doust, J. (1996). A 1% treadmill grade most accurately reflects the energetic cost of outdoor running. *J.SportSci.* 14 (4), 321–327.
- Jürimäe J, Mäestu J, Jürimäe T, et al (2011) Peripheral signals of energy homeostasis as possible markers of training stress in athletes: A review. *Metabolism* 60:335–350.
- Large AA, Owens GR, Hoffman LA (1990) The short-term effects of smoke exposure on the pulmonary function of firefighters. *Chest* 97:806–9.
- Marshall WJ, Bangert SK, Lapsley M (2012) *Clinical Chemistry*, 7th edn. 378.
- Montain, S.J., Sawka, M.N., Cadarette, B.S., Quigley, M.D. and McKay, J.M., 1994. Physiological tolerance to uncompensable heat stress: effects of exercise intensity, protective clothing, and climate. *Journal of Applied Physiology*, 77(1), pp.216-222.
- Moran DS, Shitzer a, Pandolf KB (1998) A physiological strain index to evaluate heat stress. *Am J Physiol* 275:R129–34.
- Mustajbegovic J, Zuskin E, Schachter EN, et al (2001) Respiratory function in active firefighters. *Am J Ind Med* 40:55–62.
- Peters JM, Theriault GP, Fine LJ, Wegman DH (1974) Chronic effect of fire fighting on pulmonary function. *N Engl J Med* 291:1320–2. doi: 10.1056/NEJM197412192912502

- Rejeski W, Kenney E (1987) Distracting Attentional Focus From Fatigue: Does Task Complexity Make a Difference? *J Sport Exerc Psychol* 9:66–73.
- Ribeiro M, de Paula Santos U, Bussacos MA, Terra-Filho M (2009) Prevalence and risk of asthma symptoms among firefighters in São Paulo, Brazil: a population-based study. *Am J Ind Med* 52:261–9. doi: 10.1002/ajim.20669
- Romet TT, Frim J (1987) Physiological responses to fire fighting activities. *Eur J Appl Physiol Occup Physiol* 56:633–8.
- Sawka MN, Burke LM, Eichner ER, et al (2007) American College of Sports Medicine position stand. Exercise and fluid replacement. *Med Sci Sports Exerc* 39:377–90. doi: 10.1249/mss.0b013e31802ca597
- Selkirk GA, McLellan TM (2004) Physical work limits for Toronto firefighters in warm environments. *J Occup Environ Hyg* 1:199–212. doi: 10.1080/15459620490432114
- Sheppard D, Distefano S, Morse L, Becker C (1986) Acute effects of routine firefighting on lung function. *Am J Ind Med* 9:333–40.
- Smith DL, Dyer K, Petruzzello SJ (2004) Blood chemistry and immune cell changes during 1 week of intensive firefighting training. *J Therm Biol* 29:725–729. doi: 10.1016/j.jtherbio.2004.08.046
- Smith DL, Manning TS, Petruzzello SJ (2001a) Effect of strenuous live-fire drills on cardiovascular and psychological responses of recruit firefighters. *Ergonomics* 44:244–54. doi: 10.1080/00140130121115
- Smith DL, Petruzzello SJ (1998) Selected physiological and psychological responses to live-fire drills in different configurations of firefighting gear. *Ergonomics* 41:1141–54. doi: 10.1080/001401398186441
- Smith DL, Petruzzello SJ, Chludzinski M a, et al (2005) Selected hormonal and immunological responses to strenuous live-fire firefighting drills. *Ergonomics* 48:55–65. doi: 10.1080/00140130412331303911
- Smith DL, Petruzzello SJ, Chludzinski MA, et al (2001b) Effect of strenuous live-fire fire fighting drills on hematological, blood chemistry and psychological measures. *J Therm Biol* 26:375–379. doi: 10.1016/S0306-4565(01)00047-X
- Smith DL, Petruzzello SJ, Kramer JM, Misner JE (1996) Physiological, psychophysical, and psychological responses of firefighters to firefighting training drills. *Aviat Space Environ Med* 67:1063–8.
- Smith DL, Petruzzello SJ, Kramer JM, Misner JE (1997) The effects of different thermal environments on the physiological and psychological responses of firefighters to a training drill. *Ergonomics* 40:500–10. doi: 10.1080/001401397188125

- Sothmann MS, Saupe KW, Jasenof D, et al (1990) Advancing Age and the Cardiorespiratory Stress of Fire Suppression: Determining a Minimum Standard for Aerobic Fitness. *Hum Perform* 3:217–236. doi: 10.1207/s15327043hup0304_1
- Stein KD, Jacobsen PB, Blanchard CM, Thors C (2004) Further validation of the multidimensional fatigue symptom inventory-short form. *J Pain Symptom Manage* 27:14–23.
- Thongtang N, Diffenderfer MR, Ooi EMM, et al (2013) Effects of atorvastatin on human C-reactive protein metabolism. *Atherosclerosis* 226:466–70. doi: 10.1016/j.atherosclerosis.2012.11.012
- Toner MM, Drolet LL, Pandolf KB (1986) Perceptual and physiological responses during exercise in cool and cold water. *Percept Mot Skills* 62:211–20. doi: 10.2466/pms.1986.62.1.211
- Williams BE, Petersen SR, Douglas K (1996) Physiological responses of training officers during live fire training. *Fire Eng Sep* 45–52.

Legends:

Fig 1. Schematic of the experimental design with 7wk no-heat exposure and 4wk fire instruction course for the fire service instructors (FSI) group and 11wk no-heat exposure for the control (CON) group. HR = heart rate, T_{re} = rectal temperature, NBM = nude body mass, BP = blood pressure and CO = carbon monoxide.

Fig 2: Mean \pm SD rectal temperature (bars) and heart rate (lines) pre and post Wear 1 (rectal temperature, white bars; heart rate, black dotted) and Wear 2 (rectal temperature, grey bars; heart rate, grey dotted). * denotes a significant difference ($p < 0.05$) between pre and post. † denotes a significant difference ($p < 0.05$) between Wear 1 and Wear 2.

Illustrations:

Fig 1

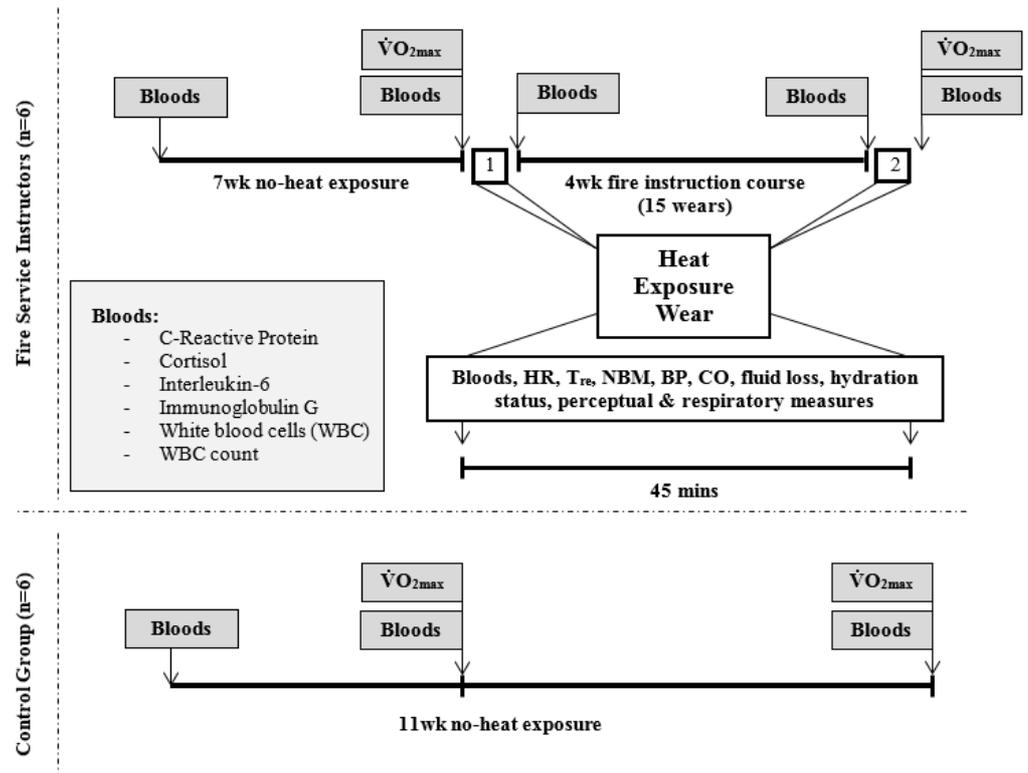
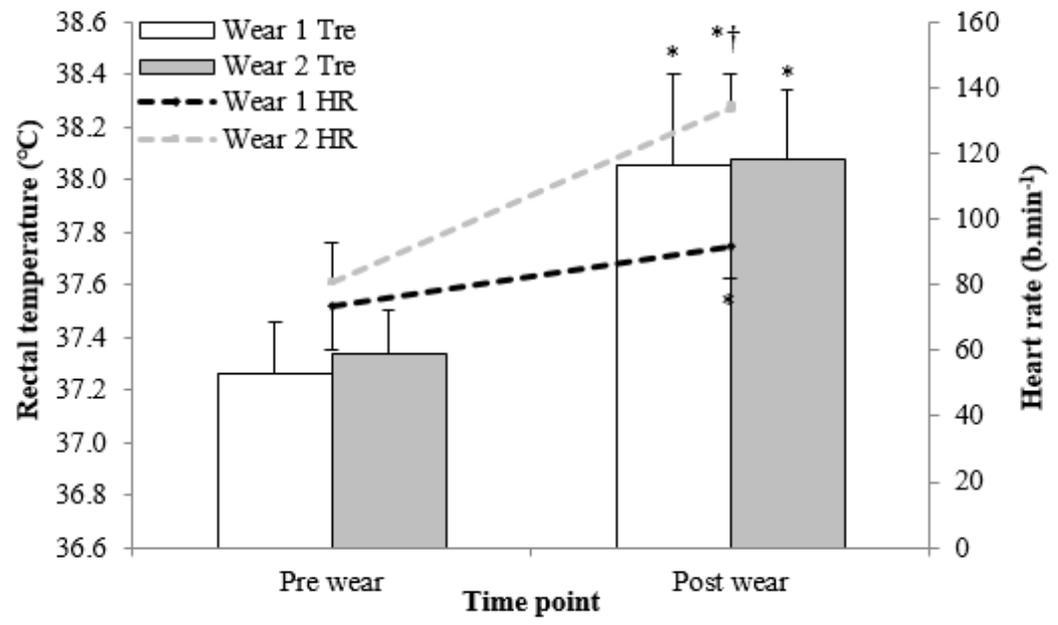


Fig 2.



Tables:

Table 1: Physiological and perceptual changes over the 4 weeks of the fire instruction course for Fire Service Instructors (FSI) and controls (CON) (mean \pm SD).

	First Assessment		Final Assessment	
	FSI	CON	FSI	CON
Age (years)	41 \pm 4	40 \pm 3	-	-
Stature (cm)	178 \pm 1	178 \pm 2	-	-
NBM (kg)	83.3 \pm 11.4	78.1 \pm 8.1	83.1 \pm 11.2	78.1 \pm 6.9
FVC (L)	4.73 \pm 0.70	5.11 \pm 0.68	4.16 \pm 0.65*	4.98 \pm 0.69
FEV ₁ (L)	3.98 \pm 0.58	4.31 \pm 0.65	3.78 \pm 0.56	4.18 \pm 0.69
FEV ₁ / FVC (%)	84 \pm 6	84 \pm 9	91 \pm 6	86 \pm 8
PEF (L)	643 \pm 106	680 \pm 127	633 \pm 119	660 \pm 151
CO (ppm)	5 \pm 8	2 \pm 3	5 \pm 5	2 \pm 0
Systolic BP (mmHg)	134 \pm 16	143 \pm 22	138 \pm 12	140 \pm 8
Diastolic BP (mmHg)	81 \pm 9	87 \pm 9	8 \pm 7	86 \pm 8
$\dot{V}O_{2max}$ (ml.kg ⁻¹ .min ⁻¹)	49.4 \pm 9.1	51.5 \pm 5.9	46.4 \pm 4.9	54.3 \pm 8.9

* denotes significant difference from pre to post 4 weeks of the fire instruction (p<0.05).

Table 2. Physiological measures of the FSI pre and post Wear 1 and Wear 2. (mean \pm SD)

	Wear 1 pre 4wk instruction		Wear 2 post 4wk instruction	
	Pre	Post	Pre	Post
NBM (kg)	83.2 \pm 11.1	82.5 \pm 11.2*	83.1 \pm 11.7	82.3 \pm 11.6*
T_{re} (°C)	37.27 \pm 0.19	38.06 \pm 0.34*	37.34 \pm 0.17	38.08 \pm 0.26*
HR (b.min⁻¹)	74 \pm 13	92 \pm 10*	81 \pm 12	134 \pm 11*†
Average HR (b.min⁻¹)		101 \pm 17		97 \pm 10
PSI		2.4 \pm 0.9		4.1 \pm 1.3
Non-urine fluid loss (L.hr⁻¹)		0.98 \pm 0.46		1.28 \pm 1.07
RPE	6 \pm 0	11 \pm 3*	7 \pm 2	14 \pm 3*
TSS	4 \pm 1	5 \pm 1*	4 \pm 0	6 \pm 1*

denotes significant difference over the 4 weeks of the fire instruction course. * denotes significant difference from pre to post wear. † denotes significant difference between Wear 1 and Wear 2 (p<0.05).

Table 3. Markers of inflammation and immune function in fire service instructors (FSI) and control (CON) groups during no-heat exposure, heat exposure and fire instruction course time periods (mean \pm SD).

Blood Measure	Group	Pre 7wks no-heat exposure period (A)	Pre Wear 1 (B)	Post Wear 1, Pre 4wk instruction (C)	Post 4wk instruction, Pre Wear 2 (D)	Post Wear 2 (E)
WBC ($10^9.L^{-1}$)	FSI	7.1 \pm 0.6 [†]	5.8 \pm 1.5*	6.9 \pm 1.5	5.8 \pm 0.9* [†]	6.9 \pm 1.30
	CON	5.2 \pm 0.5	5.4 \pm 0.8		4.9 \pm 0.3	
Neutrophil ($10^9.L^{-1}$)	FSI	4.1 \pm 0.8 [†]	3.1 \pm 0.9*	4.1 \pm 0.9	3.0 \pm 0.6*	3.8 \pm 0.8
	CON	2.9 \pm 0.6	2.9 \pm 0.6		2.8 \pm 0.4	
Lymphocytes ($10^9.L^{-1}$)	FSI	2.1 \pm 0.6	1.7 \pm 0.5*	2.0 \pm 0.7	1.8 \pm 0.6	2.1 \pm 0.6
	CON	2.0 \pm 0.3	2.1 \pm 0.4		1.7 \pm 0.3	
Monocytes ($10^9.L^{-1}$)	FSI	0.5 \pm 0.1	0.5 \pm 0.2	0.5 \pm 0.1	0.4 \pm 0.1*	0.5 \pm 0.1
	CON	0.5 \pm 0.1	0.5 \pm 0.1		0.4 \pm 0.1	
Eosinophil ($10^9.L^{-1}$)	FSI	0.1 \pm 0.1	0.2 \pm 0.1	0.1 \pm 0.1	0.2 \pm 0.1	0.2 \pm 0.1
	CON	0.1 \pm 0.1	0.1 \pm 0.1		0.1 \pm 0.0	
Basophil ($10^9.L^{-1}$)	FSI	0.03 \pm 0.01	0.03 \pm 0.01	0.04 \pm 0.01	0.03 \pm 0.02	0.03 \pm 0.01
	CON	0.02 \pm 0.01	0.02 \pm 0.01		0.02 \pm 0.01	
IL-6 (pg.ml⁻¹)	FSI	17.0 \pm 5.7 [†]	7.4 \pm 1.5*	9.9 \pm 4.4 [‡]	11.4 \pm 1.0* [†]	16.6 \pm 2.9
	CON	5.1 \pm 2.0	4.9 \pm 2.5		5.3 \pm 2.3	
Crt (ng.ml⁻¹)	FSI	4919 \pm 2605	8453 \pm 3412*	5814 \pm 1709	7540 \pm 2914	6606 \pm 2310
	CON	8976 \pm 4001	8295 \pm 4186		9114 \pm 3445	
CRP (ng.ml⁻¹)	FSI	3564 \pm 3806	8506 \pm 6392	4790 \pm 3129	7378 \pm 3770	6631 \pm 3461
	CON	3320 \pm 5163	2411 \pm 2775		3374 \pm 3403	
IgG (mg.ml⁻¹)	FSI	17.1 \pm 3.2	21.6 \pm 8.0	19.3 \pm 7.0	15.9 \pm 4.8	10.6 \pm 2.9*
	CON	19.9 \pm 6.9	17.8 \pm 4.5		14.1 \pm 3.3	

[†] denotes a significant difference (p<0.05) between FSI and CON. * denotes a significant difference (p<0.05) between pre washout measures and another time point. ⁺ denotes a significant difference (p<0.05) between time point B and D in FSI. [‡] denotes a significant difference (p<0.05) between time point B and C. Fire Service Instructors (FSI), Controls (CON). WBC = white blood cells, IL-6 = interleukin-6, Crt = cortisol, CRP = C-reactive protein, IgG = immunoglobulin G.

