Chapter 5

Protection of total body water content and absence of hyperthermia despite 2% body mass loss ("voluntary dehydration") in soldiers drinking *ad libitum* during prolonged exercise in cool environmental conditions

Article:

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5.1 Introduction

The extent to which humans need to replace their fluid losses during exercise remains contentious despite more than 60 years of focused research.[2,22,32,33,34,42,48,58,63] Prior to the 1970's athletes were encouraged to avoid drinking during exercise since it was believed that fluid ingestion impaired exercise performance.[47] This despite the evidence collected in the classic Nevada desert studies [2] which showed that soldiers benefited from ingesting fluid during 8 hour marches in the desert. At that time drinking guidelines for the US military prescribed only the need to drink within a daily range of fluid intakes, for example from 4-12 litres/day in hot environments.[37,41]

However after the mid-1990's guidelines were introduced that encouraged soldiers to drink up to 1.8 L of fluid for each hour that they were on active duty in hot environments (WBGT>30°C).[5,30,41] These guidelines which mirrored those of the American College of Sports Medicine (ACSM) [15] invoked the theory that only by maintaining the pre-exercise body mass could exercisers ensure that they would not suffer an impaired exercise performance or risk the development of heat illness.[5,30]

The adoption of these guidelines had two consequences. First was an increased prevalence of exercise-associated hyponatremia (EAH) [21,51] sometimes associated with an encephalopathy (EAHE) that proved fatal in a small number of US soldiers [56] and marathon runners.[49] Subsequently those guidelines were revised with the result that the incidence of EAH and EAHE in the US military has fallen substantially

[21] as it has also amongst marathon runners and other endurance athletes (Noakes TD, 2010). Second, these guidelines required soldiers to carry more water which led to an increased load while on active duty.[44,54]

While it is now accepted that exercisers should not be encouraged to drink "as much as tolerable" there is still no consensus of the optimum rate of fluid ingestion during exercise. The recently modified ACSM guidelines advise that exercisers should drink sufficiently to ensure that their body mass loss during exercise is less than 2%.[64] More recently the US Army Research Institute of Environmental Medicine (USARIEM) proposed sweat loss prediction equations ranging from ~575 g/hr to ~1092 g/hr (based on an individual with a body surface are of 1.9 m^2) for various workloads, environmental and clothing configurations in order to more accurately predict fluid replacement volumes during work and exercise.[22] Body mass losses of up to 2% are often regarded as "voluntary dehydration". This term refers to the observation that humans does not voluntarily drink as much water as believed to have been lost (using body mass losses as a measure) even when water is readily available. Some believe that voluntary dehydration occurs since the thirst mechanism is an inadequate stimulus to drinking.[2,34,58] Others [48,52,54,73] argue that drinking to the dictates of thirst is the biologically appropriate behaviour that optimizes performance and prevents heat illness regardless of the exact level of dehydration that develops during exercise.

This debate has special relevance for the military since soldiers ingesting fluid *ad libitum* will always drink less that those who are forced to drink in order to lose less than 2 % of their body mass during exercise. Many soldiers drinking *ad libitum* according to the dictates of their thirst will drink less, develop “voluntary dehydration” and will therefore need to carry less water during military operations. To distinguish between these possibilities, there is a need for field studies to establish the optimum rates at which soldiers should ingest fluid during exercise.

Accordingly the primary objective of this study was to evaluate the effect of an *ad libitum* fluid replacement strategy on selected hydration status markers to determine whether this approach could prevent “dehydration”. In addition we wished to determine whether body mass loss can be used as an accurate surrogate measure of the changes in total body water during exercise. For example body mass loss during exercise that may at least theoretically not contribute to body water losses include substrate oxidation and the release of water associated with the storage of glycogen (if that water does not constitute part of the total body water measured with stable isotope tracers). Thus many scientists [39,44,59,61,71] conclude that some degree of body mass loss can be explained by losses other than water so that some degree of body mass loss (not replacing all body mass losses through fluid intake) is essential to maintain normal plasma osmolality especially during prolonged exercise. This is because the POsm and not the body mass is the homeostatically regulated variable during exercise.[26] Others have argued since the Second World War, that the body contains a 2L fluid excess that can be lost before there are any effects of "dehydration".[32,33,34]

Thus the scientific hypothesis that we tested was that *ad libitum* fluid replacement is effective in protecting TBW despite body mass loss during prolonged exercise in cool environmental conditions since this method of drinking is driven to maintain plasma and tissue osmolality.

5.2 Materials and methods

Subject Selection

Ethical clearance for this study was obtained from the Research Ethics Committee from the South African Military Health Services (SAMHS) of the South African National Defence Force (SANDF). All soldiers taking part in an official military exercise (N = ~ 250) were eligible for the study; twenty Operational Emergency Care Practitioners (OECPs) were identified and invited to volunteer for this study. All the subjects were experienced and conditioned to route marches with payloads of up to 35 kg; they were medically fit to participate in the study and were without any musculoskeletal injuries. Subjects were told that they could terminate their participation at any stage without any consequences to their military careers. All subjects were required voluntarily to read and sign an informed consent. The subjects were asked to provide basic demographic information for record purposes. Three days prior to the route march, the sub-maximal oxygen consumption of each subject was directly measured using a MetaMax™ portable gas analyser (Cortex Biophysik, Germany) during the Harvard graded step-up test. Their predicted aerobic capacity was calculated from the sub-maximal exercise test results and the resulting individual regression equation for heart rate versus oxygen consumption at different workloads according to the method described in ISO 8996.[28]

Exercise intervention

The exercise intervention took the form of a competitive route march of 14.6 km. Individuals competed against each other to complete the route march in the fastest time possible without running. Each individual had to carry a mass of 26.5 kg including 4 litres of water, rifle and bush hat or cap. Participants were dressed in standard issue SANDF combat dress. Water was available for additional replenishment if required at two points along the route. The subjects drank according to the dictates of their thirst (*ad libitum*) during the march. The core body temperature of the subjects was recorded at one minute intervals with a CorTemp ^{TM}2000 (HQ Inc, USA) ambulatory remote sensing system. Core temperature data were evaluated for the potential confounding effect of fluid ingestion invalidating the ingestible sensor.[37] The ambient temperature, wind speed, relative humidity, solar radiation were recorded for the duration of the experiment (WBGT Temperature Measurement System, Questtemp, Quest Technologies, South Africa).

Hydration markers

Prior to the exercise intervention each subject emptied his/her bladder and provided a urine and saliva sample for analysis for urine specific gravity (USG), urine osmolality (UOsm) and deuterium abundance (saliva). Saliva was chosen due to the ease and non-invasive nature of its collection, as well as the fact that saliva has been proven as a valid and convenient sampling medium for determining TBW through the diluted isotope technique.[25,65,74] Furthermore it has been documented that enrichments of deuterium oxide in saliva and plasma samples were identical and reached a 2 hour plateau after administration of an oral dose of the tracer. Determining TBW through the diluted isotope technique remains the most reliable method currently available, producing lower coefficient of variation values than methods such a bioelectrical impedance.[65] Nude body mass were obtained on a scale accurate to 0.1 kg (Scale CPW 150, Adam equipment). Deuterium oxide (99%) was used to prepare a 4% (weight to weight) solution with water. This solution was then used to prepare the individual deuterium oxide doses according to individual body mass (± 0.05 g/kg body mass). Appropriate weighing of the dose bottle (to the nearest 0.1g) was performed in order to determine the exact dose consumed by each participant. After a two hour equilibration period [9,13,17,18,19,27,31,38,66,69,70,73,74], a second saliva sample was collected in order to determine the pre-exercise total body water (TBW). At the completion of the exercise each participant was provided with a towel to dry excess perspiration prior to re-weighing. A second urine sample and third saliva sample were collected and used for the determination of post-exercise USG, UOsm and deuterium abundance (saliva). The participants then received their post-exercise deuterium oxide dose followed by a two hour equilibration period. Urine voided during this period was recorded for correction of isotope loss. A final saliva sample was collected and body mass measurement performed in order to calculate post-exercise TBW.

Total body water (kg) was calculated using the preferred method of Halliday and Miller [25] according to the following equation:

TBW (Kg) = ((((T*A)/a)*((Ea-Et)/(Es-Ep)))/1000) / 1.04, in which

A = amount of dose solution drunk (grams)

a = amount of dose solution diluted in T (grams)

T = amount of tap water 'a' was diluted in

Ea = enrichment of diluted dose

Et = enrichment of tap water used to dilute the dose

Ep = enrichment of baseline sample.

Es = enrichment of post dose sample

1.04 = correction factor for over estimation due to exchange with non-aqueous hydrogen

Diluted isotope methods designed to measure TBW at the time of isotope administration are subject to systematic errors from water entering the body between the time of dosing and the sample collection.[65] Corrections were made for the ingestion of the isotope dose, metabolic water production and water added to the TBW pool through exchange with atmospheric moisture according to the methods of Schoeller et al.[65] Since most of the correction factors depend on metabolic rate, additional corrections were made for the post-exercise equilibration period during which an increased metabolic rate (excess post exercise oxygen consumption (EPOC), although marginal [8,23,35] would augment the over-estimation through increased metabolic water production. The sweat losses of the participating soldiers were calculated according to method previously described by Rogers et al.[61] Respiratory water loss was calculated by the methods of Mitchell et al.[40] For all calculations and estimations involving respiratory exchange ratio (RER) and oxygen consumption (VO_2), we assumed that the RER averaged 0.85 and that the oxygen consumption averaged 65% of VO_{2max} throughout the exercise.[16,45,61] Even if the actual RER and VO_2 of the participants differed slightly from these assumptions, the outcomes would not have been greatly different.

Statistical analyses

In order to determine which statistical test would be most suited for the comparisons of pre- and post-exercise values, the differences (paired differences) between the pre- and post-exercise results were calculated. The distributions (in the form of histograms) of paired differences of all the results were plotted with the number of classes as calculated according to the Rule of Sturge. The normality of this distribution was tested by means of the Shapiro-Wilks' W test. The statistical Rule of Sturge states that the number of classes equals N x 1.4 + 1 (where: N = sample size). Student's T-tests were used to compare results where the distribution of the paired differences was normal. Where the distribution of the paired differences was not normal, the non-parametric alternative to the student's T-test, the Wilcoxon Rank Sum Test was used to compare results. A Pearson's product moment correlation coefficient was used to determine relationships between appropriate variables. Statistical significant differences were indicated by a p-value of less than

0.05. The statistical analyses were completed using the STATISTICA© software package.[72]

5.3 Results

Twenty participants volunteered for the study. Sixteen of these subjects were male, four were female. The mean stature of the male and female participants was 1.72 ± 0.05 m and 1.58 ± 0.04 m respectively. The mean predicted VO_{2max} of the male and female participants was 45 ± 10.3 ml/kg/min and 37.8 ± 2.3 ml/kg/min respectively. Two men failed to provide sufficiently large saliva samples required for deuterium abundance analysis and their data were therefore excluded from all analyses. On average the subjects carried a pay load mass of 26.5 kg each. The mean Wet Bulb Globe Temperature (WBGT) index value for the duration of the exercise period was 14.1°C.

Body mass loss, fluid intake and sweat rates

Body mass was reduced significantly ($p<0.05$) during exercise, on average the group lost 1.3 ± 0.5 kg (Table 1). The group consumed on average 850 ± 594 ml/hr during the march (Table 1). The mean sweat rate was 1289 ± 530 ml/hr (Table 1). There was no significant relationship ($p>0.05$) between exercise time and rates of fluid intake (r = -0.13), changes in % body mass (r = 0.18) and sweat rates (r = -0.25). Figure 1 indicates the total fluid intake, hourly fluid intake as well as predicted hourly sweat rates.

Table 1: Summary of change in body mass, TBW, USG and UOsm during exercise

	Pre-exercise mean (± SD)	Post-exercise mean (± SD)	% Change mean (± SD)
Body mass [kg]	63.7 (6.7)	62.4 (6.5)	-1.98 (0.6)
TBW [kg]	37.07 (5.8)	37.26 (6.0)	0.53 (4.0)
USG	1.023 (0.002)	1.021 (0.007)	-0.15 (0.0)
UOsm [mmol/kg]	977.6 (187.3)	924.5 (164.3)	-4.10 (14.7)

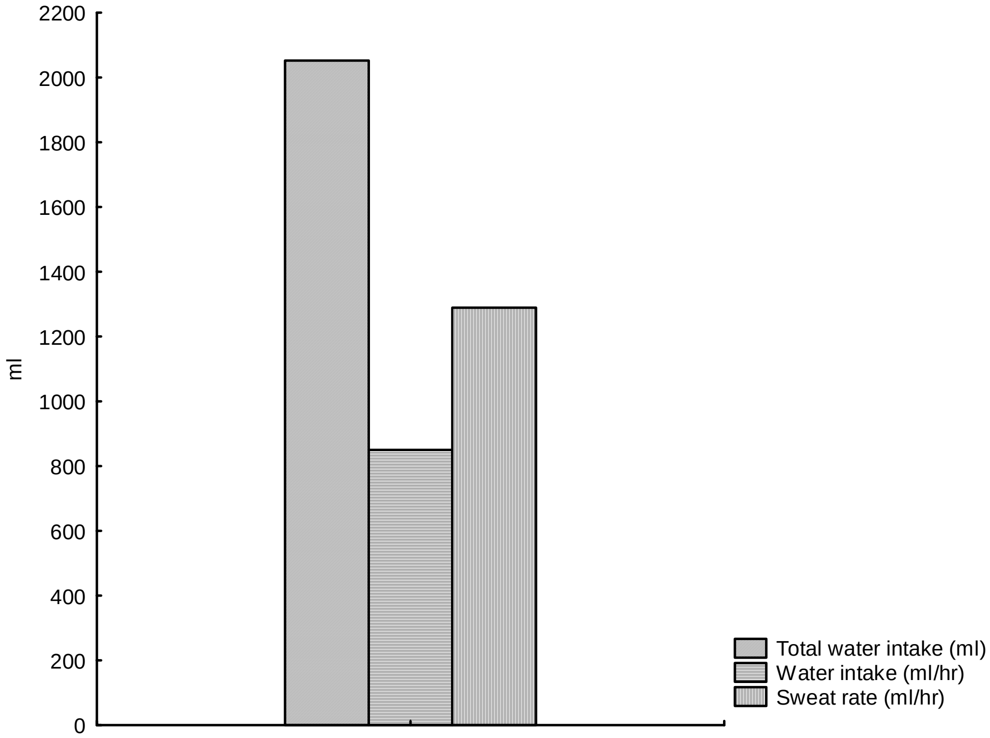

Figure 1: Mean total fluid intake, hourly fluid intake and predicted hourly sweat losses

Urine specific gravity (USG) and urine osmolality

Neither the USG nor the UOsm changed significantly during the exercise period (Table 1). Figure 2a and 2b show that there was no significant relationship between either USG or UOsm and TBW at the end of exercise or between changes in these variables during exercise.

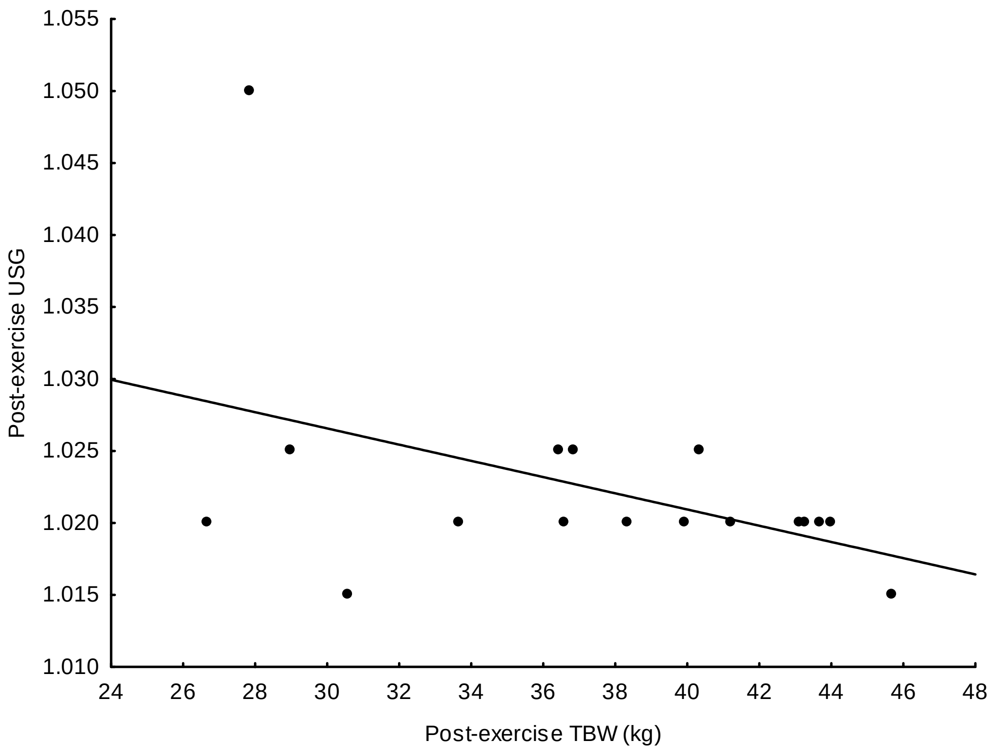

Figure 2a: The relationship between post-exercise TBW and USG ($p>0.05$; $r = 0.53$)

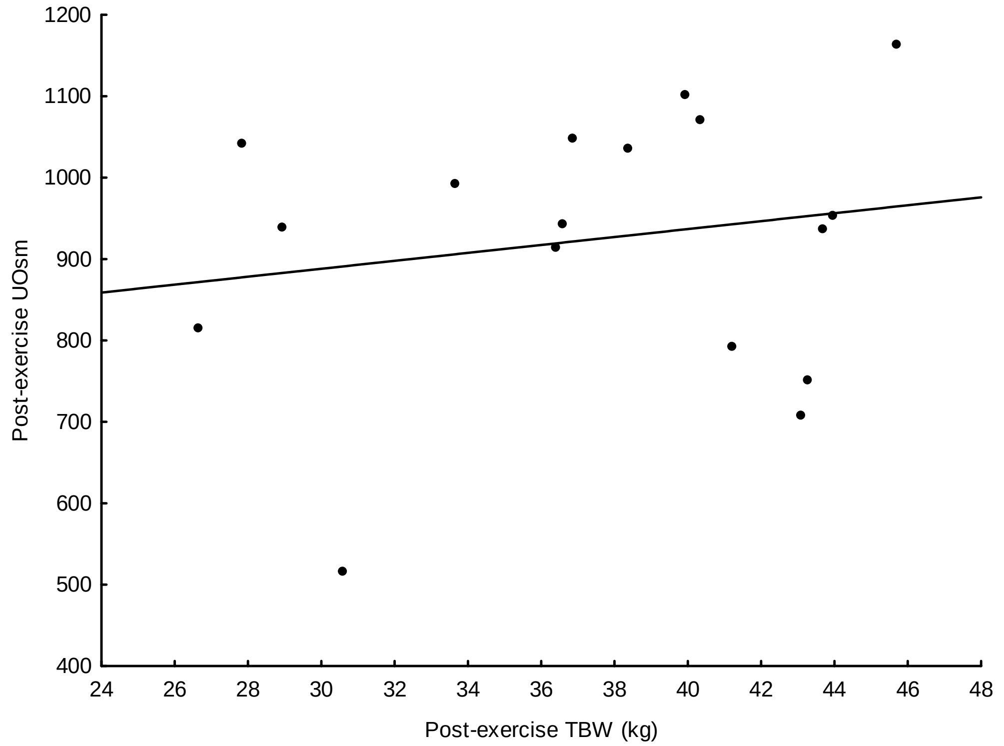

Figure 2b: The relationship between post-exercise TBW and UOsm ($p>0.05$; $r = 0.15$)

Core temperature measurements

The average peak core temperature of the soldiers during the exercise was 38.9 ± 0.3°C while the highest individual peak core temperature was 39.6°C. Figure 3a shows that there was no relationship between core temperature and the change in body mass during exercise. However there was a significant positive linear relationship between total fluid intake and core temperature during exercise (Figure 3b). This could be explained by significant relationships between finishing time (marching speed) and core temperature (Figure 3c) as well as sweat rate (Figure 3d).

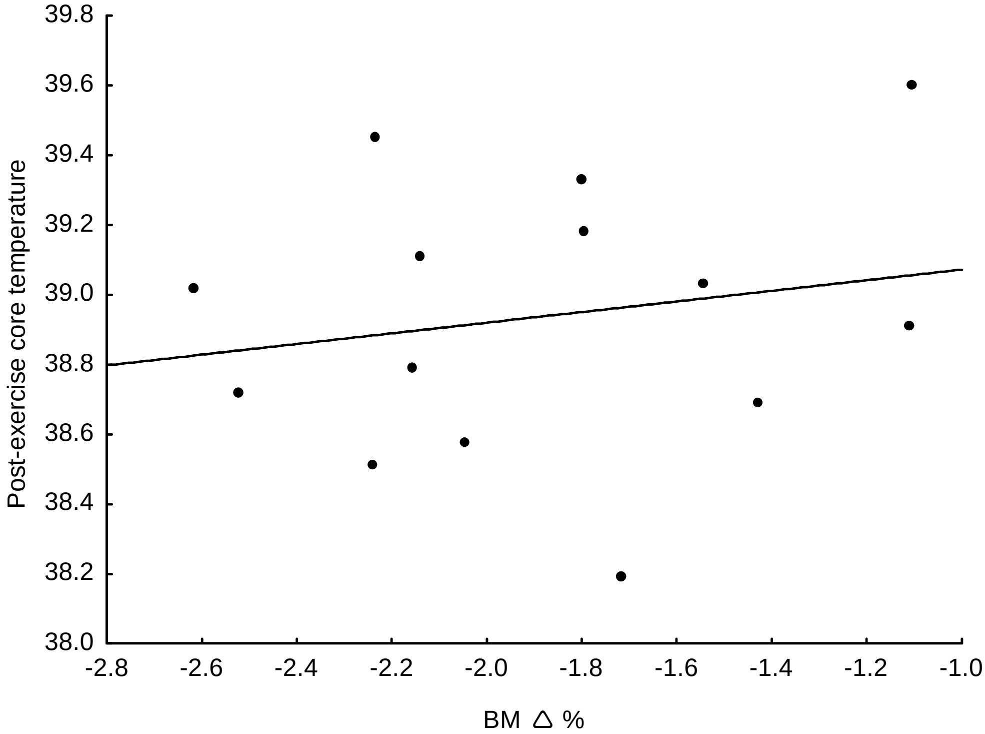

Figure 3a: The relationship between core temperature and changes in body mass ($p>0.05$; $r = 0.18$)

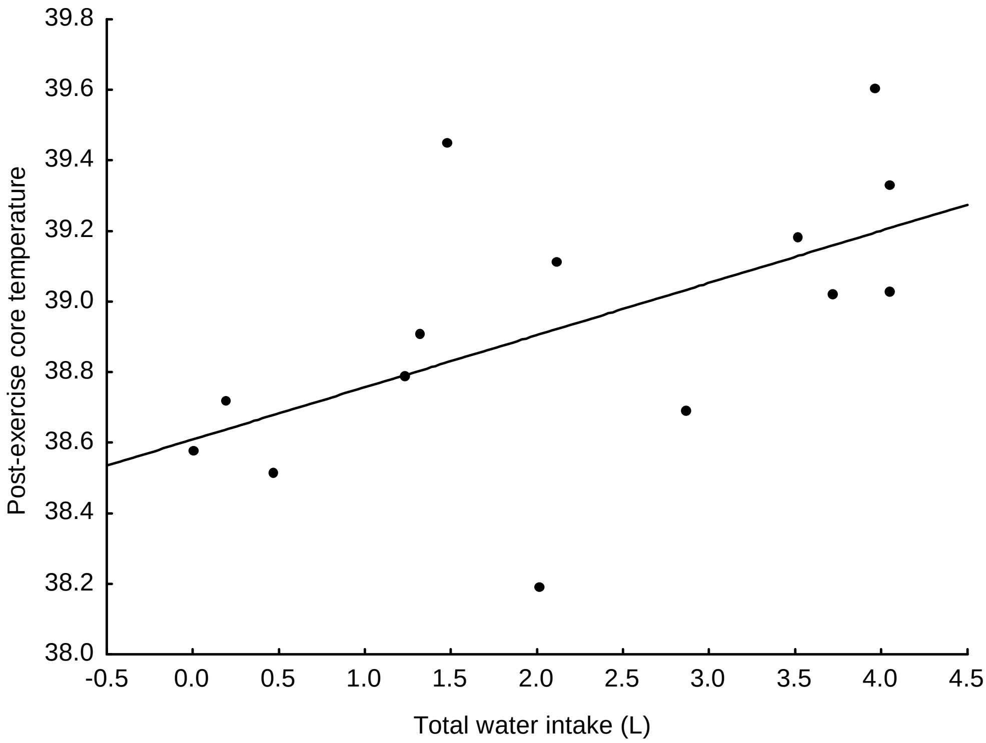

Figure 3b: The relationship between core temperature and total water intake ($p<0.05$; $r = 0.56$)

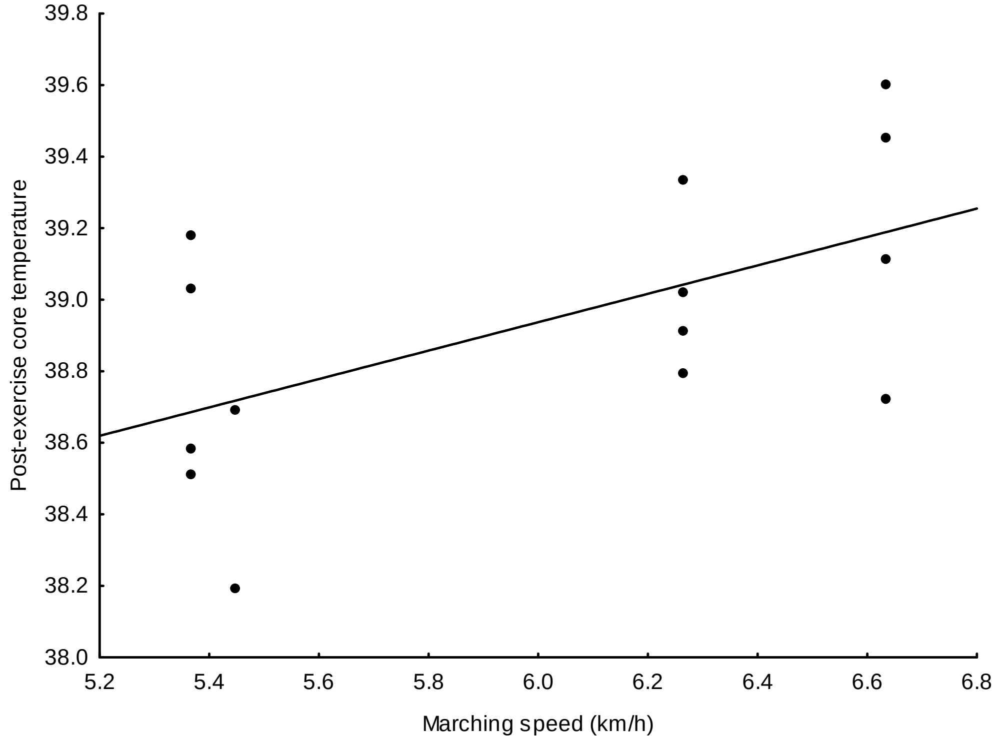

Figure 3c: The relationship between core temperature and marching speed ($p<0.05$; $r = 0.58$)

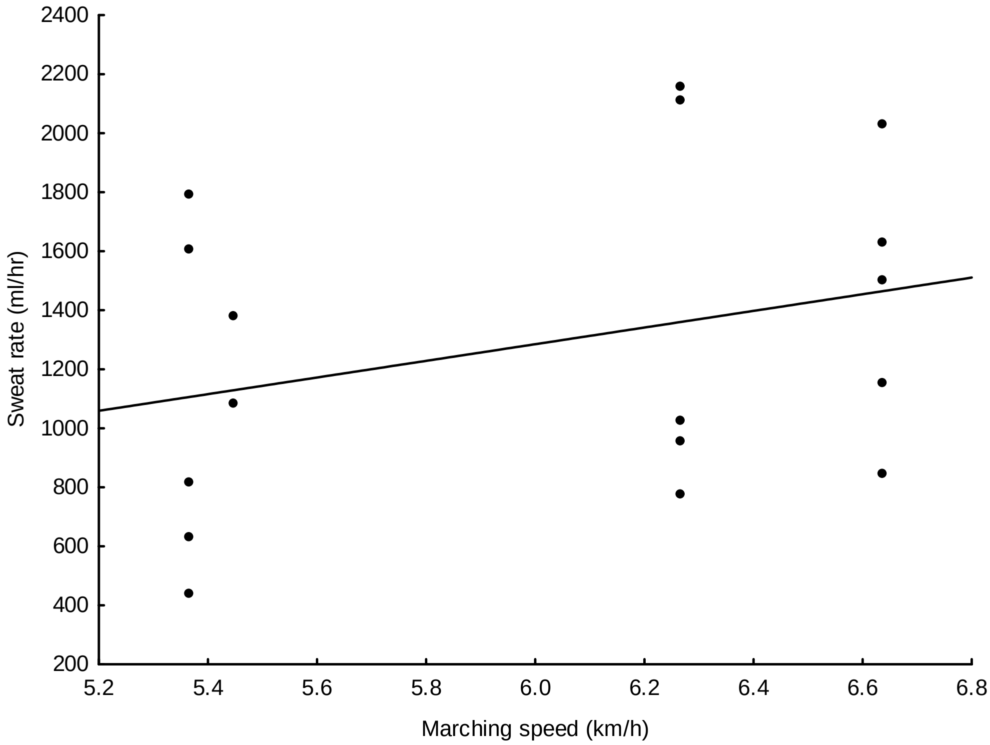

Figure 3d: The relationship between marching speed and sweat rate ($p>0.05$; $r = 0.29$)

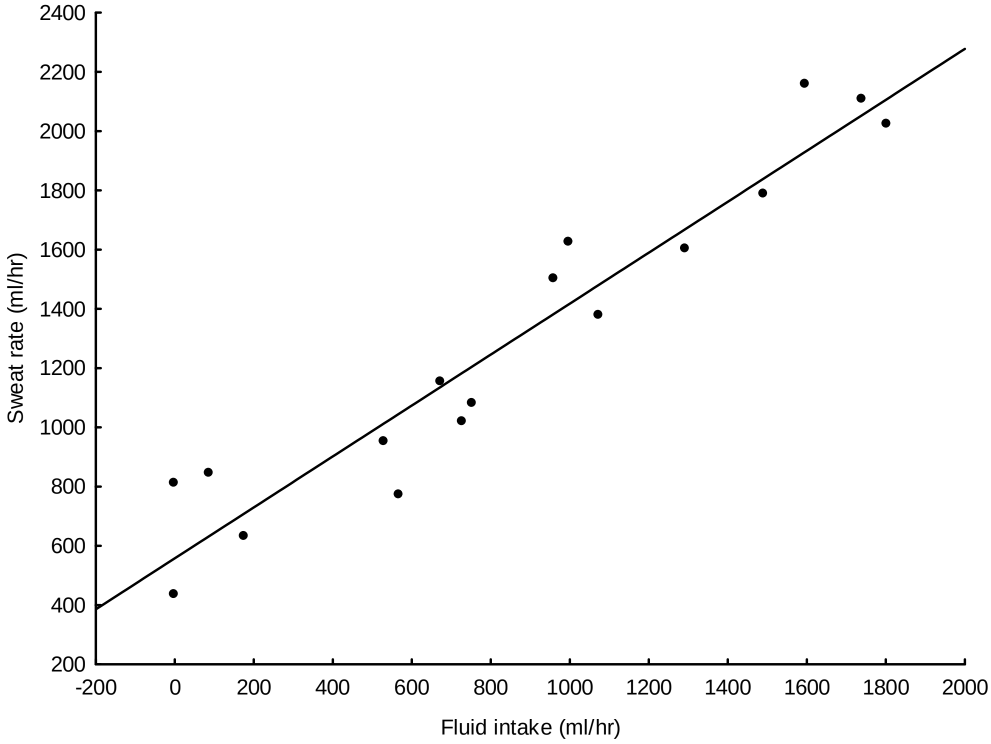

Figure 3e: The relationship between fluid intake and sweat rate ($p<0.05$; $r = 0.96$)

Total body water measurements

Table 1 presents the pre- and post-exercise TBW results. Mean TBW did not change despite 1.3 ± 0.5 kg body mass loss. Figures 4 shows that changes in these variables were unrelated. Of the 18 subjects that were tested, all lost between 0.7 and 2.4 kg body mass. Yet total body water increased in 9, stayed the same in 2 and was reduced in 6 subjects.

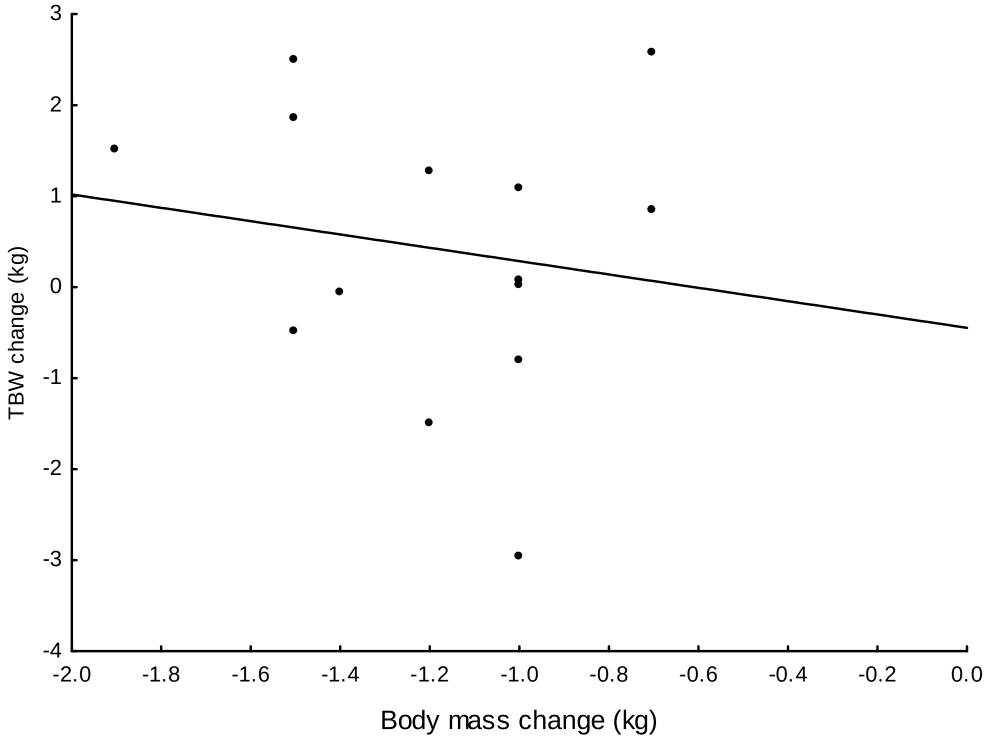

Figure 4: The relationship between body mass change and TBW change (p>0.05; r = -0.16)

5.4 Discussion

The first important finding of this study was that there were no significant changes to TBW, USG or UOsm in any of the subjects despite a significant ($p<0.05$) average body mass loss of 1.3 kg (1.98%). Thus although the subjects developed "voluntary dehydration" as classically described [2] they did not show a decrease in TBW and so were not "dehydrated". Instead TBW increased marginally by about 197 g during the route march. This increase in the TBW occurred even though the mean *ad libitum* fluid intake was only 850 ± 594 ml/h, substantially less than values of 1.2 L/hr originally prescribed by the 1996 ACSM guidelines. An important finding considering that this rate of intake was less than their hourly fluid loss. These findings suggest that changes in body mass may not accurately predict changes in TBW and raise doubts about the accuracy of using body mass losses during exercise as a surrogate marker for changes in TBW at least in the range of body mass losses that we measured.[7]

Maughan et al. [39] have recently reviewed the possible reasons why water loss alone may not explain all the mass loss during exercise. First is the production of metabolic water during fuel consumption. This endogenous source of water gain involves the transformation of one form of body mass (fat and carbohydrates) to another form (H_2O) with the exhaled loss of CO_2.

A second source of water gain during exercise is the intake of exogenous water in the form of either water or the water present in food eaten during exercise. A third theoretical source is the release of water with the breakdown of muscle and liver glycogen. It has been estimated that 3-4 grams of water may be complexed with each gram of glycogen stored in the liver or muscles.[57] Since humans can store at least 450 g of glycogen [1,20], in theory at least, 1350g of water could be stored in this way.[57] This water would become available to the TBW pool even when there is a body mass loss resulting from irreversible glycogenolysis.[61] It has been calculated that an athlete who loses 2 kg of mass during a marathon race could, in fact, be dehydrated (fluid loss) by only ~ 200 g when allowance is made for the body mass loss and alternatively body water gain from these three sources.[39,50,59] Calculations based on the method of Rogers et al.[61] predict the mean rate of carbohydrate oxidation during this study to be approximately 60 ± 15 g/hr. Considering the assumptions regarding water associated with glycogen storage and the mean duration of the route march a mean of 436 ± 113 g of water could have become available to our subjects during this march, this excludes exogenous water as well as metabolic water production.

While the presence of this fluid store remains hotly debated [7,39,53], the findings of this study indicate that the TBW was unchanged in subjects who lost an average of 1.3 kg during exercise. This was also found in our earlier study.[54] This finding is therefore compatible with the presence of an endogenous body water source that is released during exercise and which therefore "protects" the TBW despite a body mass loss during exercise. While the source of this fluid may be uncertain [42] this does not negate the importance of our finding that up to 1.3 kg of this mass lost during exercise may not be due to this loss of water from the TBW as measured with the diluted isotope (deuterium oxide) method. Indeed this finding is compatible with the theory first proposed by Ladell during and after the Second World War.[32,33,34] In particular Ladell [32,33,34] observed, as did we, the loss of 2kg body mass prior to any urine effects becoming noticeable.

There are a number of other findings in the literature which are compatible with this interpretation. Thus Astrand and Saltin [6] found that another indicator of hydration status, plasma volume, increased during an 85 km ski race despite an average 5.5% decrease in body mass. Colt et al. [14] found that TBW increased by 2.4% during a 16 km foot race in subjects who lost 2.3% of body mass. Similarly Pastene et al. [59] also reported that plasma volume did not fall in 42 km marathon runners despite a significant decrease in body mass (2.0 kg). The author suggests that the production of 402 g of metabolic water and the release of 1 280 g of water stored in glycogen complexes in muscle and liver prevented a change in TBW despite this body mass loss of ~2 kg.[59] Similar findings were reported in soldiers performing a 194 km unsupported desert march during which their mean hourly fluid intakes of 458 ml maintained thermoregulation (mean core temperature of 38.1°C) while producing a 300 g increase in TBW despite a body mass loss of 3.3 kg at the end of exercise.[44]

In addition, Speedy et al. [71] showed that serum [Na^+] was maintained despite an average body mass loss of 2.5 kg during a 226 km Ironman triathlon. Significantly subjects in that study did not regain body mass losses until 24 hours after the race compatible with the theory that the pre-race body mass is not regained until the glycogen stores with (a theoretical) associated mass of water are fully replenished. Laursen et al. [36] also found that changes in body mass were unrelated to core temperature, plasma [Na^+] or urine specific gravity at completion of another 226 km Ironman triathlon. The authors concluded that body mass losses of up to 3% are well tolerated by properly trained athletes exercising in warm conditions and who show no evidence for thermoregulatory failure.

Thus all these studies show that significant loss of body mass, perhaps between 1-3%, may be incurred without the development of significant hypohydration.[39] Indeed the most recent ACSM Position Stand acknowledges that a body mass loss of up to 2% may not incur any physiological cost. Our data are compatible with that interpretation as are the historical studies of Ladell.[32,33,34]

However, we and others have also shown that much greater mass losses occur in successful athletes who drink *ad libitum* during prolonged exercise.[67,68,71] Indeed the study of Kao et al. [29] found that the athletes who lost the most body mass (%) during a 24 hour race ran the furthest. In that study there was a linear relationship with a negative slope between the distance run in 24 hours and the extent of body mass loss. Data on marathon runners reviewed by Cheuvront et al. [12] also found that those who lost the most mass during the race also ran the fastest.

Thus we do not exclude the possibility that body mass losses greater than 2% may also not carry adverse physiological cost in those who drink according to the dictates of their thirst during prolonged exercise.[63]

Instead we conclude that these data suggests that the validity of the term “voluntary dehydration” needs to be re-considered since it now seems clear that some body mass loss can occur in athletes drinking *ad libitum* during exercise without the decrease in TBW that is necessary for the use of the term “dehydration”.

Our second important finding was that there was no relationship between % body mass loss and the peak exercise core temperature as now frequently reported.[10,46,68] The core body temperature of all the participants rose steadily from the start of the exercise, but did not exceed 40°C let alone 42°C; which is considered the danger level for core body temperature resulting in serious health consequences.[55] Instead core temperatures were homeostatically regulated within a normal range unrelated to the degree of mass loss during exercise.

Cheuvront and Haymes [11] and Byrne et al. [10] have reported similar findings in athletes drinking *ad libitum* during exercise. Paradoxically the subject who developed the highest core body temperature (39.6°C) in this study was also the subject who consumed fluid at the highest rate (1800 ml/h), thus presenting with the lowest level of dehydration. This subject lost 0.7 kg body mass while replacing 89% of his high sweat losses (2026 ml/hr). He was also amongst the first group of finishers (Figure 3c). The finding that race winners in endurance events are usually both the hottest and the most dehydrated is frequently observed [43,60,75] yet infrequently acknowledged.

Thirdly we found no relationship between changes in TBW and urinary markers of “dehydration”. Thus there were no significant correlations between either UOsm or USG and TBW changes during exercise as also reported by others.[24,62] This suggests that these markers should not be used as a measure of hydration status in athletes. This conflicts with some popular guidelines [3,4] but reflects the historical evidence.[32,33,34]

Finally, we found a relationship between the rates of fluid intake and of sweating (Figure 4e). We are unaware of other studies that have evaluated this relationship. Nor do we know whether these variables are causally related or explained by their co-dependence on a third variable, for example, the exercising metabolic rate.

Conclusion

In summary, there were a number of important findings of this study which add to the debate on the extent to which fluid and weight losses incurred during exercise need to be replaced. Until recently it was advised that full replacement of body mass losses should be achieved during exercise.[15] Currently this position has been revised so that the newest ACSM Position Stand proposes that the mass loss during exercise should not exceed 2% of the starting body mass.[64] Thus our findings could be interpreted as supportive of that proposal since we showed that subjects maintained their pre-exercise TBW despite an average body mass loss of 1.3 kg (equivalent to a 2% body mass loss) and showed no evidence for any homeostatic failure since serum [Na^+], urinary osmolality and core body temperatures did not change substantially. However this conclusion could also be an artefact of the study design in which subjects lost only ~2% body mass during the exercise intervention.

Left unanswered by our study is whether higher levels of body mass loss during more prolonged exercise, for example losses of 4-8% as reported recently in athletes competing in 160 km/ 24 hour races [29] and which losses appear to be homeostatically regulated since they remained constant and did not increase after ~8 hours, are also associated with an unchanged TBW. Alternatively, whether large changes in body mass cause changes in TBW that is associated with deleterious physiological changes such as significant reductions in plasma volume and/or electrolyte imbalances.

Regardless, our study raises questions about the validity of the term "voluntary dehydration" that was first coined more than 60 years ago. Additional studies during more prolonged exercise in which athletes undergo greater changes in body mass are required to determine whether "voluntary dehydration" does indeed occur in those who ingest fluids *ad libitum* during more prolonged exercise.

Indeed this study invites a more thorough interrogation of the use of the term "dehydration" which should be used only when there is a proven reduction in TBW and not, as this study shows, merely a reduction in body mass during exercise.

Acknowledgement

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