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Pre- and Post-Race Hydration Status in Hyponatremic and Non-Hyponatremic Ultra-Endurance Athletes

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Abstract

The monitoring of body mass, plasma sodium [Na$^+$] and urinary specific gravity (U_{sg}) are commonly used to help detect and prevent over- or dehydration in endurance athletes. We investigated pre- and post-race hydration status in 113 amateur 24-h ultra-runners, 100-km ultra-runners, multi-stage mountain bikers and 24-h mountain bikers, which drank *ad libitum* without any intervention and compared results of hyponatremic and non-hyponatremic finishers. On average, pre-race plasma [Na$^+$] and both pre- and post-race levels of U_{sg} and body mass were not significantly different between both groups. However, nearly 86% of the post-race hyponatremic and 68% of the normonatremic ultra-athletes probably drank prior the race greater volumes than their thirst dictated regarding to individual pre-race U_{sg} levels. Fluid intake during the race was equal and was not related to plasma [Na$^+$], U_{sg} or body mass changes. A significant decrease in post-race plasma [Na$^+$], body mass and an increasement in post-race U_{sg} was observed in hypo- and normonatremic finishers. Moreover, pre-race plasma [Na$^+$] was inversely associated with post-race percentage change in plasma [Na$^+$], and pre-race U_{sg} and urinary [Na$^+$] with percentage change in U_{sg} in both groups with and without post-race EAH. Thirteen (11.5%) finishers developed post-race EAH (plasma [Na$^+$] < 135 mM). The incidence of EAH in ultra-endurance athletes competing in the Czech Republic was higher than reported previously.

Key Words: mountain bikers, plasma sodium, runners, urinary sodium

Introduction

The monitoring of body mass changes, post-race plasma [Na$^+$] concentration and the assessment of urinary specific gravity (U_{sg}) are commonly used to help detect and prevent over- or dehydration in endurance athletes and provide sufficient information about hydration status in field conditions. Rapid recognition and treatment of de- and overhydration are essential to prevent morbidity and mortality in endurance activities.

A significant post-race decrease in body mass (9, 10, 14, 20, 21, 25-27, 30, 34, 36-39, 43, 46, 47, 50, 52, 53, 62, 65, 71, 73), increases in U_{sg} (26, 34, 36-39, 44, 46, 47, 53, 65) and a decrease in plasma [Na$^+$] (1, 8, 19, 20, 25, 26, 34, 39, 46, 50, 51, 54, 69-71) are generally found in ultra-endurance athletes. Kavouras (31) interprets a decrease of body mass and an increase of U_{sg} as being due to dehydration. The decrease in body mass is often attributed to dehydration (23, 30, 31) and solely fluid loss (49). However, contributions to decreases in body mass during endurance activity include substrate losses and a release of glycogen-bound water (20, 64, 71), sweat (9, 12, 18, 20, 57, 64), urine secretion (12, 18, 64) and solid mass losses (26, 34, 35, 39, 42, 43, 45, 47, 50, 55, 71).

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Table 1. Description of races: NR – number of race, TR – temperature range, AT – average temperature, AH – average relative humidity, Weather, P – precipitation

Nr	Type of race	TR (°C)	AT (°C)	AH (%)	Weather	P (mm)
1	24-h MTB race 2012	6 – 30	18 (6)	43 (1)	Sun	—
2	24-h MTB race 2012	6 – 23	15 (4)	72 (2)	Clouds	3 (2)
3	24-h running race 2012	10 – 18	12 (3)	62 (3)	Rain	15 (5)
4	MTB multi-stage race 2012	22 – 33	26 (7)	55 (9)	Sun	—
5	24-h MTB race 2013	8 – 30	17 (4)	44 (2)	Sun	—
6	MTB multi-stage race 2013	12 – 26	22 (2)	46 (9)	Sun	—
7	100 km running race 2013	-1 – +1	0 (0)	65 (4)	Rain	25 (3)

On the contrary, fluid overload (7, 16, 56, 57, 63, 64, 70) and/or hormonal regulation by arginine vasopressin (17, 43, 56, 63, 64, 69, 72) may account for decreases in plasma [Na^+]. Paradoxically, decreases in body mass and dehydration (20-22, 26, 34, 50, 57, 63, 64) and a significant relationship between post-race plasma [Na^+] and losses in body mass (20, 26) have also been shown to exist in exercise-associated hyponatremia (EAH), a condition more commonly asociated with over-hydration. Nevertheless, EAH leads most frequently to an increase in body mass (2, 7, 16, 18, 34, 56, 57, 60, 63, 64), where the body mass gain is thought to be the principal cause of reduced plasma [Na^+] (2, 7, 10, 18, 22, 43, 57, 63). Body mass losses have been associated with asymptomatic hypovolemic EAH (18). Hypovolemic EAH would be then predicted by urinary [Na^+] (29) in conjunction with plasma or serum [Na^+] (18).

Recent studies investigated possible differences in the prevalence of EAH between different types of races and endurance disciplines. The incidence of EAH in endurance runners ranges from 0 to 51.2% (1, 19, 20, 22, 33, 37, 50, 51, 60, 67), while in endurance cyclists from 0 to 39% (8, 38, 62, 66). A comprehensive study about the prevalence of EAH in Switzerland in a group of male ultra-endurance runners and cyclists reported 6% of hyponatremic finishers (34); another study investigating cyclists and runners in the 161-km Iditasport ultramarathon in Alaska reported a value of 44% (71).

A previous comprehensive study investigating athletes competing in the Czech Republic showed an occurence of EAH of 5.7% in ultra-runners and ultra-mountain bikers (26). The present study aimed to expand the data from 2012 and included the data from races held in 2013. The inclusion of number of subjects competing in 2013 increased the overall number of athletes to reach new results about the incidence of EAH in athletes competing in the Czech Republic. We intended (*i*) to examine pre- and post-race body mass, U_{sg}, plasma and urinary [Na^+] and their changes and mutually associations and determined the occurence of EAH. We compared (*ii*) a hyponatremic (plasma [Na^+] < 135 mM) (56) and a normonatremic (plasma [Na^+] ≥ 135 mM) (56) group of athletes to find potential differences between them. We hypothesized that the incidence of EAH in the Czech Republic would be similar to the recent result (26). Even though the EAH largely developes during, immediately or up to 24 h following exercise (18), we hypothesized to find any differences between pre- and post-race hydration status in hyponatremic and normonatremic ultra-athletes to establish how those factors related to changes in plasma [Na^+].

Materials and Methods

The data were collected from seven races held in the Czech Republic (24-27) and included data on athletes competing during the years 2012 and 2013 (24-27). The study comprised runners and mountain bikers from various ultra-endurance disciplines. Multi-stage mountain bikers were included despite it not being a single-stage event. Athletes were contacted prior to their races and they gave their informed written consent *via* an e-mail. Research within the project proceeded in accordance with the law (No. 96/2001 Coll. M. S. on Human Rights and Biomedicine and Act No. 101/2000 Coll. Privacy) and the study was approved by the local institutional ethics committee of Institute of Experimental Biology at Masaryk University, Brno, Czech Republic.

The Races

Temperature and relative humidity on race day (s) for the various events are presented in Table 1. The 'Czech Championship 24-h MTB race' took place during the second weekend in June 2012 and 2013 with start at noon on Saturday and finished at 12:00 on Sunday (24-27). The course comprised a 9.5 km single-track with an elevation of 220 m. The 'Bike Race Marathon Rohozec 24 h' took place on June 9th 2012 and finished on June 10th 2012 (24, 26, 27). The course comprised a 12.6 km track with an elevation of 250 m. The 'Sri Chinmoy Self-Transcendence Marathon 24-h race' took place from July 21st 2012 to July 22nd 2012 (25, 26). The lap was 1 km, situated around an athletic stadium on asphalt with 1 m ascendent. The 'Trilogy Mountain Bike Stage Race' took place the first week in July in 2012 and 2013 (25, 26). The prologue covered 3 km with 300 m difference in elevation, the first stage covered 66 km with 2,200 m of altitude to climb, the second stage was 63 km in length with 2,300 m difference in elevation and the third stage was 78.8 km with 3,593 m change in altitude. The 'Czech Championship 100-km running race' was held March 9th 2013 (25). The ultra-runners had to run 66 laps on a 1,500 m circuit.

Procedures

Body mass was recorded prior to the start of the race and immediately after crossing the finish line. When a competitor participated in more than one race or for two consecutive years, data was eliminated and no competitor was measured twice. During the multi-stage race, the measuremens were taken prior the start and after the last stage of the race. All participants were measured using a calibrated commercial scale (Tanita BC-351, Tanita Corporation of America, Inc.) to the nearest 0.1 kg. Subjects were barefoot and generally clothed in running or cycling attire for both the pre- and post-race measurements. Blood samples were drawn from an antecubital vein. One Sarstedt S-Monovette (plasma gel, 7.5 ml) for chemical analysis was cooled and sent to the laboratory and were analysed within 6 h. Blood samples were obtained to determine pre- and post-race plasma $[Na^+]$ using biochemical analyzer Modul SWA, Modul P + ISE (Hitachi High Technologies Corporation, Japan, Roche Diagnostic). Urinary samples were collected in one Sarstedt monovett for urine (10 ml) and sent to the laboratory. Urine sampling was performed as close to the start and the end of the races and was finished within two h after the 24-h races, when all finishers had completed the race and some of them were finally able to hand in urinary samples due to problems with antidiuresis. U_{sg} was detemined using an Au Max-4030 (Arkray Factory, Inc., Japan), and urinary $[Na^+]$ using biochemical analyzer Modula SWA, Modul P + ISE (Hitachi High Technologies Corporation, Japan, Roche Diagnostic). The athletes drank pre- and during each race without any intervention.

Statistical Analysis

Descriptive statistics (mean, standard deviation) were calculated for pre-race, post-race and percentage changes of body mass, plasma and urinary $[Na^+]$, U_{sg} and fluid intake during the race. Shapiro-Wilk and Kolmogorov-Smirnov test were applied to prove the data normality. When data were not normally distributed, non-parametric tests were used. The Wilcoxon signed- rank test was used to analyse differences between values obtained prior and after the race. The Mann-Whitney test was used for between-group comparisons of continuous data. The Spearman rank correlation coefficient was conducted to examine the associations between selected variables. The Scheffe and Tukey *post hoc* tests were applied to compare pairs of race types. The level of statistical significance was set at $P < 0.05$.

Table 2. Weight, blood and urine characteristics of the hyponatremic and normonatremic group (n = 113)

	Units	Normonatremic finishers (n = 100)	Hyponatremic finishers (n = 13)
Age	yrs.	38.6 (8.5)	39.5 (6.9)
Male sex	%	71	69
Body mass pre-race	kg	72.1 (9.7)	73.4 (11.5)
Body mass post-race	kg	70.8 (9.6)	72.0 (11.7)
Body mass percentage change	%	-1.8 (1.8)**	-1.9 (1.7)**
Blood $[Na^+]$ pre-race	mM	139.7 (2.8)	139.0 (2.0)
Blood $[Na^+]$ post-race	mM	138.6 (2.3)	133.3 (1.4)$^{\#}$
Blood $[Na^+]$ percentage change	%	-0.8 (1.7)**	-4.1 (1.4)$^{**,\#}$
Urinary $[Na^+]$ pre-race	mM	96.8 (54.5)	81.9 (64.4)
Urinary $[Na^+]$ post-race	mM	69.9 (49.1)	63.2 (53.0)
Urinary $[Na^+]$ percentage change	%	-3.9 (94.9)**	-15.9 (55.3)
U_{sg} pre-race	g/ml	1.017 (0.006)	1.015 (0.007)
U_{sg} post-race	g/ml	1.025 (0.006)	1.028 (0.001)
U_{sg} percentage change	%	0.7 (0.7)**	1.3 (0.6)$^{**,\#}$
Fluid intake during the race	l/h	0.555 (0.3)	0.681 (0.3)

Weight, blood and urine characteristics of the hyponatremic and normonatremic group (n = 113). Data are reported as mean (SD). Significance was set at a significance level of $P < 0.05$. ** = $P < 0.001$ * = significant difference (post-race minus pre-race) within the group; $^{\#}$ = $P < 0.05$, $^{\#}$ = significant different between the EAH and the non-EAH group.

Results

Of 145 ultra-athletes participating in this study, 113 (81.8%) volunteers (88 men and 25 women), a total of twelve 24-h ultra-runners, fifty 24-h ultra-mountain bikers, thirty-two stage mountain bikers and nineteen 100-km ultra-runners underwent pre- and post-race measurements of body mass, provided pre- and post-race blood and urine samples and post-race reports about fluid intake during the race.

Plasma $[Na^+]$, the Incidence of EAH and Fluid Intake

Pre-race plasma $[Na^+]$ was not significantly different between hyponatremic (EAH) and normonatremic (non-EAH) ultra-athletes ($P > 0.05$) (Table 2).

Table 3. Blood, urinary and weight parameters in subjects with EAH (n = 13)

Nr	Sex	PreR B [Na^+]	PostR B [Na^+]	Change in B [Na^+]	PreR U [Na^+]	PostR U [Na^+]	Change in U [Na^+]	PreR U_{sg}	PostR U_{sg}	Change in U_{sg}	PreR BM	PostR BM	Change in BM
1	M	138	129	-6.5	24	50	201.6	1.015	1.025	1.0	90.0	88.2	-2.0
2	F	137	133	-2.9	54	68	25.9	1.010	1.025	1.5	54.6	53.2	-2.6
3	M	142	134	-5.6	42	12	-71.4	1.007	1.028	2.1	73.9	71.7	-3.0
4	M	138	134	-3.1	91	44	-51.6	1.016	1.030	1.4	89.6	87.5	-2.3
5	M	138	134	-2.9	104	65	-37.5	1.016	1.030	1.4	68.6	68.0	-0.9
6	M	136	132	-2.9	145	84	-42.1	1.030	1.030	0.0	82.0	82.1	0.1
7	F	139	134	-3.6	37	55	48.6	1.007	1.027	2.0	64.3	62.3	-2.0
8	F	137	134	-2.2	58	76	31.0	1.013	1.027	1.4	58.8	57.3	-2.6
9	M	138	134	-2.9	72	19	-73.6	1.017	1.027	1.0	67.9	65.9	-2.9
10	M	142	134	-5.0	250	197	-21.2	1.028	1.030	0.2	83.4	80.5	-3.5
11	M	142	134	-5.6	133	130	-2.3	1.020	1.030	1.0	72.0	71.7	-0.4
12	F	141	134	-5.0	32	11	-65.6	1.012	1.030	1.8	65.7	62.7	-4.6
13	M	141	133	-5.7	23	10	-56.5	1.007	1.030	2.3	84.2	85.8	1.9

Note: PreR – pre-race, PostR – post-race, [Na^+] – plasma sodium, U_{sg} – urinary specific gravity, B – blood, U – urinary, BM – body mass. Units: body mass = kg, [Na^+] = mM, U_{sg} = g/ml, change = %.

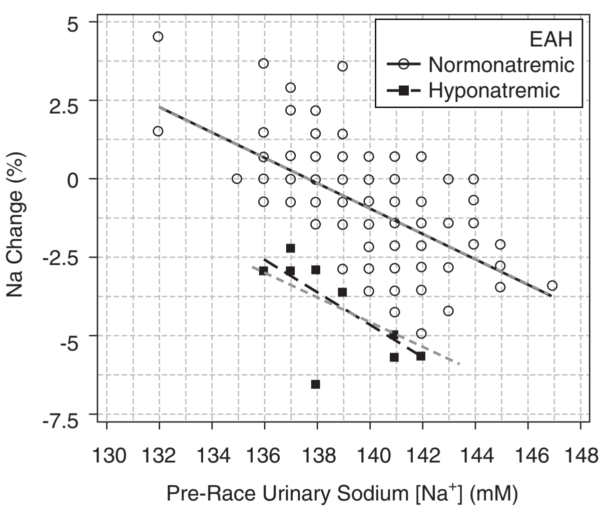

Fig. 1. The relationship of pre-race plasma [Na^+] and percentage change in plasma [Na^+] (%). EAH: n = 13, r = -0.73, P = 0.004; non EAH: n = 100, r = -0.60, P < 0.001.

The six athletes presented as being hypernatremic with plasma [Na^+] ≥ 145 mM had pre-race U_{sg} values ranging from 1009 to 1027 g/mL. On average, post-race plasma [Na^+] significantly decreased within the EAH and the non-EAH groups (P < 0.001) (Table 2) (25). In total, 13 of the 113 investigated athletes developed post-race EAH, equal to 11.5% (25) (Table 3). Between the EAH and the non-EAH groups we found differences in post-race plasma [Na^+] and percentage change in plasma [Na^+] (P < 0.001) (Table 2). Pre-race plasma [Na^+] in both EAH (r = -0.73, P = 0.004) and non-EAH (r = -0.60, P < 0.001) athletes was inversely associated with post-race percentage change in plasma [Na^+] (Fig. 1). Pre-race plasma [Na^+] was correlated with post-race plasma [Na^+] in non-EAH athletes (r = 0.57, P < 0.001). The fluid range varied from 0.4 to 1.5 L/h in EAH and from 0.2 till 1.5 L/h in non-EAH athletes (Table 2). Changes in plasma [Na^+] were not associated with reported fluid intake during the race, body weight change or change in U_{sg} in both groups (P > 0.05).

Comparing the different types of the races, statistically significant differences were found among pre-race (P < 0.001), post-race (P < 0.001) and absolute change of plasma [Na^+] (P = 0.04) (Table 4). The *post-hoc* tests did not show any of the pairs of race types to be significantly different comparing the mean value of changes in plasma [Na^+]. The comparison of plasma [Na^+] between EAH and non-EAH racers in each race was not possible due to a low number of EAH subjects (Table 4).

Body Mass and Post-Race Changes

On average, body mass significantly decreased within both groups (P < 0.001). Individual body mass changes at the finish varied from -6.6% to +2.4%. There were no significant group differences in pre- and post-race body mass, or percentage change in body mass (Table 2). We found associations between estimated fluid intake during the race and post-race percentage change in body mass in neither EAH nor non-EAH athletes (P > 0.05) (Table 2). We used cut-off points for hydration state based upon changes in body mass established by Noakes *et al*. (56, 57) where ≥ 0 change in body mass is overhydration, < 0 to -3% change in body mass is euhydration, and < -3%

Table 4. Body mass, plasma [Na^+] and U_{sg} in hyponatremic (n = 13) and normonatremic (n = 100) cases in each race

	24 MTB race		24 RUN race		100-km RUN race		MTB stage race	
	EAH (n = 6)	Non-EAH (n = 44)	EAH (n = 1)	Non-EAH (n = 11)	EAH (n = 4)	Non-EAH (n = 15)	EAH (n = 2)	Non-EAH (n = 30)
Body mass								
Pre-race	75.5 (13.4)	74.5 (10.3)	54.6	67.3 (8.4)	72.2 (7.8)	62.9 (8.2)	79.0 (7.2)	74.9 (7.2)
Post-race	74.2 (13.4)	72.8 (10.2)	53.2	66.5 (8.5)	70.2 (7.8)	61.2 (7.2)	78.7 (9.9)	73.8 (7.2)
Absolute change	-1.3 (0.8)	-1.6 (1.4)	-1.4	-0.8 (1.0)	-2.0 (1.2)	-1.6 (1.2)	-0.3 (2.6)	-1.0 (1.3)
Percentage change (%)	-1.7 (1.1)	-2.2 (1.8)	-2.5	-1.2 (1.6)	-2.8 (1.7)	-2.4 (1.7)	-0.5 (3.4)	-1.4 (1.7)
Plasma [Na^+]								
Pre-race	137.6 (1.0)	137.8 (1.8)	137.0	140.2 (1.4)	140.5 (1.7)	138.9 (2.2)	141.5 (0.7)	142.7 (2.0)
Post-race	132.8 (2.0)	137.4 (1.9)	133.0	139.1 (1.8)	134.0 (0.0)	138.2 (2.3)	133.5 (0.7)	140.3 (2.0)
Absolute change	-4.8 (2.1)	-0.4 (2.4)	-4.0	-1.0 (2.5)	-6.5 (1.7)	-0.7 (2.5)	-8.0 (0.0)	-2.4 (1.8)
Percentage change (%)	-3.5 (1.5)	-0.3 (1.7)	-2.9	-0.7 (1.8)	-4.6 (1.1)	-0.5 (1.8)	-5.6 (0.0)	-1.6 (1.3)
U_{sg}								
Pre-race	1.016 (0.000)	1.017 (0.006)	1.010	1.013 (0.004)	1.019 (0.006)	1.018 (0.008)	1.007 (0.000)	1.018 (0.007)
Post-race	1.028 (0.002)	1.024 (0.005)	1.025	1.018 (0.010)	1.029 (0.001)	1.028 (0.002)	1.029 (0.001)	1.026 (0.006)
Absolute change	0.012 (0.006)	0.006 (0.006)	1.015	0.005 (0.011)	0.010 (0.006)	0.010 (0.008)	0.022 (0.001)	0.008 (0.007)
Percentage change (%)	1.184 (0.663)	0.665 (0.593)	1.485	0.513 (1.149)	0.984 (0.646)	1.027 (0.846)	2.184 (0.140)	0.841 (0.776)

Data are reported as mean (SD) except where indicated to be a percentage. Units: body mass = kg, plasma [Na^+] = mM, U_{sg} (urinary specific gravity) = g/ml. MTB = mountain biking, RUN = running.

Fig. 2. The distribution of athletes into the three groups based on hydration status (n = 113).

change in body mass is dehydration. Fig. 2 shows the detailed distribution of ultra-athletes into the four groups on the basis of plasma [Na^+] and three groups based on hydration status. Two (15.4%) EAH finishers were dehydrated, two (15.4%) were overhydrated and nine (69.2%) were euhydrated post-race according to this classification in body mass change (Fig. 2). Twenty-seven (27%) non-EAH finishers were classified as dehydrated, thirteen (13%) as overhydrated and fifty-four (54%) as euhydrated at the finish (Fig. 2).

Statistically significant differences were found among pre- and post-race body mass ($P < 0.001$) in each of different types of race. Absolute and percentage body mass changes during the race were homogenous among the races (Table 4). Body mass comparisons between EAH and non-EAH racers in each race were not possible due to the low number of EAH subjects (Table 4).

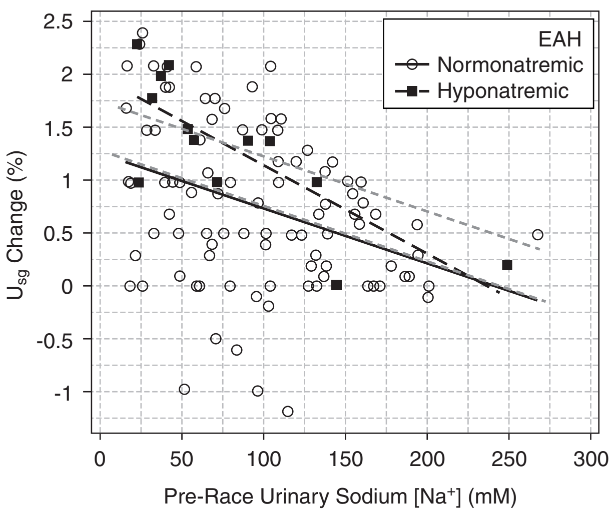

Fig. 3. The relationship of pre-race urinary [Na^+] and percentage change in U_{sg} (urinary specific gravity) (%). EAH: n = 13, r = -0.78, P = 0.002; non-EAH: n = 100, r = -0.37, P < 0.001.

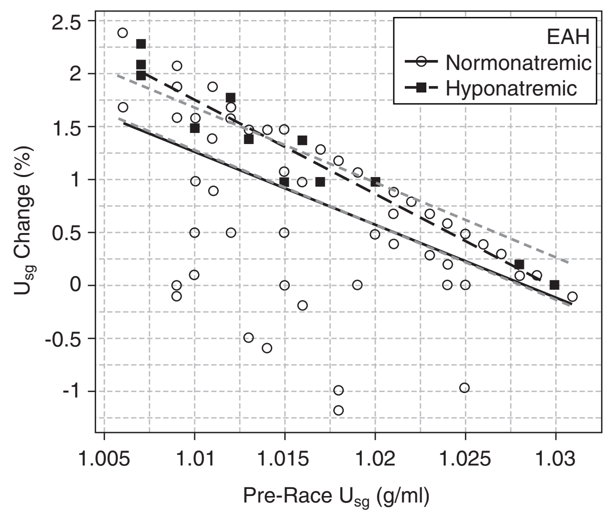

Fig. 4. The relationship of pre-race urinary specific gravity and percentage change in U_{sg} (urinary specific gravity) (%). EAH: n = 13, r = -0.96, P < 0.001; non-EAH: n = 100, r = -0.60, P = 0.001.

Pre-Race Urinary [Na^+] and U_{sg} and Post-Race Changes

Pre- and post-race urinary [Na^+] and the decrease in urinary [Na^+] did not differ between EAH and non-EAH athletes (P > 0.05) (Table 2). Nevertheless, urinary [Na^+] decreased significantly in non-EAH finishers (P < 0.001) and non-significantly in EAH finishers (P = 0.064) by Wilcoxon test, however on the level of significance (P = 0.049) by paired t-test. Pre-race urinary [Na^+] was negatively associated with percentage change in U_{sg} in non-EAH (r = -0.37, P < 0.001) and EAH (r = -0.78, P = 0.002) finishers (Fig. 3) and positively with post-race urinary [Na^+] in non-EAH (r = 0.40, P < 0.001) and EAH (r = 0.88, P < 0.001) finishers. Post-race urinary [Na^+] was negatively related to percentage change in U_{sg} in EAH (r = -0.70, P = 0.008) and positively with post-race plasma [Na^+] in non-EAH (r = 0.50, P < 0.001) ultra-athletes. We found no relationship between post-race urinary [Na^+] and percentage change in body weight or reported fluid intake during the race (P > 0.05). Individual level of urinary [Na^+] concentration below 30 mM (29) showed 9 (9%) of non-EAH athletes and 2 (15.4%) of EAH athletes pre-race and 2 (2%), and 4 (30.8%) post-race, respectively.

Pre-race U_{sg} values varied from 1.006 to 1.031 g/ml (n = 113) without significant differences between the EAH and the non-EAH groups (P > 0.05). Pre-race U_{sg} was inversely associated with post-race percentage change in U_{sg} in both post-race EAH (r = -0.96, P < 0.001) and non-EAH (r = -0.60, P = 0.001) athletes (Fig. 4). Following Armstrong *et al*. (5), a normally hydrated man exhibits the initial morning U_{sg} < 1024 g/ml; U_{sg} < 1010 g/ml is observed only after a fluid overload and U_{sg} < 1017 g/ml corresponds to extremely hyperhydrated. Fifty-four (54%) non-EAH ultra-athletes had U_{sg} values < 1017 g/ml, 14 (14%) in the range 1017-1021 g/ml, seven (7%) between 1022-1023 g/ml, ten (10%) between 1024-1026 g/ml, six (6%) between 1027-1028 /ml, eight (8%) between 1029-1031 g/ml and no athlete > 1031 g/ml prior to the race. Nine (69.2%) EAH ultra-athletes had pre-race values < 1017 g/ml, two (15.4%) in the range between 1017-1021 g/ml, none in the range between 1022-1026 g/ml, one (7.7%) had 1028 g/ml and one 1030 g/ml.

Post-race U_{sg} values varied from 1.008 to 1.031 g/mL (n = 113). On average, post-race U_{sg} increased within the EAH and the non-EAH groups (P < 0.001) (Table 2). We found significant differences in percentage change in U_{sg} (P < 0.01); however post-race levels in U_{sg} were equal (P > 0.05) in both group. Post-race U_{sg} was inversely related to change in body mass (r = -0.22, P = 0.02) in the non-EAH group. Percentage change in U_{sg} associated with percentage change in body mass in none group of ultra-athletes (P > 0.05). There was no relationship between estimated fluid intake and percentage change in U_{sg} or pre- or post-race U_{sg} within the EAH and the non-EAH groups (P > 0.05). Based on urinary indices during dehydration, exercise and rehydration (6) individual post-event U_{sg} samples are considered in the normal range between 1013 and 1029 g/ml, 1030 or higher as significant dehydration and below 1012 g/ml as hyperhydration. Seven (7%) non-EAH ultra-athletes had U_{sg} values below 1012 g/ml, 47 (47%) had U_{sg} values in the range between 1013-1029 g/ml and 46 (46%) values of 1030 g/ml or higher after the race.

None of EAH finishers developed values below 1012 g/ml, six (46.2%) had U_{sg} values in the range between 1013-1029 g/ml and 7 (53.8%) values of 1030 g/ml or higher. Regarding U_{sg}, the *post-hoc* test did not show any of the pairs of race types to be significantly different comparing the mean value of U_{sg} or urinary [Na^+] (Table 4).

Discussion

Pre- Race Hydration Status by Plasma and Urinary [Na^+] and U_{sg} Concentrations

On average, pre-race plasma [Na^+] as well as pre-race U_{sg} levels did not differ significantly between the group that developed and the group that did not develop post-race EAH. Two athletes started their races with plasma [Na^+] indicating EAH (132 mM); however, they developed no EAH at the finish. On the contrary, of the six ultra-athletes who were hypernatremic prior to the race with plasma [Na^+] ≥ 145 mM, none developed EAH and none were hypernatremic post-race. Hoffman *et al.* (20) observed at the finish of a 161-km running race in total nearly 2% of hypernatremic cases, 50% were overhydrated and nearly 17% were dehydrated. Nevertheless, Noakes *et al.* (57) described 13% post-race hypernatremic cases with 4% overhydration and 59% dehydration.

Hypernatremia can be the result of pure sodium excess, usually associated with dehydration (23, 56) and secondary due to excess losses of water or hypotonic fluids (12). However, overhydration and hypernatremia together occur only through excessive sodium intake and/or mobilization of internal sodium stores together with fluid overconsumption (20). The present athletes did not report their sodium intake; however, is not unusual to observe these sorts of practices in ultra-races (19, 63). Regarding pre-race individual U_{sg} levels following Armstrong *et al.* (5), two athletes of the present six hypernatremic cases seemed extremely hyperhydrated. For a healthy man, it is rare to achieve U_{sg} below 1010 g/mL in the morning sample (5) and one present case developed even U_{sg} of 1009 g/ml. One racer was slightly hyperhydrated, two athletes were well hydrated and only one was slightly dehydrated. Therefore, it does not seem that the present hypernatremic cases were dehydrated; it may be that hypernatremia observed was attributed to excessive pre-race sodium intake.

The interesting fact was that 54% of the current athletes who did not develop post-race EAH were considered to be extremely hyperhydrated and 14% slightly hyperhydrated compared to nearly 70% of extremely and 16% of slightly hyperhydrated post-race hyponatremic racers by categories of hydration status regarding their pre-race U_{sg} levels (5). Conversely, only 6% of the normonatremic and 8% of the hyponatremic racers were classified as being slightly dehydrated and 8% were very dehydrated (5). Moreover, a pre-race urinary [Na^+] concentration < 30 mM in 15% of the post-race hyponatremic and 9% of the post-race normonatremic athletes indicated that some athletes started the race with diluted sodium levels. Besides, lower pre-race urinary [Na^+] seemed to be a risk factor for lower post-race urinary [Na^+] in both groups, and for lower plasma [Na^+] in normonatremic finishers. We have to take into account that urinary indices are suggested as parameters of pre- and post-race hydration status (1, 2, 4, 11, 31, 36, 53, 64, 68) and it has been shown to accurately classify individuals as either hyper-, eu- or dehydrated (59).

The goal of prehydrating is to start physical activity euhydrated, with normal body electrolyte status and with urine output at normal levels (2, 11). The safest hydration strategy, following the Statement of the Third International EAH Consensus Development Conference in 2015 is to drink when thirsty prior, during and after the exercise (18). In addition, being hyperhydrated prior to exercise may enhance thermoregulatory function while exercising in the heat (61). Therefore, it is alarming that nearly 86% of the present post-race hyponatremic and 68% of post-race normonatremic athletes probably drank prior the races greater volumes than their thirst dictated, they followed a strategy to begin the race 'well hydrated' and did not drank *ad libitum*.

The next interesting finding was that lower pre-race U_{sg} and urinary [Na^+] concentrations resulted in higher increases in post-race U_{sg} levels in both normonatremic and hyponatremic finishers with even highly significant relationship in both correlations within the hyponatremic group. Moreover, lower post-race urinary [Na^+] concentrations associated with higher U_{sg} increases in hyponatremic finishers. It is difficult to explain these contradictory findings. Nevertheless, it seems that 'overhydration' prior to the race did not protect these athletes from an increase in post-race U_{sg} concentrations.

Post-Race Hydration Status by Plasma and Urinary [Na^+], U_{sg} and Body Mass Changes

On average, post-race plasma [Na^+] significantly decreased in both groups. Above that, lower plasma [Na^+] pre-race appeared to be a risk factor for lower plasma [Na^+] post-race in normonatremic finishers, which is in agreement with findings reported by Chorley *et al.* investigating non-elite marathon runners (28). The change in plasma [Na^+] was affected by pre-race plasma [Na^+] in both groups as in the mentioned study (28); however, the changes were affected inversely, not directly. Herein, intra-race hydration behaviours and/or individual physiological responses may account for the fact that post-race plasma [Na^+] did not fall under the limit level for the development of EAH in normonatremic finishers.

However, we found no differences in estimated fluid consumption during the race between both groups. Moreover, reported fluid intakes related neither to changes in body mass in agreement with Black *et al.* (8) nor to plasma [Na^+] similarly as in Knechtle *et al.* (38) or in Black *et al.* (8) nor to U_{sg} change as in Mahon *et al.* (53). Nevertheless, it also must be noted that the estimated fluid intake reported could have been inaccurate due to an error from the subjects self-reporting an average consumption. Probably, the syndrome of inappropriate antidiuretic hormone (SIADH) would cause fluid retention (17) and ADH may also be partially responsible for the fact that the majority of the present athletes had lower post-race [Na^+].

Due to different race conditions, more important than comparing fluid intake seems monitoring of the hydration status. On average, post-race U_{sg} concentration significantly increased in all ultra-athletes. Based on urinary indices during dehydration, exercise and rehydration (6), 46% of normonatremic and nearly 47% of hyponatremic ultra-athletes were considered significantly dehydrated. On the contrary, only 7% of normonatremic and even no hyponatremic finishers showed hyperhydration. The current evidence tends to favour urine indices as the most promising available marker (68) in comparison with hematological parameters which are not as sensitive (11). Nevertheless, the increase may have occured due to muscle catabolism and elevated plasma urea (41) and/or protein digestion (15), therefore the use of post-race U_{sg} concentration is limited (2, 5, 6, 13). In addition, it should be noted that it is time-dependent and shows only chronic dehydration, but not acute dehydration (2).

On average, body mass decreased significantly in hyponatremic and normonatremic finishers without differences between both groups. Nevertheless, post-race hydration status was similar only in the percentage number of overhydrated finishers as defined by individual body mass changes according to Noakes *et al.* (57). The third of normonatremic finishers was dehydrated and it was nearly twice more than in the hyponatremic athletes. It is in contrast with the hydration status regarding post-race U_{sg} values, where the number of dehydrated finishers was similar in both groups and represented approximately half of the racers. Mahon *et al.* (53) in their recent study about hydration status in mountain marathon events stated that percentage body mass loss must be considered with caution as athletes are not weighed naked and as body mass loss was not associated with U_{sg} values. It is very likely that there could be an error associated with the percentage change in body mass due to substrate oxidation (32). Body mass losses were slightly inversely related to post-race U_{sg} in the present normonatremic group; however, they were neither related to changes in U_{sg} nor to post-race plasma [Na^+] nor to changes in plasma [Na^+] in both groups, similarly as in Knechtle *et al.* (38) or Black *et al.* (8). The decrease in body mass and the increase in U_{sg} indicated dehydration (31, 68), rather than hyperhydration. On the contrary, body mass changes alone do not reflect a change in body hydration; rather a combined effect of fluid and food intake and energy losses during the race (65) and they are not a reliable measure of hydration status (56).

Nevertheless, a body mass equal to or above the normal body mass is a positive indicator for the presence of fluid overload (18, 68). Due to the wide variation in hydration levels in the present athletes, similarly as in Mahon *et al.* (53), it is likely that athletes would benefit from own individualized hydration strategies. Moreover, human water regulation is complex and dynamic, so it is difficult to assign a numerical value of euhydration, dehydration or hyperhydration (3, 63). Presumably in normonatremic ultra-athletes *ad libitum* intake, an increased activity of vasopressin and/or mobilization of sodium from internal stores (57) maintained their fluid homeostasis, despite of plasma [Na^+] losses. This should be determined in future studies.

The Incidence of EAH

The 11.5% of investigated ultra-athletes developed post-race mild EAH without any clinical implications in all present hyponatremic cases. The important fact is that the incidence of EAH in athletes competing in races held in the Czech Republic was higher than in our previous study (26). Eighty-five percent of the present hyponatremic cases were volume depleted due to body weight changes. Weight loss in hyponatremic cases may suggest volume depletion as a contributor to EAH (18). Moreover, the average increase in U_{sg} in the EAH group was significantly higher than in the non-EAH group. Hoffman *et al*. (20) demonstrated in the 5-year EAH research at a 161-km ultramarathon a relationship between post-race plasma [Na^+] and percentage change in body mass such that a lower plasma [Na^+] was more common with an increased weight loss. Also the runners developing EAH which lost even greater body mass during the race than the non-hyponatremic runners were presented by Hoffman *et al*. (19).

However, post-race plasma [Na^+] was not related to percentage change in body mass or fluid intake in the present study, similarly as in studies from Knechtle *et al*. investigating 24-h ultra-runners (41) or ultra-endurance mountain bikers (38). Notwithstanding, a lower incidence of EAH with overhydration seems to support a depletional model of EAH, when not only sodium loss, but also impairment in mobilization of osmotically inactive sodium and/or in innapropriate inactivation of osmotically active sodium are alternative explanations (18, 20). On average, urinary [Na^+] decreased in both groups; however, the number of finishers with urinary [Na^+] below 30 mM increased only in the EAH group. Hypovolemic EAH was supported by urinary [Na^+] < 30 mM (29) in conjunction with plasma [Na^+] < 135 mM in 31% of the present hyponatremic finishers (Table 2). One hyponatremic case (number 13 in Table 3) showed weight gain, in contrast with a decrease of body mass in the other finishers with urinary [Na^+] < 30 mM. The increased urinary [Na^+] losses probably expressed a reaction to a stimulation of the renin-angiotensin-aldosterone system, as reported in Knechtle *et al*. (40).

The incidence of EAH with body mass losses in the present study could influence a greater sodium loss through sweat under ambient temperature conditions in certain on the included races and the longer duration of the races, similarly as in Hoffman *et al*. (20). According to Hew *et al.* (18) we have evidence of hypovolemic EAH during the considered ultra-endurance races although the relative contribution of sweat and urine sodium losses is negligible with possible exception of volume depleted athletes with a serum or plasma [Na^+] < 135 mM. Nevertheless, the reasons remain unclear in the absence of clear evidence for overdrinking and fluid retention in all cases due to the wide variation in individual fluid consumption and hydration levels. There is paucity of data supporting sodium loss as the primary mechanism of symptomatic EAH (18) and relative over-drinking with sustained non-osmotic AVP secretion (16, 18), failure to use inactive sodium stores or osmotic inactivation of serum sodium (60) were likely involved in the development of EAH in present cases.

Limitations

A limitation was the rather small number of finishers with EAH and the low number of competitors in ultra-running races in comparison with mountain bike races. However, we have chosen the biggest, the most popular and therefore the most representative ultra-races held in the Czech Republic in the years 2012 and 2013. A further limitation was the limited number of subjects per race. It was impossible to have a high number of subjects due to the time constraints associated with the duration of pre- and post-race measurements. A further limitation is that we did not determine the sodium content in the ingested food and fluid, we did not analysed sweat sodium concentration and we did not record urine production.

Conclusions

Thirteen (11.5%) finishers developed post-race hyponatremia (plasma [Na^+] < 135 mM). The incidence of EAH in athletes competing in ultra-endurance races held in the Czech Republic was higher than previously reported. It seems that 'overhydration' prior the race did not protect ultra-athletes from an increase in post-race U_{sg} concentrations. Moreover, the present findings indicate that the determination of plasma [Na^+] remains the only viable possibility for determining EAH in ultra-endurance athletes.

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