Urinary Indices During Dehydration, Exercise, and Rehydration

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This investigation evaluated the validity and sensitivity of urine color (Uocol), specific gravity (Usg), and osmolality (Uosm) as indices of hydration status, by comparing them to changes in body water. Nine highly trained males underwent a 42-hr protocol involving dehydration to 3.7% of body mass (Day 1, -2.64 kg), cycling to exhaustion (Day 2, -5.2% of body mass, -3.68 kg), and oral rehydration for 21 hr. The ranges of mean (across time) blood and urine values were Uocol, 1–7; Usg, 1.004–1.029; Uosm, 117–1,081 mOsm · kg⁻¹; and plasma osmolality (Posm), 280–298 mOsm · kg⁻¹. Urine color tracked changes in body water as effectively as (or better than) Uosm, Usg, urine volume, Posm, plasma sodium, and plasma total protein. We concluded that (a) Uocol, Uosm, and Usg are valid indices of hydration status, and (b) marked dehydration, exercise, and rehydration had little effect on the validity and sensitivity of these indices.

Key Words: urine color, osmolality, specific gravity, plasma, sodium, sweat

Thermoregulatory sweating during strenuous exercise typically results in a body water loss of 0.8–2.0 L/hr (2). Because the thirst drive does not stimulate drinking until water loss reaches 1–2% of body mass (19), it is likely that athletes routinely train and compete in an unrecognized state of hypohydration. When water loss exceeds 2–3% of body mass, an athlete’s exercise performance, heat dissipation, and cardiovascular function are compromised (20). This phenomenon is not limited to sports that involve weight classes or hot environments, as witnessed by our observations of chronic dehydration in male and female collegiate swimmers (consuming less than 0.24 L/day of fluid) who trained for several hours each day in cool water (C.M. Maresh et al., unpublished observations, 1994).

Many authorities have advised athletes to evaluate their own fluid balance by observing sweat loss, fluid intake, and/or body mass. However, these techniques are not accurate unless all food, beverages, sweat, respiratory water loss, and excreta are carefully weighed and accurately interpreted. Other authorities (24, 26) have recommended urinalysis as a screening tool because it involves a noninvasive evaluation of a body fluid, but laboratory urinalysis is rarely performed because instruments require technical expertise and are costly. In an attempt to simplify direct

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body fluid analysis, nutritionists and exercise physiologists have advised athletes to observe their urine color \((U_{\text{col}})\) as an index of hydration status (1) because urine specific gravity \((U_{\text{sg}})\) and \(U_{\text{col}}\) are strongly correlated (2, 13). Many athletes follow this recommendation but do not realize that \(U_{\text{col}}\) may be altered by illness, vitamin supplements, exercise, and medications (10, 16, 18). This prompted our previous investigation of urinary indices of hydration status (2), which assessed the validity of using \(U_{\text{col}}\) in field and laboratory settings. Observations of 54 male and female subjects in three separate studies led to the following conclusions: (a) \(U_{\text{col}}\) was strongly correlated with \(U_{\text{sg}}\) \((r = .80, p < .0001)\) and urine osmolality \((U_{\text{osm}}; r = .82, p < .0001)\) when measured with a novel eight-color scale, and (b) \(U_{\text{col}}\) may be used by athletes as a meaningful index of hydration status when measurements of \(U_{\text{sg}}\) and \(U_{\text{osm}}\) are impractical. However, subsequent unpublished field observations of Olympic athletes (17) suggested that \(U_{\text{col}}, U_{\text{sg}}, \) and \(U_{\text{osm}}\) may be less accurate during periods of large water turnover (i.e., prolonged summer training or competition). Physiologically, this may occur because circulating hormones and the kidneys require several hours to restore fluid–electrolyte balance after severe body water perturbations (22). Therefore, the purpose of this controlled investigation (42 hr) was to evaluate the validity and sensitivity of \(U_{\text{col}}, U_{\text{sg}}, \) and \(U_{\text{osm}}\) as indices of hydration status during marked dehydration (−4% of body mass), a strenuous exercise trial in a hot environment, and a 21-hr period of oral rehydration.

**Methods**

Prior to testing, the following procedures and experimental protocol were approved by the local Institutional Review Board for Human Research. All participants received a verbal and written explanation of procedures, equipment, instruments, risks, and benefits and gave written voluntary consent to participate. The 9 male test subjects exhibited the following personal characteristics (mean ± SD): age, 23 ± 3 years; height, 180 ± 4 cm; mass, 69.8 ± 3.0 kg; body fat, 13.9 ± 2.1%; peak oxygen consumption \((\dot{V}O_2\text{peak})\), 60.3 ± 3.9 \(\text{ml} \cdot \text{kg}^{-1} \cdot \text{min}^{-1}\). Medical history questionnaires indicated that no subject had previously experienced heat illness, impaired thermoregulation, drug/alcohol abuse, or cardiovascular, respiratory, metabolic, renal, or endocrine disorders. These 9 males were trained cyclists who competed in road or off-road races and used no tobacco products.

Hydrostatic weighing was used to determine body density, corrected for residual lung volume. Percent body fat from body density was computed with the equation of Brozek et al. (5). \(\dot{V}O_2\text{peak}\) was determined in a 27 °C environment, via an incremental exercise test on a cycle ergometer (Monark, Model 818E, Stockholm, Sweden). The attainment of \(\dot{V}O_2\text{peak}\) was identified when subjects reached the following two criteria: (a) less than 150 \(\text{ml} \cdot \text{min}^{-1}\) increase in oxygen consumption following a 0.5 kp increase in ergometer resistance, and (b) a respiratory quotient greater than 1.10. The measurements of \(\dot{V}O_2\text{peak}\) and body fat were taken at least 5 days prior to the testing described below.

**Experimental Protocol**

Each subject completed one 42-hr period of observations that involved normal hydration, exercise-induced dehydration, a strenuous exercise bout in a hot environment, and a lengthy rehydration period. During the 24 hr prior to dehydration,
subjects were instructed to consume water or other noncaffeinated fluids (a volume of 30 ml · kg⁻¹ body mass) to ensure proper hydration. Also, because subjects trained regularly, they were instructed to limit their workouts to moderate to low intensities during the 48 hr prior to testing. Subjects reported to the laboratory on Day 1 (10 a.m. to noon) in a euhydrated state, at least 14 hr after eating their last meal. The hydration state of each participant was quantified by measuring $U_{50}$ body mass, and plasma osmolality; these measurements were collectively identified as the baseline phase (B) below.

In the subsequent dehydration phase of this study (D) subjects performed 2 hr of moderate-intensity cycling exercise, wearing multiple clothing layers to enhance sweating, and restricted fluid intake for $21 \pm 2$ hr. The goal of D was to decrease body mass by 4%. After a dehydration level of −4% had been verified on Day 2 (7 to 11 a.m.), subjects entered an environmental chamber ($T_{amb}$ of $36.7 \pm 0.2$ °C) and performed an exercise bout to volitional exhaustion (intensity of 70% $\dot{V}O_2$peak, $18.9 \pm 2.7$ min duration) on a cycle ergometer while consuming no fluids. Immediately following this exercise bout ($17.9 \pm 7.9$ min duration), body fluid samples were collected and relevant measurements were recorded; these values are identified as Phase E in the text below. Subjects then resumed their normal drinking and eating habits and began the rehydration phase of this protocol, during which they drank a variety of fluids ad libitum. Subjects recorded food and fluid items in a dietary log. Body mass was measured and fluids were sampled during two return visits to the laboratory, at $4 \pm 1$ hr postexercise (4H; afternoon of Day 2) and $21 \pm 1$ hr postexercise (21H; morning of Day 3). The amount of time required to complete this entire protocol (Phases B, D, E, 4H, and 21H) was $42 \pm 1$ hr (group mean ± SD), spanning three mornings (Days 1–3).

The following instruments were used during all phases of this investigation. $U_{50}$ and total plasma protein ($P_{TP}$) were evaluated in duplicate with an optical refractometer (Spartan, Model A300CL, Japan). Body mass was recorded on a platform scale (±50 g; SR Instruments, Model 700M, Tonawanda, NY). $U_{osm}$ and plasma osmolality ($P_{osm}$) were measured via the freezing point depression technique (Advanced Instruments, Model 3DII, Norwood, MA), using standard solutions appropriate for each fluid. Plasma sodium ($P_{Na}$) concentration was assessed with ion-specific electrodes (AVL Scientific Corp., Model 984-S, Roswell, GA), using appropriate standard and control solutions for each fluid. All urine and blood measurements were made in triplicate, unless otherwise noted.

Blood samples were collected at Phases 4H and 21H with a sterile needle and syringe from an antecubital arm vein without stasis. At Phases B, D, and E, a 20-gauge Teflon cannula (Critikon, Tampa, FL) was placed in a superficial forearm vein. The cannula was kept patent between D and E with a 1.5-ml volume of isotonic heparinized saline solution. Prior to all blood collections, except at E (i.e., immediately postexercise), subjects sat quietly for 20 min to allow plasma volume to stabilize.

Each urine specimen was collected in an inert polypropylene container. All $U_{col}$ and $U_{50}$ measurements were performed within 20 min of collection and were verified by two of three designated investigators. $U_{col}$ was assessed by holding each specimen container next to a color scale, in a well-lighted room. This urine color scale was developed in our laboratory and was based on observations of specimens collected during previous field (L.E. Armstrong, unpublished observations, 1988) and laboratory (2) investigations. The eight-color scale included colors ranging
from very pale yellow (#1) to brownish green (#8). Because colors are difficult to standardize, the colors in our urine scale were compared to the classic compendium of colors published by Maerz and Paul (14) and matched the following standardized samples (plate/grid number): Color 1, 17/B1; Color 2, 9/H1; Color 3, 17/J1; Color 4, 17/L1; Color 5, 9/I3; Color 6, 9/L3; Color 7, 12/K6; Color 8, 23/L1.

Statistical Analyses

The outcome variables were evaluated for significance across time (i.e., from Phase B to Phase 21H) via a one-way analysis of variance with repeated measures. Group data were expressed as means ± SD. Significance for statistical tests was established at the .05 level of confidence. The Newman-Keuls post hoc test was used to determine specific differences among the sample means in the event of a significant F ratio. Pearson product moment correlation coefficients were computed for selected variables across all time points. These r values were transformed to z scores; the mean z score was then calculated and transformed again to a value that represented the mean correlation coefficient for all time points (S. Owen, University of Connecticut, personal communication).

Results

All baseline (B) urine and plasma measurements (Table 1, Figures 1 and 2) indicated that subjects complied with pretest drinking instructions and were well hydrated at the beginning of testing. The 21-hr period of dehydration and fluid restriction (D) resulted in a mean body mass loss of 2.64 kg; expressed as a percentage of the initial body mass, this corresponded to a loss of 3.7 ± 0.9%. Subsequent exercise in a hot environment resulted in an additional body mass loss of 1.04 kg (−5.2% total).

Table 1  Body Mass and Urine Volume (mean ± SD) During All Phases of the Protocol

<table>
<thead>
<tr>
<th>Measurement (units)</th>
<th>Protocol phasea</th>
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<tbody>
<tr>
<td></td>
<td>B</td>
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<tr>
<td>--------------------------</td>
<td>-----</td>
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<tr>
<td>Body mass (kg)</td>
<td>70.77b</td>
</tr>
<tr>
<td>±3.89</td>
<td>±3.78</td>
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<tr>
<td>Body mass change (%)b</td>
<td>0.0b</td>
</tr>
<tr>
<td>±0.0</td>
<td>±0.9</td>
</tr>
<tr>
<td>Urine volume (L)c</td>
<td>0.87</td>
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<tr>
<td>±1.10</td>
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aB = baseline; D = dehydration; E = immediate postexercise; 4H = 4 hr postexercise; 21H = 21 hr postexercise. bSignificantly different from all other means (p < .05 to .001). cSignificantly different from all other means (p < .05 to .005), except 4H versus 21H. dChange relative to Phase B. eDurations (hr) of urine volume collections were B, 17; E, 0.6; 4H, 4; 21H, 21. fNot measured; only a single specimen was collected.
The fluids consumed during phases 4H and 21H were selected by personal preference and involved a variety of beverages (i.e., water, milk, sport drinks, soda, juices); the group mean was 4 ± 3 different beverages. The mean volume of fluid consumed was 5.56 ± 1.80 L per 21 hr of rehydration. This mean 5.56 L fluid intake closely approximates the sum of the mean maximal body weight loss (Phase E, 3.68 kg, Table 1) plus the total 21-hr urine volume during rehydration (2.02 L, Table 1). Thus, after Phase E, ad libitum drinking resulted in nearly complete restoration of body mass at 21H (restored to -0.8% of initial body mass; -0.6 kg).

Figures 1 and 2 depict changes in body mass, urine, and hematological variables during this investigation. Figure 3 illustrates the relationship between the three primary outcome variables (i.e., $U_{col}$, $U_{ur}$, and $U_{om}$) during all phases of testing (B through 21H); the dashed line represents a theoretical linear relationship.
Figure 2 — Mean (±SD) changes in plasma indices of hydration status during the 42-hr protocol.

Discussion

Maintenance of fluid—electrolyte balance by the kidneys is accomplished by simultaneous volume regulation, osmoregulation, and circadian rhythms (6). **Volume regulation** (i.e., circulatory homeostasis) is associated with water and sodium balance and may be altered by diet, posture, exercise, and the size of the extracellular space (21). **Osmoregulation** is affected by changes in water and electrolyte excretion, under the influence of arginine vasopressin. **Circadian rhythms** of electrolyte excretion may be altered by environmental cues (e.g., ratio of light or dark exposure) but are independent of posture, exercise, environmental temperature, and water or salt deprivation (23). Although controlling urine tonicity by the renal tubules is the primary means by which the body regulates fluid—electrolyte balance, no universally accepted method exists to determine hydration status (7) from measurements of renal concentration (i.e., $U_{\text{osm}}$, $U_{\text{sg}}$).

Our previous research (2) showed that $U_{\text{osm}}$ and $U_{\text{sg}}$ were strongly correlated with $U_{\text{col}}$ ($r = .80$ and .82, respectively), and that $U_{\text{col}}$ may be used by athletes as a meaningful index of hydration status. However, subsequent observations of British
Figure 3 — Specific relationships between $U_{col}$, $U_{osm}$, and $U_{sg}$ during all experimental phases. The dashed line represents unity.

athletes competing at the 1996 Summer Olympic Games in Atlanta (17) found that $U_{col}$, $U_{osm}$, and $U_{sg}$ were less accurate during periods of heavy training and large water turnover. This important theory could not be challenged by our previous research, which included resting measurements, a maximal fluid loss of only 1.8% of initial body mass, and no direct observations of rehydration (2). However, this theory was addressed by the present experimental design, which simulated the intense training and competition of highly trained cyclists.

**Urinary and Plasma Indices**

The change in body mass was used as the “gold standard” by which all hydration indices were evaluated, because it represented body water fluctuations. Body mass loss, as depicted in Figure 1, revealed that changes in $U_{col}$, $U_{osm}$, and $U_{sg}$ followed a
pattern similar to that of fluid loss. Minor exceptions to this pattern in $U_{\text{osm}}$ and $U_{\text{s}}$ at Phase D (i.e., they rose higher than in Phase E) were observed. This may have occurred because of the differential hormonal control of water (i.e., via arginine vasopressin) versus solutes (i.e., sodium via aldosterone). By definition, $U_{\text{osm}}$ and $U_{\text{s}}$ are determined by solute particle concentrations. Urine color, in contrast, is influenced by changes in both solutes and water. Overall, $U_{\text{osm}}$ tracked body water loss as effectively as, or more effectively than, $U_{\text{osm}}$ and $U_{\text{s}}$ (Figure 1). Further, Figure 2 illustrates that changes in plasma variables approximated the changes in body mass loss (Figure 1).

Clinical and lay publications often note that $U_{\text{osm}}$ changes in response to illness, medications, and vitamin supplementation (10, 13, 18). Even exotic urine colors (i.e., pink, red, green, blue, black) may result from physiological pigments in abnormal concentrations, food derivatives, or bacterial growth in stored specimens (8). The influences of these factors on $U_{\text{s}}$ in the present study were not controlled, but their effects appear to have been minimal because (a) no exotic colors were observed, (b) the correlations between $U_{\text{col}}$, $U_{\text{osm}}$, and $U_{\text{s}}$ were strong (range: $r = .68$ to $.99$), and (c) the time between specimen collection and urine color determination was brief (20 min). Previously, we had frequently noticed that the properties of specimens changed when they stood at $T_{\text{amb}}$ of 21–23 °C, or were refrigerated, for several hours. Thus, practitioners and athletes should measure urinary indices soon after samples are collected because bacterial contamination, standing, or refrigeration result in increased turbidity, precipitation, darkened $U_{\text{col}}$, and an offensive odor (4, 10).

Prior to all blood collections, except at E (i.e., immediately postexercise), subjects sat quietly for 20 min to allow plasma volume to stabilize. Because the blood samples at E were collected immediately postexercise, it is possible that plasma constituents were altered by exercise-induced fluid shifts. This is a possible limitation in the design of the present investigation and in the interpretation of plasma values at Phase E (Figure 2). Although plasma total protein would be the most likely variable to reflect exercise-induced plasma volume changes (i.e., plasma proteins do not leave the circulation readily), previous research indicates that plasma volume shifts of up to −10% do not alter $P_{\text{N}}$ or $P_{\text{osm}}$ values (11, 25).

**Effects of Dehydration, Exercise, and Rehydration**

Figure 3 (bottom panel) illustrates that all mean (±SD) values for $U_{\text{sg}}$ and $U_{\text{osm}}$ fell near or on the line of unity, indicating that experimental manipulations (i.e., dehydration, exercise, rehydration) had little effect on the relationship between $U_{\text{sg}}$ and $U_{\text{osm}}$. Indeed, their correlation coefficient across time points was $r = .99$ ($p < .001$, $n = 9$ at each point), despite the fact that $U_{\text{osm}}$ depends on the number of particles in solution and $U_{\text{sg}}$ depends on the number and mass of the solutes (10). Previously (2), we reported that $U_{\text{osm}}$ and $U_{\text{col}}$ could be used interchangeably ($r = .97, p < .001, n = 45$) in research settings to assess euvolemia versus hypohydration but that the variance of this relationship increases at $U_{\text{sg}} > 1.024$ and $U_{\text{osm}} > 900$ mOsm · kg⁻¹. Both of these conclusions are supported by this graph.

The two uppermost panels in Figure 3 illustrate the relationships of $U_{\text{col}}$ versus $U_{\text{osm}}$ and $U_{\text{sg}}$. During dynamic periods of great or rapid fluid loss or gain (i.e., D, E, 21H), the strength of the relationship between $U_{\text{col}}$ and $U_{\text{sg}}$ or $U_{\text{osm}}$ diminished only slightly. However, the mean correlations of these urinary indices across time remained strong ($r = .68$ and .72). We previously reported that these correlation coefficients
were $r = .82$ (U_{col} vs. U_{osm}) and $r = .80$ (U_{col} vs. U_{eg}) when data from three substudies were combined (2).

The standard deviation bars in Figures 1 and 3 show that the greatest variance in U_{col}, U_{sp}, and U_{osm} occurred during rehydration phases 4H and 21H. Based on the research of Shirreffs et al. (22), this variance can be attributed to differences in the volume of fluid consumed by these highly trained cyclists during ad libitum rehydration. Had this experimental protocol involved controlled fluid intake (i.e., 20 ml·h^{-1}·kg body mass^{-1}) of only one fluid, it is likely that the variance in urinary indices at 4H and 21H would have been smaller. Such a protocol, however, would not have simulated athletes' actual drinking practices. The reader is referred to a recent investigation (3) for consideration of physiological mechanisms that affect drinking behavior. In addition, differences in the composition of rehydration fluids may alter either U_{col} or U_{osm}. For example, Gonzalez-Alonso et al. (9) and Maughan et al. (15) reported that fluids containing sodium chloride or potassium chloride decreased U_{col} and increased U_{osm} versus pure water (they did not report U_{col} and U_{eg}). However, in the present study, drink composition apparently had a minor effect on urinary indices, because although subjects drinking ad libitum consumed a variety of fluids, the relationships between both U_{col} and U_{eg} ($r = .68$) and between U_{col} and U_{osm} ($r = .72$) were strong.

It has been noted that the composition of individual urine specimens is not likely to be identical to that of a composite urine sample collected over many hours (12). This is especially relevant to athletes because a dehydrated athlete can rehydrate with a large bolus of pure water or hypotonic fluid that results in one or two urine specimens with a low U_{col}, U_{osm}, or U_{eg}, before the body water deficit has been replaced completely. This occurs because the ingested fluid rapidly dilutes the blood, and the kidneys, in turn, temporarily excrète a dilute urine. Theoretically, such an individual specimen might falsely indicate that an athlete has rehydrated successfully. In contrast, when pure water or other fluids are consumed gradually, the extracellular fluid equilibrates with intracellular fluid gradually, and differences between single specimens and composite samples are smaller. Because composite urine samples were not analyzed in the present investigation, a separate pilot study was conducted of five healthy males (unpublished observations, 1997). Statistical analyses verified that single specimens were different from composite samples (collected during 5 ± 3 hr). Correlations (of single urine specimens vs. composite samples) were as follows: $U_{col}, r = .81, p < .001, n = 53, range 1–6$; and $U_{eg}, r = .64, p < .001, n = 53, range 1.008–1.028$; U_{osm} was not measured. This pilot study and Figure 1 suggest that, after athletes rapidly ingest a large quantity of pure water or hypotonic fluid without food, collection of three or more consecutive individual specimens (which exhibit low values for U_{col}, U_{osm}, and U_{eg}) will verify adequate whole-body rehydration.

**Practical Applications**

The foregoing observations of highly trained cyclists indicate that urinary indices of hydration may be used in the following ways:

1. During daily training sessions and postexercise recovery or rehydration periods, athletes may utilize U_{col} as an index of hydration status that tracks body water changes as well as or better than U_{osm}, U_{eg}, P_{osm}, P_{Nat}, and P_{tp} (Figures 1 and 2).
2. Exercise, large gains and losses of body water, and intake of a variety of rehydration fluids apparently do not alter the validity of \( U_{\text{col}} \), \( U_{\text{sg}} \), or \( U_{\text{osm}} \) as indices of hydration status. Throughout this investigation, \( U_{\text{col}} \) was strongly correlated with both \( U_{\text{osm}} \) and \( U_{\text{sg}} \) (Figure 3).

3. The strength of the relationship between \( U_{\text{osm}} \) and \( U_{\text{sg}} \) \((r = 0.98, p < .001)\) indicates that they may be used interchangeably in laboratory studies to assess hydration status during dynamic periods of large water turnover.

4. If athletes achieve a \( U_{\text{col}} \) of 1 or 2 (i.e., "very pale yellow" or "pale yellow"), their body mass will be within 1% of their well-hydrated baseline body mass (Figure 1). It is likely that this state will minimize the detrimental effects of dehydration on exercise performance.

5. When dehydrated athletes consume a large bolus of pure water or hypotonic fluid rapidly (i.e., >1.5 L·hr\(^{-1}\)), they should collect three or more consecutive specimens to verify that whole-body rehydration has been accomplished.

References


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