Comparison of Common Field/Clinical Measures to Standard Laboratory Measures of Hydration Status

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Context: Accurately determining hydration status is a preventative measure for exertional heat illnesses (EHI). **Objective:** To determine the validity of various field measures of urine specific gravity (U_{sg}) compared to laboratory instruments. **Design:** Observational research design to compare measures of hydration status: urine reagent strips (URS) and a urine color (U_{col}) chart to a refractometer. Setting: We utilized the athletic training room of a Division I-A collegiate American football team. Participants: Trial 1 involved urine samples of 69 veteran football players (age=20.1+1.2yr; body mass=229.7+44.4lb; height=72.2+2.1in). Trial 2 involved samples from 5 football players (age=20.4+0.5yr; body mass=261.4+39.2lb; height=72.3+2.3in). Interventions: We administered the Heat Illness Index Score (HIIS) Risk Assessment, to identify athletes at-risk for EHI (Trial 1). For individuals "at-risk" (Trial 2), we collected urine samples before and after 15 days of pre-season "two-a-day" practices in a hot, humid environment(mean on-field WBGT=28.84+2.36°C). Main Outcome Measures: Urine samples were immediately analyzed for U_{sg} using a refractometer, Diascreen 7[®] (URS1), Multistix[®] (URS2), and Chemstrip10[®] (URS3). U_{col} was measured using U_{col} chart. We calculated descriptive statistics for all main measures; Pearson correlations to assess relationships between the refractometer, each URS, and U_{col}, and transformed U_{col} data to Z-scores for comparison to the refractometer. **Results:** In Trial 1, we found a moderate relationship (r=0.491, $p \le .01$) between URS1 (1.020 $\pm 0.006 \mu g$) and the refractometer $(1.026+0.010\mu g)$. In Trial 2, we found marked relationships for U_{col} (5.6+1.6 shades, r=0.619, *p*<0.01), URS2 (1.019±0.008µg, r=0.712, *p*≤0.01), and URS3 (1.022±0.007µg, r=0.689, p<0.01) compared to the refractometer (1.028+0.008µg). Conclusions: Our findings suggest that URS were inconsistent between manufacturers, suggesting practitioners use the clinical refractometer to accurately determine U_{sg} and monitor hydration status. **Key Words:** urine color, urine reagent strip, urine specific gravity

Both the National Athletic Trainers' Association and American College of Sports Medicine have released position statements regarding hydration in athletes and the risks associated with hypohydration.^{1,2} Hypohydration and heat related injuries may result in loss of playing time, decrease in performance, decrease in overall health, and death.³ The National Center for Catastrophic Sports Injuries reports since 2000 there have been 13 fatalities due to heat stroke, and in 2005 alone 5 heat related fatalities.⁴ Because hypohydration is one of the leading signs of exertional heat illness,¹ allied health professionals treating high risk populations should be measuring hydration status as a preventative priority.

Change in body mass, urine color (U_{col}) , urine osmolality (U_{osm}) , plasma osmolality (P_{osm}) , and urine specific gravity (U_{sg}) are all common measures of hydration status, and each method presents advantages and limitations. Based upon the methods available, clinicians should develop assessment protocols to assess pre-participation hydration levels and subsequently monitor high risk athletes over the course of activity.³

Some measures of hydration status, osmolality, are more appropriate for a laboratory setting, yet others, change in body mass and urine color, are more commonly associated with

clinical measures of hydration. Researchers often look for tools that provide the greatest accuracy and reliability for laboratory investigations.⁵ However, in a clinical setting, tools that require little technician expertise and expense are more customary. The "gold standard" for the measurement of hydration status in the clinical setting is unclear. Several researchers state that the use of a clinical refractometer for U_{sg} is more accurate than urine reagent strips (URS),⁶⁻¹⁰ while others state that URS are equally reliable.¹¹⁻¹³ As research continues to examine hydration indices to determine the most reliable and valid measurement of hydration status, the purpose of this research was to determine the validity of URS compared to refractometers in detecting accurate measures of U_{sg} in the clinical setting.

Methods

Participants

For Trial 1, 69 Division I-A collegiate American football players (age= 20.1 ± 1.2 yr; body mass= 229.7 ± 44.4 lb; height= 72.2 ± 2.1 in) participated in the pilot implementation of the Heat Illness Index Score(HIIS) Risk Assessment to identify athletes at-risk for exertional heat illness. Five football players (age= 20.4 ± 0.5 yr; body mass= 261.4 ± 39.2 lb; height= 72.3 ± 2.3 in) were identified as moderate-risk, and urine samples and body mass changes were continuously monitored throughout preseason "two-a-day" practices (Trial 2) in a hot, humid environment (mean on-field WBGT= $28.84\pm2.36^{\circ}$ C).

Research Design

We used a non-experimental, observational research design to compare three brands of URS and U_{col} to a refractometer.

Experimental Procedures

As part of the HIIS, participants were asked to provide a urine sample (Trial 1). The sample was immediately analyzed for U_{sg} using a clinical refractometer (model 300CL, Atago Inc., Japan) and the Diascreen 7® (Hypogaurd, Minneapolis, MN) reagent strip (URS1). Urine samples were then collected before and after each practice for fifteen days of practices (Trial 2). U_{sg} was measured using the clinical refractometer, Multistix® (Bayer Corporation, Elkhart, IN) reagent strip (URS2), and Chemstrip 10® (Roche Diagnostics, Indianapolis, IN) reagent strip (URS3). Urine color was measured using a U_{col} chart (Human Kinetics, Champaign, IL). U_{sg} and U_{col} were immediately measured with the refractometer and U_{col} chart, respectively. Then, two researchers analyzed the specimens with the URS. One researcher read the strip in the specimen, as recommended by each manufacturer and the other researcher read the results at the times specified by the manufacturer.

Statistical Analysis

Descriptive statistics were calculated for all urine indices. Pearson correlations were calculated to assess the strength of the relationship between the clinical refractometer and the URS. Subsequently, z-scores were calculated for the urine color and the clinical refractometer measures of U_{sg} . Pearson correlations were then calculated to identify the strength of the relationship between these variables. Significance was set *α-priori* at *p*<0.05.

Results

Measures of urine indices demonstrated that our participants were significantly hypohydrated (Table 1). A moderate correlation was found for the relationship between URS1 and the clinical refractometer (Table 2). Marked correlations were found for the relationship between URS2 and URS3 (Table 2). The correlation analysis of z-scores revealed the clinical refractometer measure of U_{sg} and urine color were moderately correlated (r=0.619, *p*<0.01).

Discussion

Urine Specific Gravity

The NCAA suggests U_{sg} as the most practical, cost-efficient measurement of hydration status in athletes.¹⁴ U_{sg} is a measure of the ratio between the density of urine and the density of water.^{3,5,6,15} Urinary concentration depends on the presence of particles (electrolytes, phosphate, urea, uric acid, proteins, glucose, and radiographic contrast media) per unit of urine volume.^{3,7} Small amounts of urine can be used for rapid, non-invasive, and inexpensive measurements.^{5,7} Fluid denser than water will have a measurement greater than 1.000Gm/mL.⁵ In the average healthy person U_{sg} ranges between 1.002 to 1.030,¹⁵ with minimal hypohydration measure set at 1.010 to 1.020 (1% to 3% loss of body weight)^{1,16} and severe hypohydration above 1.030.^{1,5,17} Two popular U_{sg} measurement techniques are URS and refractometry.

URS are cost-efficient, easily accessible, and easy to use^{7,10} to estimate U_{sg} .³ When immersed in urine, an analytic reaction occurs in the small pads between an ion exchanger, bromthymol, and buffers with the urine concentration of hydrogen ions and sodium ions. The protons are released in the presence of cations and react with the bromthymol blue, changing the color of the strip.^{3,6,7,10}

Urine refractometry identifies when concentrated fluid breaks normal light differently than water and measuring the refraction of the beam,^{3,7} detecting particles according to weight rather than number.⁷ Research has demonstrated that U_{sg} measurement by refractometry is a more sensitive indicator of hydration status than blood plasma or hematocrit measurements¹⁸⁻²⁰ and should be used when determining hydration status of athletes.¹

Refractometry is the preferred method of U_{sg} measurement as compared to URS.^{9,10} However, previous literature is contradictory concerning the use of URS. Some studies have shown a positive correlation between the two types of measurements, while others have shown no correlation. Correlation between URS and refractometry has been found to be low (r=.573).¹⁵ Refractometry is consistently more accurate and reagent strip results are often unpredictable.⁶ We also ascertained that URS were unpredictable between manufacturer types. Further research is necessary to determine the validity of URS as a measure for U_{sg} .⁵ Our research confirmed inconsistent results for U_{sg} within different URS tested and the overall moderate correlations leads us to suggest that refractometry continues to be the preferred clinical method of U_{sg} measurement. However, URS are a psychomotor skill required in educational competencies and therefore, a common clinical practice taught in athletic training education programs.²¹ *Change in Body Mass*

Water comprises 50-70% of the body's total mass.³ Change in body mass is the most common clinical technique used to determine hydration status in athletes and utilizes pre and post-exercise body mass measurements to calculate the amount of body water lost. Although the most commonly used, change in body mass has limitations. Immediate ingestion of fluid equal to 5% body loss did not return P_{osm} to baseline measurements.⁹ Fluids ingested following 6% body loss required 48-72 hours to demonstrate a euhydrated status.²² These results show that while change in body mass is an inexpensive, practical, method for hydration measurement, it may not be valid or reliable.

Urine Color

 U_{col} is an inexpensive, yet reliable⁵ method for hydration measurement. Normal urine color should be described as light yellow^{3,5} with severe hypohydration described as brownish green.⁵ U_{col} does not provide the accuracy or precision of U_{sg} or U_{osm}^{5} and has a tendency to underestimate the level of hydration,¹⁷ but it may be a valid self-assessment of hydration level.

However, U_{col} can be misleading if a large amount of fluid is consumed rapidly, causing the kidneys to excrete dilute urine even if hypohydration exists.⁵ *Urine Osmolality*

 U_{osm} and U_{sg} are accurate measures of hydration status.¹⁹ U_{osm} measures the amount of osmoles of dissociated solute particle per kilogram of solution.^{3,6} U_{osm} measurements require a osmometer and trained technician, therefore may not be practical for clinical use.⁵ Normal U_{osm} equals <500mOsm/L.³ While osmolality is the most accurate measurement of total solute concentration, U_{osm} may not accurately determine hydration status immediately after activity. Water turnover, intercultural differences, regulative mechanisms all contribute to an inaccuracy of U_{osm} measurements.⁵

Plasma Osmolality

 P_{osm} is the most widely used hematological index of hydration and is considered the "gold standard" of hydration status.⁵ P_{osm} accurately tracks acute changes in hydration status during exercise in a warm environment. P_{osm} linearly increased from 283 mosmo/kg when euhydrated by > 30mosmol/kg after hypohydrated by 15% of total body water.²³ This shows that P_{osm} is positively correlated with hydration status. When hypohydrated, P_{osm} will proportionally decrease and increase when euhydrated. Osmolality increases because sweat is ordinarily hypotonic relative to plasma.²³ P_{osm} measurements increase with progressive hypohydration and return toward euhydration.^{9,16} As with U_{osm} , P_{osm} is measured using an osmometer, requiring expensive equipment and training, which again may not be clinically practical.⁵ Also, P_{osm} can be influenced by sports drinks or meals.⁹

Urine Reagent Strips

A variety of manufacturers produce reagent strips, each with specific instructions for proper techniques for immersion and reading of results. Failure to follow manufacturer's specifications is a common cause of inaccurate test results.^{10,28} Leaving the reagent strip immersed in the specimen too long may cause the reagents to dissolve and become inaccurate.¹⁰ Reagent strips must also be protected against ambient moisture, heat, and light, may not be stored in alternate containers, and will be inaccurate if expired.²⁸ In addition, specimen containers must be free of detergents and other contaminants.¹⁰

URS measurements were typically greater than refractometer measurements (mean difference of 0.002 ± 0.007) and although the URS were specific (83%) they were not sensitive (38%).¹⁵ One researcher reported strong correlations (r=.906 and r=.911) when comparing URS and 2 refractometers.¹¹ Several other researchers suggest that reagent strips are not a valid measure for U_{sg}, reporting correlations well below .800,²⁴⁻²⁹ which are not suitable for clinical practice. Further, a wide dispersion of data has been demonstrated when comparing reagent strips (Clinitek-50) with a refractometer.⁷ This research, among our moderate correlations, further support the use of refractometry for measuring U_{sg} instead of inconsistent and unreliable URS.

Conclusions

Hydration status is instrumental in preventing heat related illness. When choosing a measurement technique in the clinical setting it is important the clinician use a tool that is practical, inexpensive, and does not require technical operation, yet also provides the practitioner with a reliable and accurate measure. We suggest a urinary refractometer is a more reliable clinical measure of U_{sg} and should be used in conjunction with change in body mass and U_{col} to monitor the hydration status of at-risk athletes exercising in extreme environmental conditions.

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Table 1. Urine Indices

Trial 1 n=55	Mean	SD
USG Clinical Refractometer	1.026	0.010
RS1	1.020	0.006
Trial 2 n=5	Mean	SD
USG Clinical Refractometer	1.028	0.008
RS2	1.019	0.008
RS3	1.022	0.007
Urine Color	5.6	1.6

Table 2. Pearson r Correlations

	Trial 1 n= 69	Trial 2 n= 55		
USG Clinical	URS1	URS2	URS3	
Refractometer				
	r= 0.491 p< 0.01	r= 0.712 p< 0.01	r= 0.689 <i>p</i> < 0.01	