

University of Arkansas, Fayetteville
ScholarWorks@UARK

Theses and Dissertations

12-2015

The Validity of Urine Color Self-Assessment as an Index of Hydration in Males

Daniel Layne Nixon

University of Arkansas, Fayetteville

Follow this and additional works at: <http://scholarworks.uark.edu/etd>

 Part of the [Human and Clinical Nutrition Commons](#), and the [Other Kinesiology Commons](#)

Recommended Citation

Nixon, Daniel Layne, "The Validity of Urine Color Self-Assessment as an Index of Hydration in Males" (2015). *Theses and Dissertations*. 1406.

<http://scholarworks.uark.edu/etd/1406>

This Thesis is brought to you for free and open access by ScholarWorks@UARK. It has been accepted for inclusion in Theses and Dissertations by an authorized administrator of ScholarWorks@UARK. For more information, please contact scholar@uark.edu.

The Validity of Urine Color Self-Assessment as an Index of Hydration in Males

A thesis submitted in partial fulfillment
of the requirements for the degree of
Master of Science in Kinesiology

by

Daniel Layne Nixon
Bachelor of Science in Kinesiology, 2013

December 2015
University of Arkansas

This thesis is approved for recommendation to the Graduate Council.

Dr. Stavros Kavouras
Thesis Director

Dr. Matthew Ganio
Committee Member

Dr. Brendon McDermott
Committee Member

Abstract

While water intake is so important, there are few practical hydration assessment techniques for the general population to use on a daily basis. The present study examined the accuracy of self-assessed urine color (U_{col}) as a potential hydration assessment tool. Male college aged subjects provided a urine sample into a custom built urinal ($n=76$; 1.79 ± 0.76 m, 83.9 ± 16.0 kg). The urinal contained a picture of the 1-8 color scale and a light and dark urine color scale. Subjects were asked to give their urine color estimation as a whole number integer and to estimate if their urine was light or dark. For each sample, osmolality (U_{osmo}), specific gravity (U_{sg}) and urine color (U_{col}) were measured in the laboratory. Participant's U_{col} was determined from a researcher by comparing the color of the urine sample to the 1-8 color urine color scale and the light and dark scale. Based on the ROC analysis the overall accuracy of the self-assessment of U_{col} was calculated to be 65% (area under the curve). The analysis further resulted in 35% specificity and 91% sensitivity. On the light and dark scale only 8 participants choose dark while the other 68 chose light. Additionally, of the 68 people that chose light 18 were categorized as hypohydrated. Of the 8 participant's that chose dark, 4 were categorized to be euhydrated. Bland-Altman analyses were used to calculate the agreement between self- and laboratory-assessed U_{col} ratings ($r=0.31$; $P<.005$). The mean difference between self- and laboratory-assessed U_{col} was $-.26$ urine color units. This means it was found participants tended to rate their U_{col} slightly lighter than the researchers. Furthermore, it was found as U_{col} became increasing darker, so did the discrepancy between the participant and researchers U_{col} ratings. In conclusion, self-assessed U_{col} had "poor" capabilities to identify hypohydration overall. Also, self-assessed U_{col} had a reasonable ability to diagnose hypohydration with a U_{col} of 3 or greater. Based on these data, young adult males can moderately assess their U_{col} accurately.

Acknowledgements

Special thanks are extended to the faculty, staff, and students of the University of Arkansas Human Performance Laboratory and the College of Education and Health Professions for supporting this research.

Table of Contents

Table of Contents	
I. Introduction	1
II. Methods	3
Subjects	3
Procedures	3
Urine Analysis	4
Data Analysis	4
III. Results	5
Validity of the self-assessed urine color to measure urine concentration.....	6
Self-assessment of urine color.....	6
Agreement between self- and lab-assessed urine color.....	7
IV. Discussion	8
References	11
Appendix	13

I. Introduction/Review of Literature

Water has been called the most essential nutrient for the human body (Kavouras & Anastasiou, 2010; Manz, Wentz, & Sichert-Hellert, 2002). Sufficient water intake and hydration status is vital to exercise performance, cognition, and general health (Bar-David, Urkin, & Kozminsky, 2005; Kavouras et al., 2012; Thornton, 2010). Heat illness, death, and frequent heat injuries among athletes and the general population during summer months emphasize the need for proper hydration among individuals prone to high levels of hypohydration. While water intake is so important, there is no universally accepted measure of hydration status. There are multiple urinary measurements (i.e., osmolality, volume, urine specific gravity, and color) researchers can use to measure hydration status. Researchers consider these measurements to be time saving, relatively easy, and accurate but the average population does not have access to laboratory equipment. A more feasible approach for the mass population to assess their hydration status is urine color (U_{col}).

Normal U_{col} is primarily due to the presence of urochrome, a byproduct of hemoglobin breakdown (Ehrig, Waller, Misra, & Twardowski, 1999). Variations in U_{col} are mostly due to the differences in urine concentration, but may also be affected by changes in pH, ingested substances, and metabolic abnormalities (Raymond & Yarger, 1988). Researchers have recommended the general population to observe their U_{col} each day to assess their hydration status because studies have shown U_{col} is strongly correlated with urine osmolality ($r = 0.82$, $p < 0.0001$) and U_{sg} ($r = 0.80$, $p < 0.0001$) (Armstrong et al., 1998; Kavouras et al., 2015). The thirst drive has been found to not stimulate drinking until water loss reaches 1-2% of body mass (Adolph & Rothstein, 1947). This means individuals could, at times, be in a state of hypohydration without knowing it. With water needs for optimal hydration not being

consistently defined, it is necessary for the individual to use a hydration marker on a regular basis to know their hydration status. 24-h urine is a commonly used indicator of hydration status in laboratory settings (Morimoto et al., 2014; Guerra et al., 2014). Many researchers prefer it to individual urine samples because it accounts for the daily fluctuations in urine concentration due to eating, drinking, and exercise (Shephard, Penberthy, & Fraser, 1981). Unfortunately, using 24-h urine samples in real life situations is not realistic due to the need for laboratory equipment. It has been suggested that early afternoon urine concentration is closely correlated with that of total 24-h urine samples (Perrier et al., 2013). This suggests this period may be appropriate as an alternative sample to estimate 24-h hydration status.

To date no study has found if the general population can accurately assess their urine color compared to an experienced researcher in the lab. One justification of a possible discrepancy between self- and lab-assessed U_{col} is associated with the general perception of color. Roy and Colleagues (1991), suggested age plays a role in color perception, with individuals between the ages of 20 to 50 best able to accurately recognize differences in colors. Kavouras and Colleagues (2015), advised self-validated U_{col} only be used to distinguish between eu- and hypohydrated in the case of children. With U_{col} seeming to be the best option for real life situations no study has determined if the general population can accurately assess their hydration status from their urine color. The purpose of this study is to examine the accuracy of self-assessed urine color as an index of hydration in males. It is hypothesized self-assessed urine color is evaluated accurately compared to an evaluation of the urine sample by a researcher.

II. Methodology

Subjects

76 male subjects were recruited for this study from a convenient sample within the city of Fayetteville, Arkansas. The participants were between 18 and 36 years of age. Institutional review boards at the University of Arkansas approved this study. This study only included physically healthy participants free from any renal or physiological diseases. After being informed of the purpose of this study, all subjects gave written consent to participate.

Procedures

Subjects came in between the times of 7:00 AM and 3:00 PM to participate in the study. The participants started by being provided detailed verbal and written instructions of the procedures of the study. Then the participants completed the International Physical Activity Questionnaire (IPAQ) and Water Intake Questionnaire (WIQ). After the questionnaires, the participant's body weight and height without shoes and minimal clothing were recorded to the .1 kg and .01 m.

Next, subjects were provided with a classic 1-8 urine color scale (Armstrong et al., 1994) in order to evaluate the color that best describes their urine color. Subjects were informed on the correct use of the urine color scale and instructed to urinate into the urinal to assess their urine color. Numbers were assigned to the colors from 1, representing the lightest, to 8, representing the darkest. Also, subjects were provided with a light and dark scale with the light respecting a color of a euhydrated urine sample and a dark color representing a hypohydrated urine sample. Subjects were instructed to give a urine sample into a custom built urinal. The urinal contained a picture of the 1-8 color scale and the light and dark scale. The urinal drained into a dark plastic container on the bottom so the researcher could collect the sample, where the subjects would not

be able to see the color or the volume of their sample. Subjects were then asked to provide a sample and give their urine color estimation as a whole number integer and to estimate if their urine was light or dark. After the participants had given the researcher their self-validated estimates of the two charts the participant was asked if he observed the urine from the stream or urine pooled in the urinal. All subjects received financial compensation after the completion of their participation.

Urine Analysis

Urine samples were analyzed within 24-h after the collection. U_{osmo} was measured multiple times, by freezing point depression (3D3 Advanced Osmometer, Advanced Instruments, Inc., MA). U_{sg} was measured using a hand-held clinical refractometer (ATAGO SUR-NE, TOKYO, Japan). Each participant's U_{col} was determined from an experienced researcher by comparing the color of the urine sample placed in a clear, glass 15-mL tube against a white background, under fluorescent lighting next to the classic 1-8 color urine color scale and the light and dark scale.

Data Analysis

The association of the urine color scale in males as a marker of hydration status was tested by regression analysis, performed between the participant U_{col} vs. U_{osmo} . Threshold analysis was based on the receiver operation characteristic (ROC) curve to evaluate the optimal value of U_{col} to predict hypohydration (i.e., $U_{\text{osmo}} \geq 800 \text{ mmol kg}^{-1}$). The Bland-Altman comparison method evaluated the accuracy between the researcher- versus participant-evaluated U_{col} values. A probability (P) level of 0.05 was defined for statistical significance. Statistical analyses were performed using SPSS (version 22, SPSS Inc., Chicago, IL).

III. Results

Subject characteristics of the 76 participants are presented in Table 1, while mean values and ranges of measured hydration markers are presented in Table 2.

Table 1

Subject characteristics

	Mean \pm SD	Range
Sample size (#)	76	76
Height (m)	1.79 \pm 0.76	1.57-1.98
Weight (kg)	83.9 \pm 16.0	60.1-134.4
BMI (kg·m ⁻²)	26.2 \pm 4.8	18.8-38.4

Table 2

Urinary hydration markers

	Mean \pm SD	Range
U _{osmo}	661 \pm 247	49-1121
U _{sg}	1.017 \pm 0.007	1.001-1.032
Lab- U _{col}	3.4 \pm 1.3	1-6
Self- U _{col}	3.1 \pm 1.0	1-6
Stream (#)	27	
Urinal (#)	45	

Validity of the self-assessed urine color to measure urine concentration

Linear regression analysis revealed that self-assessed U_{col} was significantly positively associated with U_{osmo} (Fig. 1). U_{col} ratings explained 21% of the variance in U_{osmo} ($r = 0.46$, $P < 0.0001$).

This is a weak predictor of U_{osmo} in healthy men.

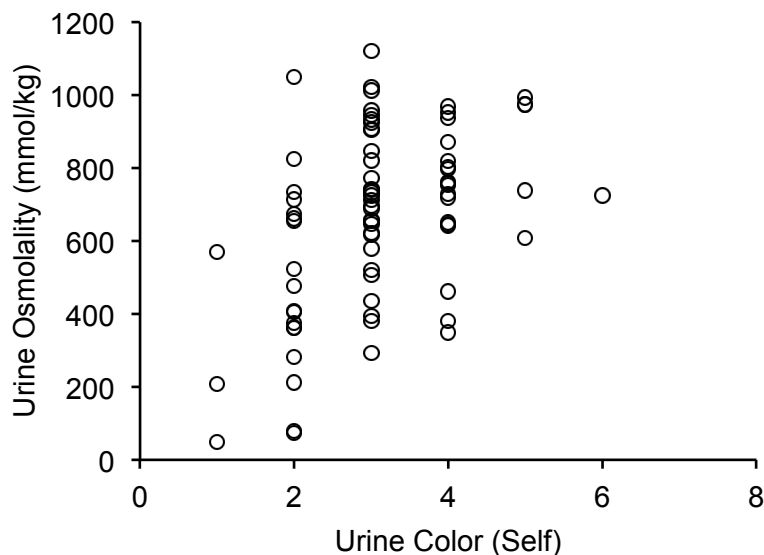


Fig. 1 Regression analysis of urine color as a predictor of urine osmolality.

Self-assessment of urine color

Based on the ROC analysis the overall accuracy of the self-assessment of U_{col} was calculated to be 65% (area under the curve). Self-assessment of U_{col} illustrated poor ability to identify hypohydrated samples (Table 3; $P < 0.005$). The term “poor” is a ranking used to define the ability of the test (i.e., urine color) to identify the condition (i.e., hypohydration). This is when the area under the curve (AUC) is between 0.60 and 0.69 (Tape, 2015). The analysis further resulted in 35% specificity and 91% sensitivity. It was also calculated the optimal self-assessed urine color threshold value for hypohydration was ≥ 3 (i.e., a self-assessed rating as U_{col} 3 or higher indicated hypohydration). Additionally, on the light and dark scale it was found that only 8 participants choose dark while the other 68 choose light. Also, of the 68 people that chose light 18 were categorized as hypohydrated. Of the 8 participant’s that chose dark, 4 were categorized to be euhydrated.

Table 3 Receiver operating characteristic evaluation of U_{col} measured in the self-assessment as a diagnostic tool for identifying hypohydration standard.

Predictive variable	Diagnostic Standard	Threshold	AUC	Sensitivity %	Specificity %
Self- U_{col}	U_{osmo}	3	0.65	91	35

Predictive variable was tested against the corresponding hypohydration diagnostic standard $U_{osmo} \geq 800$ mmol kg⁻¹.

Agreement between self- and lab-assessed urine color

Bland-Altman analyses were used to calculate the agreement between self- and laboratory-assessed U_{col} ratings (Fig. 2; $r = 0.31$, $P < .005$). Since U_{col} is an interval scale, numerous coordinates comparing mean U_{col} rating versus U_{col} rating difference happened more than once (i.e., multiple data points for each dot). The frequency of each coordinate's repetition is presented with the diameter of each marker in Fig. 2. Furthermore, the values for the x-axis are displayed in 0.5 integers. In this circumstance, the 0.5 integers allow for comparison of the measurement techniques and should not be mistaken as a U_{col} that was assessed to be between two other integers. The mean difference between self- and laboratory-assessed U_{col} was -.26 UC units. This means it was found participants tended to rate their U_{col} slightly lighter than the researchers. Furthermore, it was found as U_{col} became increasing darker, so did the discrepancy between the participant and researchers U_{col} ratings. Another finding of this study was that there was almost no difference in the accuracy between participants observing the urine through the stream or pooling in the urinal. The difference between U_{col} Self-Lab for observing the stream was -0.29 while observing into the urinal was -0.24.

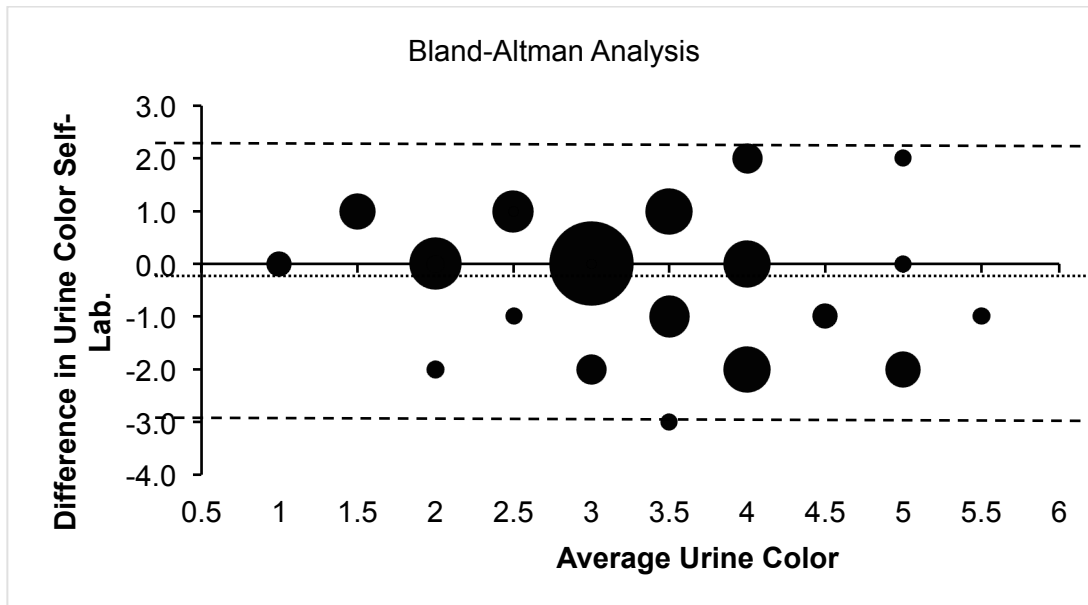


Fig. 2 Bland-Altman analysis of self-assessed urine color versus laboratory-assessed urine color. The x-axis, average urine color, is the mean of each self-assessed and laboratory-assessed sample. The y-axis, urine color difference, represents the difference between laboratory-assessed and self-assessed urine color for each sample. The area of markers indicates the relative frequency of corresponding data point (i.e., larger circles designate more occurrences). Upper and lower dashed lines represent 95% limits of agreement. Middle dotted line represents mean difference between respective Self- U_{col} and Lab- U_{col} .

IV. Discussion

There were three primary findings for this current study. First, it was found that U_{col} -self versus U_{osmo} was significantly positively correlated. Second, self-assessed U_{col} had “poor” capabilities to identify hypohydration overall. Last, self-assessed U_{col} had a reasonable ability to diagnose hypohydration with a U_{col} of 3 or greater. To our knowledge this is the first study showing the capabilities of self-assessed urine color as an index of hydration in adult males.

These findings show the capabilities of using urine color to assess hydration in males. Numerous studies have shown the strong positive relationships of U_{col} ratings in comparison with other hydration markers such as U_{osmo} and U_{sg} (Armstrong et al., 1994). Furthermore, studies have validated the classic urine color scale with changes in total body water during dehydration, rehydration, and exercise (Armstrong et al., 1998). Additionally, it has been found

that U_{col} is valid through visual assessment of changes in U_{col} , which was associated with the changes of body water change across different states of de- and rehydration. U_{col} has been shown to differ between adults habitually consuming low or high amounts of fluid on a day to day basis (Perrier et al., 2013). Our results show that men should have a moderate ability to assess their hydration state throughout their day.

Numerous experts have advised athletes to evaluate their own U_{col} to assess their hydration status (Armstrong et al., 1998). Our data suggest that on a normal daily basis, self-assessment of U_{col} has a moderate ability to identify hypohydration. The ROC analysis indicated a self-assessed U_{col} threshold value of 3 or greater to be constant with hypohydration. This is in agreement with the study by Kavouras and Colleagues that found Lab-evaluated U_{col} having a threshold value of 3 or greater for diagnosing hypohydration (2015). It must be stated though, that other studies such as Cheuvront and colleagues have calculated hypohydration to have a diagnostic value of up to 5.5 or greater in athletes (2010).

The difference between the lab- and self-assessed U_{col} thresholds was confirmed through the agreement plots that verify self-assessments tended to slightly underestimate U_{col} ratings (Fig. 2). This is in disagreement with a previous study by Kavouras and Colleagues that found children overestimated their urine color by 1 integer (2015). This discrepancy could be due to the children assessing their U_{col} in a plastic cup while the current study had the participants assess their U_{col} in a urinal. Also, the children were instructed to assess their U_{col} by looking at the stream while the current study allowed participant's to look at their stream or pooling of urine in the urinal. The Beer-Lambert Law states that light absorbance is equal to the product of three things; the concentration of the solution the light is passing through, the length of the solution the light passes through, and the absorption coefficient. As a result, there are two physical factors of

the sample container; the diameter of the urine cup and the material of the container. This can influence the amount of light absorbed by the container, influencing the color of the sample. This is why current studies transfer the samples into a 15 mL glass test tube to minimize any influence of light on the sample. Since, the participants observed the urine going into a urinal this could limit the amount of light passing through the urine causing participants to evaluate their U_{col} darker than the researcher.

The classic U_{col} scale is a well-known tool for evaluating hydration status. We believe the current study has achieved the task of testing its validity in young adult males. One limitation of this study is that the majority of participant's were college students between the ages of 18-22. Further studies should attempt to gather a more diverse age range. In conclusion, young adult males can moderately assess their U_{col} accurately.

References

- Adolph, E. F., & Rothstein, A. (1944). Voluntary dehydration. *Federation Proceedings. Federation of American Societies for Experimental Biology*, 3, 1-1.
- Armstrong, L. E. (2007). Assessing hydration status: The elusive gold standard. *Journal of the American College of Nutrition*, 26(5 Suppl), 575S-584S. doi:26/suppl_5/575S [pii]
- Armstrong, L. E., Maresh, C. M., Castellani, J. W., Bergeron, M. F., Kenefick, R. W., LaGasse, K. E., & Riebe, D. (1994). Urinary indices of hydration status. *International Journal of Sport Nutrition*, 4(3), 265-279.
- Armstrong, L. E., Soto, J. A., Hacker, F. T., Jr, Casa, D. J., Kavouras, S. A., & Maresh, C. M. (1998). Urinary indices during dehydration, exercise, and rehydration. *International Journal of Sport Nutrition*, 8(4), 345-355.
- Bar-David, Y., Urkin, J., & Kozminsky, E. (2005). The effect of voluntary dehydration on cognitive functions of elementary school children. *Acta Paediatrica (Oslo, Norway : 1992)*, 94(11), 1667-1673. doi:U1051312R8102RJ8 [pii]
- Chevront, S. N., Ely, B. R., Kenefick, R. W., & Sawka, M. N. (2010). Biological variation and diagnostic accuracy of dehydration assessment markers. *The American journal of clinical nutrition*, 92(3), 565-573.
- Ehrig, F., Waller, S., Misra, M., & Twardowski, Z. J. (1999). A case of 'green urine'. *Nephrology, Dialysis, Transplantation : Official Publication of the European Dialysis and Transplant Association - European Renal Association*, 14(1), 190-192.
- Guerra, A., Folesani, G., Mena, P., Ticinesi, A., Allegri, F., Nouvenne, A., & Meschi, T. (2014). Hippuric acid in 24 h urine collections as a biomarker of fruits and vegetables intake in kidney stone formers. *International Journal of Food Sciences & Nutrition*, 65(8), 1033-1038. doi:10.3109/09637486.2014.950210
- Kavouras, S. A., Arnaoutis, G., Makrillos, M., Garagouni, C., Nikolaou, E., Chira, O., & Sidossis, L. S. (2012). Educational intervention on water intake improves hydration status and enhances exercise performance in athletic youth. *Scandinavian Journal of Medicine & Science in Sports*, 22(5), 684-689. doi:10.1111/j.1600-0838.2011.01296.x
- Kavouras, S. A., Johnson, E. C., Bougatsas, D., Arnaoutis, G., Panagiotakos, D. B., Perrier, E., & Klein, A. (2015). Validation of a urine color scale for assessment of urine osmolality in healthy children. *European Journal of Nutrition*, doi:10.1007/s00394-015-0905-2 [doi]
- Kavouras, S. A., & Anastasiou, C. A. (2010). Water physiology: Essentiality, metabolism, and health implications. *Nutrition Today*, 45, S27-S32. doi:10.1097/NT.0b013e3181fe1713

- Manz, F., Wentz, A., & Sichert-Hellert, W. (2002). The most essential nutrient: Defining the adequate intake of water. *The Journal of Pediatrics*, *141*(4), 587-592. doi:S0022 3476(02)00147-6 [pii]
- Morimoto, Y., Sakuma, M., Ohta, H., Suzuki, A., Matsushita, A., Umeda, M., . . . Arai, H. (2014). Estimate of dietary phosphorus intake using 24-h urine collection. *Journal of Clinical Biochemistry and Nutrition*, *55*(1), 62-66. doi:10.3164/jcbn.14-15
- Perrier, E., Demazieres, A., Girard, N., Pross, N., Osbild, D., Metzger, D., . . . Klein, A. (2013). Circadian variation and responsiveness of hydration biomarkers to changes in daily water intake. *European Journal of Applied Physiology*, *113*(8), 2143-2151. doi:10.1007/s00421 013-2649-0 [doi]
- Perrier, E. T., Armstrong, L. E., Daudon, M., Kavouras, S., Lafontan, M., Lang, F., & Klein, A. (2014). From state to process: Defining hydration. *Obesity Facts*, *7 Suppl 2*, 6-12. doi:10.1159/000360611 [doi]
- Raymond, J. R., & Yarger, W. E. (1988). Abnormal urine color: Differential diagnosis. *Southern Medical Journal*, *81*(7), 837-841.
- Shephard, M. D., Penberthy, L. A., & Fraser, C. G. (1981). Short- and long-term biological variation in analyses in urine of apparently healthy individuals. *Clinical Chemistry*, *27*(4), 569-573.
- Tape, T. The Area Under an ROC Curve. In: *Interpreting Diagnostic Tests*. University of Nebraska Medical Center. <http://gim.unmc.edu/dxtests/ROC3.htm>. Accessed 08/02/2015.
- Thornton, S. N. (2010). Thirst and hydration: Physiology and consequences of dysfunction. *Physiology & Behavior*, *100*(1), 15-21. doi:10.1016/j.physbeh.2010.02.026 [doi]

Appendix



UNIVERSITY OF ARKANSAS

Office of Research Compliance
Institutional Review Board

February 23, 2015

MEMORANDUM

TO: Stavros Kavouras J.D. Adams
Evan Johnson Catalina Capitan
Lisa Jansen Rebecca Mishler
Thomas Vidal Mickey Hammer
Joseph Robillard LynnDee Summers
Ainsley Huffman Weldon Murry
Layne Nixon Lauren Smith
Berkley Vaughn Alison Schroeder
Jillian Fry Cameron Nichols

FROM: Ro Windwalker
IRB Coordinator

RE: PROJECT MODIFICATION

IRB Protocol #: 14-11-247

Protocol Title: *The Validation of Urine Color in Free-Living Adults*

Review Type: EXEMPT EXPEDITED FULL IRB

Approved Project Period: Start Date: 02/20/2015 Expiration Date: 11/19/2015

Your request to modify the referenced protocol has been approved by the IRB. **This protocol is currently approved for 400 total participants.** If you wish to make any further modifications in the approved protocol, including enrolling more than this number, you must seek approval *prior to* implementing those changes. All modifications should be requested in writing (email is acceptable) and must provide sufficient detail to assess the impact of the change.

Please note that this approval does not extend the Approved Project Period. Should you wish to extend your project beyond the current expiration date, you must submit a request for continuation using the UAF IRB form "Continuing Review for IRB Approved Projects." The request should be sent to the IRB Coordinator, 109 MLKG Building.

For protocols requiring FULL IRB review, please submit your request at least one month prior to the current expiration date. (High-risk protocols may require even more time for approval.) For protocols requiring an EXPEDITED or EXEMPT review, submit your request at least two weeks prior to the current expiration date. Failure to obtain approval for a continuation *on or prior to* the currently approved expiration date will result in termination of the protocol and you will be required to submit a new protocol to the IRB before continuing the project. Data collected past the protocol expiration date may need to be eliminated from the dataset should you wish to publish. Only data collected under a currently approved protocol can be certified by the IRB for any purpose.

If you have questions or need any assistance from the IRB, please contact me at 109 MLKG Building, 5-2208, or irb@uark.edu.