

Letter to the Editor

A Discussion of Creatinine Analysis in Single Collection Urine Specimens

Dear Sir:

In reference to the article of Needleman et al. [1], which implies that very low urine creatinine values are most likely due to adulteration treatment rather than to water intake, the following case is presented to indicate where water consumption was the interfering factor.

A pre-employment screen on EB was reported positive for THC (confirmed with alternate methodology) using a 20 ng/dL cutoff and estimated at greater than 50 ng/dL, relative to higher calibrator results. The individual, disbelieving our results and naturally concerned about his rejection for employment, returned two days later as a "walk-in" for a drug screen (pay up-front, results to subject only). During the information recording and sample collection (with standard collection protocol of door ajar, no running water, temperature tape evaluation), the subject revealed to the collector that he had been advised to drink 'lots of water' to dilute the drug. He had consumed 'about a gallon.' This sample was clear, measured 1.002 sp. gr. and 10.9 mg/dL creatinine. The sample tested negative for all drugs (negative for THC at 20 ng/dL cutoff). The report to the subject noted the unacceptably low sp. gr. and creatinine values and that these invalidated the test results. The subject again disputed the report and in a non-standard procedure was advised to return in the AM for a first morning void collection for testing. This sample was positive for THC in the 20 to 50 ng/dL range (confirmed) with a sp. gr. of 1.026 and creatinine of 238 mg/dL. We do not know the time frame during which the water was consumed by EB prior to the second sample analysis. Based on the in-lab voluntary water consumption by two male technologists of approximately 0.9 and 1.0 gallon of water, respectively, in a four hour period, we are aware that the sp. gr. of urine can be reduced to 1.000. In our experience a low creatinine value was a result of water intake and influenced the drug analysis results.

George M. Homer, Ph.D.
Barbara Born, B.S.
North Central Laboratories
436 E. River St.
Elyria, OH 44035

Reference

[1] Needleman, S. B., Porvaznik, M., and Ander, D., "Creatinine Analysis in Single Collection Urine Specimens," *Journal of Forensic Sciences*, Vol. 37, No. 4, July 1992, pp. 1125-1133.

Author's Response

Dear Sir:

Our manuscript was directed to the question of whether or not the creatinine level of a single urine specimen might be used as an indication of deliberate dilution of that specimen with water. It was not directly concerned with measurement of the specific gravity of the urine and the implication that measurement might have on interpreting the drug analysis results. The comments by Dr. Homer and Ms. Born, however, should provide additional impetus to examination of the issue of dilution of urine with respect to its meaning on analytical results.

The information provided by Dr. Homer, however, does raise a few questions. He

says the original analytical value for THC was screened "estimated at greater than 50 ng/dL (actual confirmation value not provided). The time between possible drug use and the first analysis, and, as stated, the time frame during which water was consumed is unknown. In our studies, near a gallon of water also produced a marked drop in creatinine (to 10–30 mg/dL) in six hours. Three days later, on the third analysis, after the subject consumed a gallon of water on Day 2, the individual provided a specimen which was still positive for THC in the 20–50 ng/dL (again the confirmation value was not provided). This does not appear to be realistic. Incidentally, it should be noted that the confirmation cutoff value used in our laboratory for THC is 15 ng/mL. The cutoff of 20 ng/dL cited by Dr. Homer should never have been used as a basis for assigning a positive finding for THC. This is probably the limit of linearity value. I assume Dr. Homer meant ng/mL for THC and ng/dL for creatinine.

To continue the argument above, considering the normal disappearance half-life for THC in urine of 22–24 hours, three or four days after drug use, one might reasonably expect the analytical value to have fallen to well below the cutoff value, not show positive at essentially the same value as on Day 1, barring new use of drug. Thus, the information provided for this undocumented specimen and presumed non-use of drugs, is seriously flawed and does not lend itself to rigid comparison with the controlled data provided in our manuscript—which still remains only a guideline for what might be taking place.

Saul B. Needleman, Ph.D.
Navy Drug Screening Lab
Bldg. 38H
Great Lakes, IL 60088

A Discussion of Creatinine Analysis in Single Collection Urine Specimens

Dear Sir,

Dilution (internal and external) of urine specimens that can alter drug testing outcome has been a problem since the advent of urine drug testing [1]. Detection of urine dilution is complicated by the limited size and number of studies which report mean creatinine concentrations for randomly collected urine specimens. Dr. Saul Needleman et al.'s study in the July 1992 issue of *Journal of Forensic Sciences* addressed this subject. They reported a mean urine creatinine value of 171.6 mg/dL for randomly collected urines [2]. Their results were based upon the analysis of 350 specimens obtained from military recruits [2]. While this is a relatively large study, the reported mean creatinine concentration is considerably higher than the mean urine creatinine values reported in a number of smaller studies [3–5]. There are a total of 372 measurements in these three reports with a weighted mean urine creatinine concentration of 96 mg/dL [3–5]. It is important to note that Needleman et al.'s mean concentration would have been higher had they accurately measured the 26 urine specimens with creatinine values > 300 mg/dL which they chose to report as 300 mg/dL [2]. There are two studies (total $N = 40$) which agree with Needleman et al.'s mean urine creatinine value [6,7]. The weighted mean urine creatinine concentration of these studies is 183 mg/dL [6,7]. The specimens used in the study by Hasday and Grum were not "true" random collections since all were evening collections collected for comparison with first morning specimens [6].

Concomitant with the publication of Needleman et al.'s article the abstract of a much larger study ($N = 7705$) appeared in *Clinical Chemistry* [8]. This study by Mayer and Hemphill reported a mean urine creatinine value of 120 mg/dL with a standard deviation of 72 mg/dL [8]. While the standard deviations in these two studies are quite large, the very large sample sizes more than compensate and the two means are *significantly* different ($P < 0.001$).

Several factors might account for the observed difference. For instance, the populations

may be different with regard to age or sex [9]. The sample collection techniques may differ with regard to time of collection or amount of notification (for example, no notice, first-morning specimens versus randomly collected specimens with notice of greater than 4 hours).

If one assumes that the specimens analyzed by Mayer and Hemphill (working at a commercial laboratory) are largely pre-employment tests, it could be hypothesized that these individuals had sufficient time to internally dilute their specimens to avoid detection of drug use. If a significant number of individuals undertook this dilutional effort, it might account for the lower mean creatinine concentrations. However, Mayer and Hemphill re-analyzed all negative specimens with creatinine concentrations below 20 mg/dL ($N = 262$) for evidence of THC or cocaine [8]. They found only 24 specimens with measurable amounts THC or cocaine [8]. While the positive rate of these "diluted" specimens was higher (9.1%) than the positive rate of specimens with a creatinine concentration above 20 mg/dL (3.4%), the overall percent of the dilute samples containing drug metabolites was relatively low [8]. What else might account for this difference?

I believe, the answer to the difference lies in some of the articles cited above and in some additional articles. If one examines a large enough number of truly random urine specimens and compares them to 24 hour specimens, one would expect a much greater range and variation in creatinine concentration among the randomly collected specimens. However, both types of studies yield a time-averaged concentration. Therefore, the mean urinary creatinine concentration for a *large* number of randomly collected specimens would be expected to approximate the mean concentration found for a large number of 24 hour urine specimens.

Seven studies reporting urine creatinine concentrations from 24 hour urine specimens ($N = 613$) yielded mean urine creatinine values of 89, 90, 93, 101, 108, 132, and 154 mg/dL [5,7,9-13]. Calculation of the weighted mean for these seven studies yields an average urine creatinine concentration of 108 mg/dL. These time-averaged values agree with the mean urine creatinine value for the large number of randomly collected specimens reported by Mayer and Hemphill.

Three reports of mean urine creatinine concentration for first morning specimens ($N = 156$) yielded a weighted mean value of 176 mg/dL [4,6,9]. This is very close to the value obtained by Needleman et al.

For these reasons I suggest that the "true" mean creatinine concentration for randomly collected specimens is most likely to be close to 120 mg/dL. I suggest that the higher values reported in Needleman et al.'s article may be due to the non-randomness of collection and/or some other factor such as the situational stress of being a new military inductee causing concentration of the urine specimens.

William E. Ottinger, Ph.D.
Assistant Professor
Department of Clinical Laboratory Sciences
University of Texas Health Science Center at San Antonio
7703 Floyd Curl Drive
San Antonio, Texas 78284-7772

References

- [1] Hoffman, A. and Silvers, J., *Steal This Urine Test: Fighting the Drug Hysteria in America*, 1987, Viking Penguin Press.
- [2] Needleman, S. B., Porvaznik, M., and Ander, D., "Creatinine Analysis in Single Collection Urine Specimens," *Journal of Forensic Sciences*, Vol. 37, No. 4, July 1992, p. 1125.
- [3] Gibb, J. A., Ogston, S. A., Paterson, C. R., and Evans, J. R. "Discriminant Functions in Differential Diagnosis of Hypercalcemic Patients," *Clinical Chemistry*, Vol. 36, 1990, p. 358.
- [4] Lafolie, P., Beck, O., Blennow, G., Boreus, L., Borg, S., Elwin, C.-E., Karlsson, L., Odelius,

- G., and Hjemdahl, P., "Importance of Creatinine Analyses of Urine When Screening for Abused Drugs," *Clinical Chemistry*, Vol. 37, 1991, p. 1927.
- [5] Gokce, C., Gokce, O., Baydinc, C., Ilhan, N., Alasehirli, E., Ozkucuk, F., Tasci, M., Atikeler, M. K., Celebi, H., and Arslan, N., "Use of Random Urine Samples to Estimate Total Urinary Calcium and Phosphate Excretion," *Archives of Internal Medicine*, Vol. 151, 1991, p. 1587
- [6] Hasday, J. D. and Grum, C. M., "Nocturnal Increase of Urinary Uric Acid:Creatinine Ratio," *American Review Respiratory Diseases*, Vol. 135, 1987, p. 534.
- [7] Shephard, M. D. S., Penberthy, L. A., and Fraser, C. G., "Short- and Long-Term Biological Variation in Analytes in Urine of Apparently Healthy Individuals," *Clinical Chemistry*, Vol. 27, 1981, p. 569.
- [8] Mayer, B. and Hemphill, P., "Evaluation of Urine Creatinine as a Marker to Identify Dilute Specimens Being Analyzed for Drugs of Abuse," *Clinical Chemistry*, Vol. 38, 1992, p. 1008.
- [9] Hesse, A., Classen A., Knoll, M., Timmermann, F., and Vahlensieck, W., "Dependence of Urine Composition on the Age and Sex of Healthy Subjects," *Clinica Chimica Acta*, Vol. 160, 1986, p. 79.
- [10] Kreig, V. M., Gunsfer, K.-J., Steinhagen-Thiessen, E., and Becker, H., "Vergleichende quantitative Alalytik Klinisch-chemischer Kenngrößen im 24-Stunden-Urin und Morgenurin," *Journal of Clinical Chemistry and Clinical Biochemistry*, Vol. 24, 1986, p. 863.
- [11] Araki, S., Hiroshi, A., and Murata, K., "Adjustment of Urinary Concentration to Urinary Volume in Relation to Erythrocyte and Plasma Concentrations: An Evaluation of Urinary Heavy Metals and Organic Substances," *Archives of Environmental Health*, Vol. 41, 1986, p. 171.
- [12] Bingham, S. A., Williams, R., Cole, T. J., Price, C. P., and Cummings, J. H., "Reference Values for Analytes of 24-h Urine Collections Known to be Complete," *Annals of Clinical Biochemistry*, Vol. 25, 1988, p. 610.
- [13] Gowans, E. M. S. and Fraser, C. G., "Biological Variation in Analyte Concentrations in Urine of Apparently Healthy Men and Women," *Clinical Chemistry*, Vol. 33, 1987, p. 847.

Author's Response

Dear Sir:

"Normal" values for creatinine in urine vary considerably between 100–200 mg/dL [1–3], placing our value (171.6 mg/dL) [4] at the upper "normal" end and Dr. Ottinger's suggested weighted mean value of 96 mg/dL at the lower end. Different "normal" ranges may in fact exist as a function of the population from which they were derived.

The highest standard used in our study was one of 300 mg/dL creatinine. Because the actual values for the 26 specimen in our study having greater than 300 mg/dL creatinine were only about 3% greater than the 300 mg/dL reported (established by dilution), our mean of 172 mg/dL is representative of the real specimen pool we used. Further, by his own literature citations, Dr. Ottinger reports three small studies which gave a lower final mean, but two which gave a larger mean value than ours. Thus, the final mean we obtained lies well within the generally accepted range for creatinine in urine.

Where we differed significantly from most published studies is in basing our mean finding on single urine specimen in contrast to the general practice of using 24 hour pool collections. The purpose of our study was to determine whether a single urine specimen could be used diagnostically to point to possible adulteration of urine specimen in drug testing. Almost all of the specimen used in our analytical selection were identified as being first or early morning samples. Since most sampling in the military is done in the morning, our sampling mimicked actual specimen collection procedures, and we felt justified in equating our specimen pool with the population it was intended to challenge.

We recognized that these selection criteria in themselves, however, might yield a slightly higher value than might be obtained from pooled urine or from a urine specimen obtained later in the day. Further, since our specimen were drawn from an essentially non-random, dominantly male population, the creatinine values would be higher than in a totally random population.

In selecting our criteria for suggesting that a specimen might be adulterated, however, we adopted a very conservative approach in using the value of 100 mg/dL as the 'high'

normal, a value not too far different from Dr. Ottinger's calculated weighted mean derived from seven studies—108 mg/dL, and permitting as much as a three-fold dilution of urine to be possible (down to 30 mg/dL) to provide the "low" end of normal. Thus, only specimen with creatinine values below 30 mg/dL come under consideration as being adulterated.

The study of Mayer and Hemphill [5] would appear to prove the contention of our own work in indicating that specimen with measurable quantities of drug below established cutoff values, might be the result of individual donors attempting dilution in hope of avoiding detection.

Thus, the main thrust of our study, the determination of whether or not single collection specimen can be used to detect possible adulteration of urine by dilution is answered. In addition, the question of the extent to which extensive ingestion of liquid might effect the final analytical drug results also is indicated on the basis of the controlled ingestion studies reported therein.

Saul B. Needleman, Ph.D.
Navy Drug Screening Laboratory
Great Lakes, IL 60088

References

- [1] Wallach, J., "Interpretation of Diagnostic Tests," *A Handbook Synopsis of Laboratory Medicine*, Little, Brown and Co., New York, 1974, p. 16.
- [2] Thompson, R. H. S. and Wootton, I. D. P., "Biochemical Disorders in Human Disease," Academic Press, p. 263.
- [3] The Merck Index (Martha Windholz, Ed.), 10th edition, Merck and Co., Rahway, N.J., 1983.
- [4] Needleman, S. B., Porvaznik, M., and Ander, D., "Creatinine Analysis in Single Collection Urine Specimens," *Journal of Forensic Sciences*, Vol. 37, No. 4, July 1992, p. 1125.
- [5] Mayer, B. and Hemphill, G., "Evaluation of Urine Creatinine as a Marker to Identify Dilute Specimens Being Analyzed for Drugs of Abuse," *Clinical Chemistry*, Vol. 38, 1992, p. 1008.