A Positive Urine Alcohol with Negative Urine Ethyl-Glucuronide

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ABSTRACT

Urine ethanol is a common finding in urine drug-screening results. An unexpected finding of alcohol in urine can have serious implications for patients who have committed to pain-management contracts or are being counseled for substance abuse. Although laboratory screening for urine ethanol is highly specific and sensitive, the source of the ethanol can sometimes be unclear. This case report describes a 44-year-old Caucasian man with positive urine ethanol results who reported having abstained from alcohol. A test for urine ethyl-glucuronide and ethyl sulfate was used to validate the information in the patient history stating that the patient had not consumed alcohol for 1 year. Evaluation of the urinalysis results from the patient revealed fermentation in the context of glucosuria as the source of the urine ethanol.

Keywords: ethanol, ethyl-glucuronide, urine glucosuria, false positive alcohol, fermentation, drug screen

Clinical History

An ambulatory care pharmacist contacted the laboratory concerning an unexpected positive urine-ethanol test result. The patient was a 44-year-old Caucasian man who was being managed for diabetes mellitus type 2, lumbar radiculopathy, and shoulder and rib pain. The patient had had paraplegia for the past 20 years. He presented to the primary care department with concern about his blood glucose levels which, when measured at home, were consistently approximately 300 mg per dL. He was taking insulin (70 units twice daily and 45 to 50 units before meals). The patient reported that he had had a sweet taste in his mouth for the past few days. He also reported an increased urge to urinate and frequent urinary-tract infections.

Abbreviations

OTP, opioid therapy plan; THC, tetrahydrocannabinol; ADH, alcohol dehydrogenase; NAD, nicotinamide adenine dinucleotide; ETG, ethyl-glucuronide; ETS, ethyl sulfate; LC-MS/MS, liquid chromatography–mass spectrometry; NA, nonapplicable; RBC/HPF, red blood cells per high power field; WBC, white blood cells; Squam epi, squamous epithelial cells

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The patient had been prescribed hydrocodone-acetaminophen (the Rx is for tablets containing 10 mg hydrocodone and 325 mg acetaminophen [both drugs are in each tablet]), taken as needed for back and joint pain. At our institution, opiate prescriptions require a patient to consent to an opioid therapy plan (OTP). The OTP is a provider-patient contract that requires the patient to abstain from alcohol and other recreational drugs (such as tetrahydrocannabinol [THC]), while taking opioid medications. To monitor compliance with the OTP, routine urine drug screening is performed on patients before their opiate prescriptions are refilled. The drug screening results for this patient, obtained during this visit, are shown in **Table 1**.

The urine creatinine levels of the patient were greater than 20 mg per dL; those levels were sufficiently concentrated to merit drug screening. Our laboratory uses an Emit II Plus Ethyl Alcohol Assay (Beckman Coulter Inc), which utilizes alcohol dehydrogenase (ADH) and the coenzyme nicotinamide adenine dinucleotide (NAD). ADH catalyzes the oxidation of ethanol to acetaldehyde; this process reduces NAD to NADH, which is then measured at 340 nm, allowing for the quantitation of alcohol in the specimen.[1]

The patient tested positive for opiates, as expected. He also tested positive for ethanol, in violation of his OTP. The patient reported that his most recent alcohol use was 1 year

Table 1. Screening Results for the Patient, a 44-Year-Old Caucasian Man

Drug Screen Results		
Drug (Urine)	**Result**	**Positive Cutoff Value**
Amphetamines	Negative	500 ng/mL
Barbiturates	Negative	200 ng/mL
Benzodiazepines	Negative	200 ng/mL
THC	Negative	50 ng/mL
Cocaine	Negative	150 ng/mL
Ethanol	Positive	10 mg/dL
Creatinine	41.1 mg/dL	NA
Urinalysis		
Component	**Value**	**Reference Range**
Specific gravity	1.030	1.003–1.035
pH	5.0	5.0–8.0
Leukocytes	Negative	Negative
Nitrite	Negative	Negative
Protein	Negative	Negative
Glucose	>500 mg/dL	Negative
Ketones	Trace	Negative
Hemoglobin	1+	Negative
RBC/HPF	0–3	None
WBC	0–5	None
Squam epi	Many	None
Bacteria	4–10	None

THC, tetrahydrocannabinol; NA, nonapplicable; RBC/HPF, red blood cells per high power field; WBC, white blood cells; Squam epi, squamous epithelial cells.

ago. Based on his symptoms, urinalysis and microscopy were also performed (**Table 1**). A urine ethyl-glucuronide (ETG) and ethyl sulfate (ETS) test was ordered on an aliquot of the urine (the same specimen that was already obtained and had tested positive for ethanol). The ETG/ETS test result came back 7 days later as being negative.

Discussion

Given the marked glucosuria and the presence of bacteria in the urine, the possibility of urine fermentation was considered. With the presence of sugar and microbes and with the passage of time, it was hypothesized that the presence of ethanol detected in the urine was not due to ingestion of ethanol but, rather, that ethanol had formed as the product of microbial fermentation of glucose. To test this hypothesis, ETG/ETS was ordered on the specimen and sent to our reference laboratory. A cutoff value of 500 ng per mL was used to delineate a positive ETG result and 250 ng per mL for a positive ETS result. ETG and ETS are direct metabolites of ethanol that are present in urine after ingestion of products containing ethyl alcohol. These metabolites can also result from the use of products that contain alcohol but were not intentionally consumed, such as mouthwash or hand sanitizer. Measurement of ETG can be performed using immunoassay or liquid chromatography–mass spectrometry (LC-MS/MS). If using an immunoassay screen, a positive result is then confirmed by LC-MS/MS. Our reference laboratory uses LC-MS/MS to directly measure ETG and ETS, with no initial screening step.

ETG arises from the detoxification of alcohol by its conjugation with activated glucuronic acid (**Figure 1**). Testing urine for ETG to detect recent alcohol use has been studied since the 1960s.[2] This glucuronide detoxification pathway is estimated to account for only 0.5% to 1.5% of ethanol elimination.[3] Despite being the result of a minor elimination pathway, ETG has become useful as a marker for alcohol consumption. Unlike serum, blood, or urine alcohol, ETG remains detectable in urine for days after alcohol consumption. A positive urine ETG result strongly suggests that a patient has consumed alcohol in the past 80 hours.[4]

By measuring urine ETG/ETS concentration, one can obtain a second independent, longer-duration marker of alcohol use. In a patient with a positive urine ethanol screening result, confirmation via mass spectrometry of ethanol is often carried out. However, ordering of ETG/ETS to confirm a positive urine alcohol result is not common practice in our medical system, nor is it routinely used in other clinical laboratories in our region. In my experience, most health care providers are not aware of the option of ETG/ETS testing.

In this case, the patient was adamant that he had not consumed alcohol. Because the OTP and further prescription of opioids were contingent on a negative urine alcohol result, it was believed that urine ETG/ETS testing was warranted in this case. If the ETG and ETS results had been positive, those results would have strongly suggested that the patient had recently consumed alcohol. However, ETG by itself still does not deliver definitive proof of alcohol use. By adding the marker ETS, the specificity of testing for alcohol use is increased: unlike ETS, ETG can arise from microbial fermentation or metabolism.[5] Bacterial contamination also can cause a false-negative ETG result, due to bacterial decomposition of ETG[6]; ETS, however, is not affected by these contamination issues. For this reason, ETS is a useful addition to ETG testing. In this patient, ETG and ETS results were negative. These findings strongly suggest that the ethanol was present in the urine due to bacterial fermentation.

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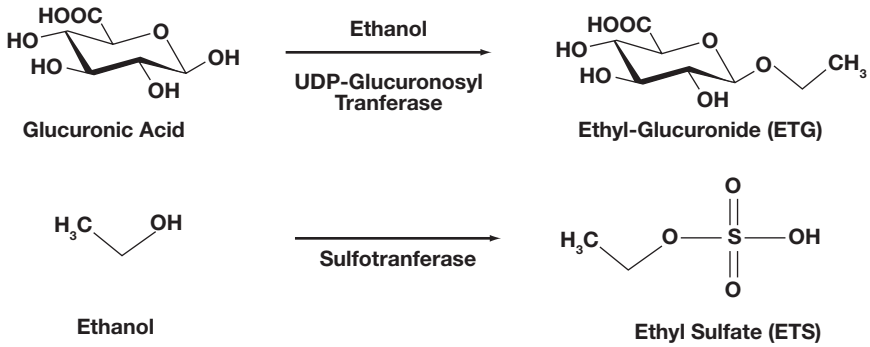

Figure 1

Description of the creation of ethyl-glucuronide (ETG) from the detoxification of alcohol. UDP indicates urine diphosphate; ETS, ethyl sulfate.

As brewers and winemakers are aware, *fermentation* is the biochemical process of converting sugars, in the absence of oxygen, into alcohol (and carbon dioxide). Bacteria and yeast can ferment sugars. If a liquid has a sufficient amount of sugar (>500 mg/dL glucose, in this case), along with the presence of bacteria and the passage of time, fermentation in vitro can occur.[7] In this case, the urine was collected at 10:30 AM, transported to our main laboratory across the city, and tested at 5:00 PM. The specimen was transported at ambient (room) temperature. A period of 6.5 hours seems to be an ample amount of time to allow for fermentation. As Sulkowski et al[8] showed, fermentation of urine glucose is temperature dependent and could be prevented by freezing or by using sodium fluoride as a preservative. This case highlights the fact that specimens that cannot be tested immediately should be transported and held in refrigerated conditions, to reduce the chances that fermentation will occur in the specimens.

Laboratory Role in Diagnosis

The laboratory played a key role in the course of treatment for this patient. The initial urine ethanol result generated by the laboratory was not a false positive but rather was a true positive for ethanol, albeit ethanol produced in vitro.

Because of a culture of strong communication between the laboratory staff and health care providers, the pain management pharmacist working with the patient felt comfortable asking about this unexpected result. The laboratory director then suggested ETG/ETS testing on the already obtained specimen. The negative ETG result, coupled with the negative ETS result, lent credence to the assertion of the patient that he had not consumed alcohol. Although it is possible that ETG or ETS were present below the positive cutoff concentrations, it was deemed more likely that the patient had complied with the terms of his OTP, and that the presence of urine ethanol in his specimen had resulted from fermentation of the tested specimen rather than alcohol consumption by the patient.

Patient Follow-Up

The patient was prescribed cephalexin (500 mg, 4 times per day for 7 days) and topical silver sulfadiazine (1%) for his urinary tract infection. He recovered from the infection and continued to comply with his plan for pain management via opiate therapy. **LM**

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