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Urine Samples Tampering Pattern for Drugs of Abuse Testing: Experience of the Saudi Arabia Poison Control Centers

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Abstract

Recently, urine substance of abuse (SOA) testing in the pre-employment/workplace and suspected SOA settings has become common in many countries all over the world. There have been multiple published research recommending the performance of the urine sample validity test (SVT) for substance of abuse testing administered in the pre-employment/workplace and suspected SOA settings. On the opposite side, very little researches focusing on variable procedures of urine adulteration in (SOA) testing process, including diluted, substituted, adulterated, and invalid tests. The current research investigated 7985 submitted urine drug test samples for sample validity test from pre-employment/workplace and suspected SOA settings in Saudi Arabia over one year. All preliminary immunoassay screen-positive urine sample drug tests were confirmed by gas chromatography/mass spectrometry and liquid chromatography/mass spectrophotometry. This article found that the prevalence of tampering (diluted, substituted, or invalid tests) in urine samples from the pre-employment/workplace and suspected settings were 0.87% and 0.69%, respectively. The percentage of diluted, substituted, adulterated and invalid urine specimens from the pre-employment/workplace and suspected cases were 75%, 21.4%, 1.7%, 1.7% and 63.6%, 36.4%, 0%, 0% respectively. The most common substance of abuse detected from the pre-employment/workplace and suspected specimens were cannabis, followed by amphetamines. We recommend that all urine samples taken for substance of abuse testing from both the pre-employment/workplace and suspected settings need to be investigated for validity.

Keywords: Substances of abuse tests; Adulterant agents; Pseudo-negative results

Introduction

Urine sample adulteration is very serious problem in forensic urine drug testing process. Sample adulteration is usually performed by substitution, dilution or the addition of adulterants agents including so called "masking agents" sold commercially. Adulteration process is defined as the tampering or manipulation of a urine sample with the intention of changing the test results [1].

The use of adulterant agents can cause false negative results in drug tests by either interfering with the screening test procedure and/or destroying the drugs present in the urine sample. Dilution attempts may also be employed in an attempt to release false negative drug test results. From the clinical point of view, the accepted method to test for adulteration or dilution attempts is to determine certain urinary characteristic parameters such as creatinine level, pH degree, and specific gravity and to investigate the presence of glutaraldehyde, nitrite and oxidants/pyridinium chlorochromate (PCC) in urine sample [2].

Drug abuse has become one of the major public health problem all over the world. In Saudi Arabia, Bassiony [3] reported that cannabis, amphetamine and opiates were the most widely used illicit drugs detect in urine samples collected from suspects who were arrested for possessing and/or taking illicit drugs. Over the past few decades, pre-employment drug testing has become a common mandatory routine practice in the world workplace [4-8]. Pre-employment drug testing poison control laboratories certified by the Saudi Arabia governmental authority are performing roughly 46,000 sample per year [3].

In urine samples for drug abuse testing, collected in Canada, by checking the dilution rate only, the researchers [9] documented that 6.7% of 38,431 urine samples were dilute. To the best of our knowledge, there were a few researches concerning further urine sample validity tests for urine sample of suspected substance of abuse criminal cases, including substituted, invalid, or adulterated procedures. For pre-employment/workplace drug testing of urine samples, there have been several reports concerning urine specimen validity tests for substances abuse tests [10-12]. The aim of this article was to clarify our findings from urine samples adulteration attempts detected from specimen validity tests results, involving the pattern adulteration; the rates of dilution, substitution, adulteration, and finally invalid samples prevalence, in cases of pre-employment/workplace and suspected drug of abuse testing for urine samples in Saudi Arabia over one years.

Materials and Method

Materials

Ministry of health/Saudi Arabia have nine urine drug abuse-testing laboratories inside poison control centers, certified by the legal authority in Saudi Arabia. 7,985 urine specimens from pre-employment/workplace drug testing and suspected settings for drug

abuse testing were investigated by urine specimen validity tests during the period of November 1, 2015 to October 31, 2016. Of these urine specimens, 6,402 (80.2%) came from pre-employment and workplaces. The other 1,583 (19.8%) urine specimens came from General Directorate of Police Forces. Urine sample collection was guided by the Drug Abuse Urine Collection Guideline of the TFDA, which was applied in August 1999. Urine donors were witnessed and placed in a room with no access to water or any other type of detergents. Higher research committee of general directorate of poison control center/ Saudi Arabia (HRC-GDPCC-SA- 0002) approved this study.

Drug of abuse test protocol and technique

Regarding protocol of urine collection sample for drug of abuse tests, we utilised a revised Mandatory Guidelines for Federal Workplace Drug Testing Programs (Guidelines), 73 FR 71858 (November 25, 2008) for urine testing.(10) We investigated immunoassay procedure by ARCHITECT Version ci 4100 with ARCHITECT ®D.A.U (drug of abuse in urine) ci 4100 Opiate, Benzodiazepine, Cannabis, Amphetamines, Cocaine and Barbiturates assay kits, for qualitative analysis in human urine.

If the immunoassay test result was below the cutoff point, the specimen was reported as negative. If the immunoassay test result at or above the cutoff point, result was positive, laboratories further confirmed the identity of the drug or drug metabolite definitively by using gas chromatography/mass spectrometry (GC/MS) (Agilent, 6890/5973N, Hewlett-Packard, Palo Alto, CA, USA) and Thermo Finnigan LTQ FT Ultra High Performance Mass Spectrometer. The cutoff levels of each drug in urine for immunoassay preliminary procedure and GC/MS confirmatory procedures were mention in (Table 1).

Drugs of Abuse Type	Cut-off Level (ng/ml)	
	Preliminary Procedures	Confirmatory Procedure
Opiates	300 ng/ml	200 ng/ml
Amphetamines	300 ng/ml	250 ng/ml
Benzodiazepines	200 ng/ml	100 ng/ml
Cannabinoids	25 ng/ml	15 ng/ml
Cocaine	300 ng/ml	150 ng/ml
Tramadol	300 ng/ml	100 ng/ml
Barbiturates	200 ng/ml	200 ng/ml

Table 1: Cut-off levels in ng/ml for drugs of abuse tested for urine specimen.

Drug of abuse test protocol and technique

For every sealed urine sample submitted for a drug abuse test from the pre-employment, workplace or suspected drug testing of urine samples, the collection process was under the chain of custody principle and then the samples were sent to poison control centers/ drug of abuse department. For every sample that underwent urine SVT, we measured the creatinine level, degree of specific gravity, pH degree, presence of the next mentioned adulterant agents Nitrite, Glutaraldehydes and Oxidants with Abon Biopharm Multi-Drug Screen rapid detection kits.

The analyst used rapid detection kit with a pH range of 0-14. Of all the urine specimens, >99% were in the range of pH 5-8 and only three cases were reported had pH >10.

The reporting results for adulteration attempts on urine samples were classified as follows [11].

(1) A urine sample was documented as diluted when the creatinine concentration level was ≥ 2 mg/dL but <20 mg/dL and the specific gravity degree was >1.0010 but <1.0030 on a single aliquot. A diluted sample is a urine sample with creatinine and specific gravity levels lower than expected for human urine sample.

(2) A urine sample was documented as substituted sample when the creatinine concentration level was <2 mg/dL and the specific gravity degree was <1.0010 or >1.0030. A substituted specimen is a urine specimen with creatinine and specific gravity values that are so decreased or strange that they are not consistent with normal human urine sample.

(3) A urine sample was recorded as adulterated sample if the pH degree was <3 or >11. An adulterated sample is a urine sample containing a substance that is not a normal constituent of urine composition or containing an endogenous substance not present at a normal physiological concentration of urine.

(4) A urine sample that did not meet any of the above criteria (diluted, substituted, or adulterated) but was clearly not normal was reported as invalid urine sample.

The drug test panels for the urine samples from the pre-employment/workplace and suspected settings were investigated according to the accredited protocol in Saudi Arabia. For employment/ workplace samples, accredited drug test panel were amphetamines, cannabinoids, opiates, cocaine, barbiturate, benzodiazepines. While for the suspended cases of drugs of abuse as previous mentioned panel plus ethanol and other tests were involved as necessary.

Results

Table 2 describes the prevalence of dilute, substituted, and invalid urine samples from the pre-employment/workplace and suspected settings during the investigated duration. The prevalence of diluted, substituted, adulterated or invalid urine samples from the pre-employment/workplace was (56 cases) 0.87%, higher than that of the suspected specimens, which was (11 cases) 0.69%. Dilution was the predominant procedure of tampering in both pre-employment/ workplace and police suspected urine samples settings.

The prevalence of diluted, substituted, adulterated and invalid urine samples for pre-employment/workplace and suspected cases are mentioned in Table 2 as the following (75%, 21.4%, 1.7%, 1.7%) and (63.6%, 36.4%, 0%, 0%), respectively.

Pre-employment/workplace settings total no. (56)	Suspected settings total no. (11)
Diluted	
42 cases (75.0%)	7 cases (63.6%)
8 cases with mild degree of dilution and creatinine level 15-20 mg/dl.	2 cases with moderate degree of dilution and and creatinine level 10-15 mg/dl.
15 cases with moderate degree of dilution and and creatinine level 10-15 mg/dl.	5 cases with high degree of dilution and the creatinine level <10 mg/dl
19 cases with high degree of dilution and the creatinine level <10 mg/dl	
Substituted	
12 cases (21.4%)	4 cases (36.4%)
12 cases with water substitution	1 cases with water substitution
	2 cases with ethanol substitution
	1 case with orange juice substitution
Adulterated	
1 case (1.7%)	———
One case with detergent.	
Invalid	
1 case (1.7%)	———
One male case replaces the sample with a female pregnant urine sample with positive HCG test	

Table 2: Prevalence of diluted, substituted, adulterated and invalid urine samples from pre-employment/workplace and suspected settings.

These data indicate that dilution was the most common method of tampering in both pre-employment/workplace and suspected urine samples (69.6%) and (63.6%), respectively. There were two adulterated urine samples by addition of highly concentrated liquid detergent to the investigated urine sample, from the workplace setting. Among all of investigated urine specimens, two specimens (2.4%) had a pH value outside the range of 5-8. One had a pH value <3 (as adulterated with orange juice) and the second one had a pH value >10 (as adulterated with detergent).

By confirmatory procedure by GC/MS and LC/MS the urine drug positive rates were 3.6% and 69.6% for the pre-employment/workplace and suspected settings respectively. The most common drug identified from suspected samples was cannabis (48.3%), followed by amphetamine (37%), followed by alcohol (13.9%) finally opiates including morphine and codeine (1.5%). The most common drug identified from the pre-employment/ workplace sample was cannabis (56.1%), followed by amphetamine (22.7%). No specimens from either the pre-employment/workplace or suspected setting were positive for cocaine.

Discussion

Fraser and Zamecnik [13] in Canada observed that 6.8% of urine samples from the offenders on conditional release in the community were dilute. On the same aspect, United States a toxicology laboratory revealed that, 4.94% of 4227 specimens were associated with dilutional attempt [14]. In current study, the rate of dilutional attempts of the collected specimens were 0.87% and 0.69% from pre-employment/workplace and suspected specimens respectively; this may be attributed to a highly strict regulatory rules in the collecting protocol of the investigated urine specimens.

In this study, the urine specimen tampering procedure (diluted, substituted, adulterated and invalid) prevalence from the pre-employment/workplace and the suspected specimens were (75%, 21.4%, 1.7%, 1.7%) and (63.6%, 36.4%, 0% 0%), respectively. In one study in United States, there were 6,800,000 urine specimens were collected for drug of abuse testing protocol under federal law. Of these urine samples, 2.1% gave a drug positive result and 0.15% were reported with diluted, substituted, or invalid tampering attempts [1]. For the pre-employment/workplace urine samples in the current research, the researchers revealed 56 cases (0.87%) with diluted, substituted, adulterated or invalid, higher than the 0.15% mentioned in the results of Bush [1]. The higher tampering rate (0.87%) in pre-employment/workplace current research specimens compared to that from the United States (0.15%) may be attributed to that most of the investigated cases were pre-employment 90.3% with long preparatory chance to perform the tampering procedure.

In the current research, dilution was the most common procedure of tampering of urine samples from both pre-employment/workplace and the suspected specimens settings. Diluting urine sample is often the easiest way to make a false negative drug test result [15]. In the same side of our results, Beck et al. mentioned that 11% of all urine

samples delivered to toxicology laboratory for drug abuse testing were diluted (creatinine <4 mmol/L) [16].

In Cook et al. [17] reported that the measurement of urine pH degree is useless indicator in detecting the dilutional status of urine sample and recommended that the measurement of pH degree is a valuable indicator for assessing chemical adulteration attempts. Because the kidneys are limited to producing urine within the pH range of 4.5-8, pH values beyond this range are highly indicator for adulteration.

Burrows et al. [18] mentioned that urine should be reported as adulterated if the pH was <3 or >11. In this study, only one adulterated cases were found. Jaffe et al. [19] mentioned that, the assumption that adulteration rarely occur. They also found that, in a healthy case, the specific gravity of a urine sample is expected to be ≥1.003 and have a pH between 3 and 11; thus, a pH or specific gravity outside of this range may indicate directly a chemical adulteration [19].

The most common drug identified from the suspected samples in the current study was cannabis 48.3% followed by amphetamines (37%). Concerning substance of abuse problems in Asia, Bart [20] mentioned that, there has been an increase in amphetamine to be appear as a globalized abuse throughout Asia.

Conclusion

In conclusion, for the pre-employment/workplace and suspected urine samples, we reported our experience in urine specimen validity test including diluted, substituted, adulterated and invalid prevalence in Saudi Arabia. For the pre-employment/workplace and suspected urine specimens, we reported urine tampering attempts (diluted, substituted, adulterated or invalid) prevalence from the pre-employment/workplace and suspected specimens was 0.87% and 0.69, respectively, in current study. We recommend that all urine samples for substance abuse testing from the pre-employment/workplace and suspected settings mandatory undergo a specimen validity test.

Conflicts of Interest

All authors declare no conflicts of interest.

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