Dosage des amphétamines dans les cheveux, la salive et les urines

Analysis of amphetamines in hair, oral fluid and urine

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(Réçu le 7 novembre 2005 ; accepté le 17 décembre 2005)

RÉSUMÉ
Des révisions proposées aux directives fédérales incluant l'utilisation potentielle des cheveux, de la salive ainsi que des urines pour le dépistage des conduites addictives en entreprise ont été publiés dans le registre fédéral en 2004. Cette étude a été conçue pour déterminer le taux de positivité de différents types d'échantillons dans une population toxicomane et dans une autre niant sa consommation de méthamphétamine. Cette étude a été menée sur 200 sujets, la moitié admettant une consommation de méthamphétamine, l'autre moitié la niant. Chaque sujet a donné un échantillon d'urine, de salive et de cheveux prélevés au moment de l'entrevue. Pour chaque sujet, des informations sur leur consommation de stupéfiants, incluant la fréquence de consommation, leur appartenance ethnique, leur âge, leur sexe et la couleur de leurs cheveux ont été enregistrées. Les échantillons ont été analysés pour la méthamphétamine, l'amphétamine, la 3-4 méthylendioxy- méthamphétamine (MDMA), son métabolite la méthylendioxy-amphétamine (MDA) et la

SUMMARY
Proposed revisions to the Federal guidelines, which include the potential use of hair and oral fluid, as well as urine, for workplace drug testing were published in the Federal Register in 2004. This study was designed to determine the positivity rate in various specimen types, both in a drug using population and a population denying methamphetamine use. The study enrolled 200 subjects, half of whom admitted to methamphetamine use, half who did not. Each subject provided a urine sample, an oral fluid and a hair specimen taken from the head at the time of interview. Information on drug use, including time of last use, frequency of use, ethnicity, age, sex and hair color were recorded for each subject. The hair specimens were analyzed for methamphetamine,amphetamine, 3-4 methylenedioxy-methamphetamine (MDMA), its metabolite methylenedioxy-amphetamine (MDA) and methylenedioxy ethylamphetamine (MDEA). The oral fluid specimens were confirmed for the presence of methamphetamine, amphetamine, MDA and MDA. Hair
métahylandoxide éthylamphétamine (MDEA). Les échantillons de salive ont servi à valider la présence de méthamphétamí-
ne, d’amphétamine, de MDMA et de MDA. Les cheveux ont identifié le plus grand nombre de toxicomanes dans les deux 
populations. Les urines ont permis d’identifier un plus grand 
nombre de sujets positifs que la salive. Bien que l’analyse des 
amphétamines dans les cheveux, la salive et les urines 
aient déjà été publiée, c’est la première étude où les trois 
matrices ont été collectées simultanément sur une population 
toxicomane et analysées en suivant les directives fédérales.

Introduction

Methamphetamine acts as a stimulant, increasing phy-
sical activity and decreasing appetite, thereby contribut-
ing to lack of sleep, and weight loss. Long-term use of 
amphetamines can lead to malnutrition, as well as skin 
disorders, ulcers and vitamin deficiency. Other adverse 
effects include anxiety, violent behavior, hallucina-
tions, addiction, paranoia, mental issues and even 
amphetamine induced psychosis and stroke. Methamphetamine has been the dominant drug in 
Southern California for many years. According to the 
2000 National Household Survey on Drug Abuse, an 
estimated 8.8 million people (4% of the population) 
have tried methamphetamine at some time in their 
lives. Data from the 2000 Drug Abuse Warning 
Network (DAWN), which collects information on 
drug-related episodes from hospital emergency depart-
ments in 21 metropolitan areas, reported that metham-
phetamine-related episodes increased by 30% from 
approximately 10,400 in 1999 to 13,500 in 2000, 
although there had previously been a significant 
decrease in methamphetamine-related episodes reported 
between 1997 (17,200) and 1998 (11,500).

Hair

Human hair grows at approximately 1 -1.3 cm per 
month, and has been measured using various biomar-
kers (1,2). Hair, due to its growth rate and stability, 
offers a much longer history of drug exposure than any 
other matrix. Hair has been increasingly chosen as a 
workplace test specimen, since, in contrast to urine, the 
collection is observed, and the possibility of adulter-
ation or substitution of the specimens is reduced. There 
are many publications regarding the incorporation of 
methamphetamine into hair (3-5). Methamphetamine, 
amphetamine, 3-4 methylenedioxyamphetamíne 
(MDMA), its metabolite methylenedioxyamphetamine 
(MDA) and methylenedioxy ethylamphetamíne 
(MDEA) are incorporated well into the hair shaft and 
have been detected and analyzed using established pro-
cedures (6,7). For the screening of drugs in hair, both 
methanol and aqueous extraction procedures have been 
identified the highest number of drug users in both the admit-
ted using population and those who denied use. Urine identified a higher number of positive subjects than oral fluid. 
While the analysis of amphetamines in hair, oral fluid and 
urine has been previously published, this is the first study 
where all three matrices were collected simultaneously from 
a drug using population, and analyzed according to the 
proposed Federal guidelines.

Materials and Methods

Subjects

The study enrolled 200 subjects, from the Drug and 
Alcohol Recovery Team (DART) in Fullerton, Orange 
County, Southern California. The study was approved 
under Immunalysis Institutional Review Board IRB # 
2004-05-001. All subjects participating in the study 
were made aware of the purpose of the study, and 
signed a consent form. While information on drug use, 
including time of last use, frequency of use, ethnicity,
age, sex and hair color were recorded for each subject, names, addresses or other identifying information was not collected on the interview sheet. Complete anonymity was established during the sample collection and laboratory testing procedures. Each subject provided a urine sample, an oral fluid and a hair specimen taken from the head at the time of interview. Information on drug use, including time of last use, frequency of use, ethnicity, age, sex and hair color were recorded for each subject.

One hundred of the subjects admitted methamphetamine use in the recent past; the others denied drug use. The subjects ranged in age from 21 to 65 years old, and frequency of use ranged from daily to once a month.

**Experimental**

Hair and oral fluid screening kits were obtained from Immunalysis Corporation (Pomona, CA). The Methamphetamine Direct ELISA Kit (Catalog # 211-0480) was used for screening both the hair and oral fluid specimens according to the manufacturer’s instructions. The urine specimens were screened using enzyme immunoassay with the Diagnostic Reagents, Inc. (DRI, Sunnyvale, CA) reagents for amphetamines (Catalog # 0018). For confirmatory procedures, deuterated \( \text{d}_{14} \)-methamphetamine, \( \text{d}_{11} \)-amphetamine, \( \text{d}_{5} \)-MDMA, \( \text{d}_{2} \)-MDA and \( \text{d}_{5} \)-MDEA as internal standards as well as the unlabelled drug standards were obtained from Cerrilliant (Round Rock, TX). Solid phase mixed mode cation exchange - hydrophobic phase extraction columns (ZSDAU020, 200 mg) were obtained from United Chemical Technologies Inc. (Bristol, PA). The derivatizing agents trifluoroacetic anhydride (TFAA) and heptafluorobutyric anhydride (HFBA) were obtained from Pierce Chemical Co. (Rockford, IL). All solvents were HPLC grade or better, and all chemicals were ACS grade. The Toxi-Lab system was purchased from Varian Inc, Laguna Beach, CA.

**Hair**

**Sample preparation**

The hair was cut into small segments (3-5 mm), and an aliquot of 10 mg was weighed. The hair was washed briefly with methanol (2 mL/10 min), the solvent was decanted and the hair was allowed to dry. To the hairs, 0.025 M monobasic phosphate buffer (pH 2.7 with 0.1% BSA; 0.5 mL) was added, the tubes were capped and incubated at 60°C for one hour. 0.5 M dibasic phosphate buffer (pH 9.0; 50 uL) was added to neutralize the acid environment and the liquids were transferred to corresponding clean glass tubes. All specimens, calibrators and controls were then diluted 1:5 by adding 400 uL of phosphate buffer saline (PBS with 0.1% BSA, pH 7.0) to 100 uL of extract.

**Screening Assay**

An aliquot of the diluted hair extract (20 uL), along with Horseradish peroxidase enzyme labeled methamphetamine derivative (100 uL) was added to the individual micro-plate well coated with a methamphetamine specific antibody. After incubation (60 min, room temperature), the micro-plate wells were washed with deionized water (6 x 300 uL). Tetramethyl benzidine (TMB, 100 uL) was added to each well and the plate incubated in the dark for 30 minutes. The reaction was stopped using 1N hydrochloric acid (100 uL) and the plate read at 450 nm with a reference wavelength of 620 nm. The sample size of 20 uL of the diluted hair extract gave good separation at the screening cut-off concentration of 500 pg of methamphetamine equivalents per milligram of hair. A specimen was considered to be presumptively positive if it screened higher than the recommended cut-off of 500 pg/mg.

**Confirmation Assay**

Presumptive positive samples identified using the screening assay were carried forward to confirmation using gas chromatography-mass spectrometry (GC/MS) operating in electron impact mode. The internal standard solution contained methamphetamine - \( \text{d}_{14} \) amphetamine - \( \text{d}_{11} \) MDMA - \( \text{d}_{5} \) MDA - \( \text{d}_{5} \) and MDEA - \( \text{d}_{5} \). Low and high controls were also prepared. A separate aliquot of hair was weighed out (2 - 10 mg) depending on the screening result) and methanol (2 mL) was added. The samples were incubated for 1-5 min at room temperature, then the methanol was discarded. Internal standard (100 uL) was added to each calibrator, control or hair specimen. Methanol (2 mL) was added to the hairs and the tubes were heated at 75°C for 2 hours. The methanol was then transferred to clean glass tubes, and evaporated at 45°C under nitrogen until approximately 50 - 100 uL was remaining. Phosphate buffer (0.1 M, pH 6, 2 mL) was added to each tube. Solid-phase mixed mode extraction columns (CleanScreen DAU) were placed into a vacuum extraction manifold. Each column was conditioned with methanol (3 mL), deionized water (2 mL) and 0.1 M phosphate buffer (pH 6.0; 2 mL). The samples were allowed to flow through the columns using no vacuum. The columns were washed with deionized water (2 mL), 0.1M hydrochloric acid (2 mL) and methanol (3 mL). The columns were allowed to dry at high pressure for 5 minutes. The drugs were finally eluted using fresh methylene chloride : isopropanol : ammonium hydroxide (80:20:1.8; 2 mL). The extracts were evaporated to dryness under nitrogen and reconstituted in heptafluorobutyric anhydride (HFBA, 50 uL) and ethyl
acetate (50 μL). The tubes were heated at 75°C for 15 min, cooled and 50% ammonium hydroxide (0.5 mL) was added. Following mixing, iso-octane (100 μL) was added and the tubes centrifuged (3000 rpm; 5 min). The tubes were frozen in dry ice to freeze the aqueous layer, and the upper organic layer was transferred to autosampler vials for analysis by GC/MS.

**Analytical Procedure (GC/MS)**

An Agilent Technologies 6890 gas chromatograph coupled to a 5973 mass selective detector (MSD) operating in electron impact mode was used for analysis (GC/MS). The gas chromatographic column was 1% phenyl-99% methyl silicone DB-1, 0.20 mm ID, 0.33 μm film thickness, 12 m length and the injection temperature was 200°C. The injection mode was splitless. The oven was programmed from 70°C; ramped at 15 °C/min to 145°C; then ramped at 45°C/min to 280°C. The transfer line was held at 280°C. The ions monitored for each internal standard and drug are shown in Table I.

The limit of quantitation of the procedure was 20 pg/mg for amphetamine and 40 pg/mg for the other analytes. The linearity range was from 20 - 40,000 pg/mg and the precision at the cut-off was less than 3.0% for all analytes. A sample was not considered positive, unless it contained at least 300 pg/mg of methamphetamine, and also contained amphetamine at equal to or greater than 50 pg/mg.

**Table I : Ions monitored in the SIM profile using gas chromatography-mass spectrometric (GC/MS) analysis.**

<table>
<thead>
<tr>
<th>Compound</th>
<th>Quantitation Ion</th>
<th>Qualifying Ions</th>
</tr>
</thead>
<tbody>
<tr>
<td>Methamphetamine</td>
<td>254</td>
<td>210, 118</td>
</tr>
<tr>
<td>Metamphetamine – d₆</td>
<td>261</td>
<td>213</td>
</tr>
<tr>
<td>Amphetamine</td>
<td>240</td>
<td>118, 91</td>
</tr>
<tr>
<td>Amphetamine – d₆</td>
<td>244</td>
<td>128</td>
</tr>
<tr>
<td>MDMA</td>
<td>254</td>
<td>210, 162</td>
</tr>
<tr>
<td>MDMA – d₆</td>
<td>258</td>
<td>213</td>
</tr>
<tr>
<td>MDA</td>
<td>162</td>
<td>240, 375</td>
</tr>
<tr>
<td>MDA – d₆</td>
<td>167</td>
<td>244</td>
</tr>
<tr>
<td>MDEA</td>
<td>268</td>
<td>240, 162</td>
</tr>
<tr>
<td>MDEA – d₆</td>
<td>274</td>
<td>244</td>
</tr>
</tbody>
</table>

**Oral fluid**

**Sample Preparation**

The oral fluid specimens were collected using a Quantisal™ collection device (Immunoanalysis, Pomona CA). When the absorbent collection pad had absorbed 1 mL of neat oral fluid (+10%), a blue dye was visible in the indicator window on the plastic stem of the collection pad. The pad was placed in a polypropylene transport tube containing 3 mL of extraction buffer solution, capped and taken to the laboratory.

**Screening Assay**

An aliquot (10 μL) of the oral fluid + buffer was added to the micro-plate wells for analysis following the protocol described above for the hair specimens, and according to the manufacturer’s instructions in the package insert. A specimen was considered to be presumptively positive if it screened higher than the recommended cut-off of 50 ng/mL of methamphetamine equivalents.

**Confirmation Assay**

Presumptive positive specimens were confirmed using gas chromatography-mass spectrometry (GC/MS). Calibrators and control were prepared in 25% synthetic saliva and 75% Quantisa™ buffer. For the specimens, 1 mL was removed from the Quantisa™ collector. Internal standard (200 μL) containing deuterated d₆-amphetamine and d₆-methamphetamine was added. Low and high controls were included in each confirmation batch. Concentrated ammonium hydroxide (150 μL) and 1-chlorobutane (3 mL) were added to each sample. The specimens were mixed for 10 minutes and then centrifuged. The top layer was transferred to glass screw cap tubes, trifluoroacetic anhydride (TFAA, 100 μL) was added, and the tubes were capped. Following mixing, the samples were heated at 70°C for 15 min and evaporated to dryness. The specimens were reconstituted in ethyl acetate (50 μL) and transferred to auto-sampler vials for analysis using GC/MS. The ions monitored were 144 and 123 for deuterated amphetamine (d₆); 140, 118 and 91 for amphetamine; 161 and 123 for deuterated methamphetamine (d₆); 154, 118 and 110 for methamphetamine. MDMA and MDA were not detected in any of the samples. MDEA was not monitored in the oral fluid specimens.

**Analytical Procedure (GC/MS)**

A Shimadzu QP5000 instrument was used for analysis. The column was a HP1-MS (30 m length x 0.25 mm diameter x 0.25 μm film thickness). The injector was operated in splitless mode and the injection temperature was 200°C. The oven was programmed from 70°C to 180°C at 20°C/min, ramped at 40°C/min to 240°C and held for 1.2 min. The transfer interface temperature was 280°C. The instrument was operated in electron impact mode. The limit of quantitation was 15 ng/mL for amphetamine and methamphetamine; and the upper limit of linearity was 2500 ng/mL for amphetamines; 5000 ng/mL for methamphetamine of neat oral fluid equivalents. A sample was not considered positive, unless it contained at least 50 ng/mL of methamphetamine, and also contained amphetamine at equal to or greater than the limit of detection of the assay.
Urine

The urine specimens were screened using Diagnostic Reagents, Inc. (DRI) EIA assay for amphetamines at a cut-off of 1000 ng/mL. The DRI assays were performed on the Hitachi 717, a fully automated chemistry analyzer. Specimens screening positively were confirmed using the Toxi-Lab thin-layer chromatography system (Toxi-Lab, Inc.) thin-layer chromatography system, for the confirmation of amphetamine and methamphetamine, according to the manufacturer's protocol (14). The detection limits for amphetamines and methamphetamine were 400 and 200 ng/mL respectively.

Results and discussion

Methamphetamine use has become a major problem among many communities, frequently associated with violence among men and women (15). As far back as 1995, Kipke et al. reported that 62% of Los Angeles street youth admitted taking methamphetamine, in fact it was the second most popular drug after marijuana (16). In a study of juvenile arrestees reported in 2002, 11.9% of girls and 9.2% of boys reported using amphetamines, while 12.4% of girls and 8.5% of boys reported the use of crystal methamphetamine specifically. Crystal methamphetamine or “crank” has longer lasting effects and is less expensive than heroin or cocaine. The ethnicity of the subjects was approximately equal through African American, Hispanic and Caucasian juveniles (17). More recently, methamphetamine has been reported as the drug of choice in communities of gay and bisexual men, where it has been associated with high-risk behaviors. It was shown to be a commonly used drug among these groups in New York City where it is often combined with other drugs that may increase its risks and adverse health consequences. The reports indicate that methamphetamine is widely used by men across all age groups, educational level, race and HIV status, but that rates of methamphetamine use can be reduced with methamphetamine dependence treatment (18,19).

In our study, hair and oral fluid specimens were considered positive when they were confirmed using GC/MS procedures at or above the levels dictated by the proposed Federal guidelines (20). The results for the positive specimens are shown in Table II.

Self-reported Drug Users: In the self-reported drug using population, 16 individuals were not positive by any specimen type. Of the remaining 84 subjects found to be positive, 56 (66.6%) were positive via the hair test, 52 (61.9%) using urinalysis and 48 (57.1%) via oral fluid.

Self-reported Non-drug Users: In the self-reported non-drug taking population, 25 of 28 subjects (89.2%) who tested positively had a positive hair test, while only 12 (42.8%) were positive through urinalysis and 7 (25%) were positive via oral fluid. Both urine and oral fluid were only able to identify a low number of methamphetamine users, urine identified twelve subjects; oral fluid identified seven, but hair identified 26 individuals as positive, more than twice as many subjects as urine, and more than three times as many subjects as oral fluid. This is in agreement with other publications, indicating hair identifies more positive drug users than the other specimens tested (21).

Overall: Eighty-eight (88) of the 200 subjects were negative in all sample types, but of the 112 individuals who were positive in at least one matrix, 82 (73.2%) had a positive hair assay, while only 64/112 (57.1%) had a positive urine and 55/112 (49.1%) provided a positive oral fluid test. Hair identified 56% of admitted methamphetamine users; urine detected 52% of users, and oral fluid identified 48% of subjects admitting methamphetamine use. Overall, the hair identified 18 more specimens than urine, and 27 more samples than oral fluid (Figure 1).

Correlation: It is difficult to determine whether a correlation exists between the frequency of dose reported

Table II: Positivity by matrix from a population admitting methamphetamine use, and from subjects denying methamphetamine use.

<table>
<thead>
<tr>
<th>Matrix Type</th>
<th>Self-reported Drug Users</th>
<th>Self-reported Non-Drug Users</th>
<th>Overall</th>
</tr>
</thead>
<tbody>
<tr>
<td>All matrices negative</td>
<td>16</td>
<td>72</td>
<td>88</td>
</tr>
<tr>
<td>All matrices positive</td>
<td>26</td>
<td>5</td>
<td>31</td>
</tr>
<tr>
<td>+ Hair only</td>
<td>20</td>
<td>15</td>
<td>35</td>
</tr>
<tr>
<td>+ Hair and oral fluid</td>
<td>3</td>
<td>1</td>
<td>4</td>
</tr>
<tr>
<td>+ Hair and urine</td>
<td>7</td>
<td>5</td>
<td>12</td>
</tr>
<tr>
<td>+ Urine only</td>
<td>9</td>
<td>1</td>
<td>10</td>
</tr>
<tr>
<td>+ Oral fluid and urine</td>
<td>10</td>
<td>1</td>
<td>11</td>
</tr>
<tr>
<td>+ Oral fluid only</td>
<td>9</td>
<td>0</td>
<td>9</td>
</tr>
<tr>
<td>Total</td>
<td>100</td>
<td>100</td>
<td>200</td>
</tr>
</tbody>
</table>

Figure 1: Number of positive tests in each population (n=200).
by the subjects and the amounts of drug and metabolites detected in the specimens. Variation in the amount of drug, purity of drug, time after sampling, as well as the truthfulness of subjects all affect the outcome. However, within this study group, it is clear that hair is the best specimen to test in order to identify the highest number of drug users, whether or not self-reported. In general, urine identified a higher number of drug users than oral fluid in both populations.

**Hair Color and Ethnicity:** The hair color and ethnicity of the subjects was recorded at interview. The state of the art in hair testing is that it is most likely the melanin content of hair is critical in determining the basic drug content in hair, not ethnicity. In our subjects, the predominant hair color was black across all ethnic types (55%). Dark brown and gray accounted for approximately 20% each of the subjects, with only a few subjects having red, blonde, or light brown hair. Since there was little variation in hair color and melanin content, it was difficult to form any conclusions on the effect of pigmentation on methamphetamine incorporation.

The ethnicity of the subjects also had little effect on the identification of methamphetamine users. Hair identified a much higher number of drug users than either oral fluid or urine across all ethnicities. A breakdown of the demographics from the subjects testing positively showed that males accounted for 66/112 (58.9%) of the positive subjects; females for 41.1%.

Whites accounted for the highest number of positives in all specimen types, with 43 of 82 hair specimens (52.4%) being positive, 34 of 64 (53.1%) of urines and 33 of 55 (60%) of oral fluids. African American subjects accounted for 18 of 82 (21.9%) of all hair positives, 10/64 (15.6%) of urines and 5/55 (9%) of oral fluid samples. In the Hispanic population, all three matrices were similar in detection rate. Hair accounted for 11 of 82 (13.4%), urine for 9/64 (14%) and oral fluid for 9/55 (16.3%) of the total positive samples. Finally, those describing themselves as "other" accounted for 12.1% of the hair positives, 17.1% of the urine positives, and 14.5% of the oral fluid positive samples (Figure 2).

**Concentrations detected**

**Hair:** The range of concentrations detected in hair was extremely wide, with both the mean and median values being substantially higher than the proposed cut-offs for regulatory samples in both populations (0.3 ng/mg). Among the self-reported drug users, the mean methamphetamine level was 13.8 ng/mg and the median was 7.8 ng/mg. For amphetamine, the levels were 1.0 ng/mg for the mean, and 0.53 ng/mg for the median.

In the population reporting no use of methamphetamine, the average levels were lower than in the self-reported population. The mean methamphetamine level was 11 ng/mg; median value was 6.8 ng/mg. The mean amphetamine level was 0.72 ng/mg; median concentration 0.35 ng/mg.

Figure 3 shows the extracted ion chromatogram of a hair specimen taken from a subject admitting to methamphetamine use twice a week. The system would only allow 6 extracted ion chromatograms to be shown on one page, but the other ions were collected for each sample and used in the determination of positivity according to accepted GC/MS validation protocols. The hair contained 8.8 ng/mg of methamphetamine and 0.24 ng/mg of amphetamine.

**Oral fluid and Urine:** In our study, the positive rates for oral fluid and urine were similar within each population of admitted drug users and the non-drug using subjects. The range of concentrations measured in oral fluid was from the low nanograms per milliliter (ng/mL) to over 5000 ng/mL of methamphetamine and over 500 ng/mL of amphetamine. Obviously the timing of collection following recent methamphetamine use is a factor in
the level detected. As expected, the highest levels of methamphetamine were detected in subjects admitting to very recent methamphetamine use.

Four individuals had methamphetamine values of over 5000 ng/mL with associated high amphetamine levels. All admitted to using methamphetamine in the recent past. Three of the four had used on the day of collection, but one stated his last use was three days before collection and he only used once a month. The corresponding hair results from the other subjects are shown in Table III. No specimens contained MDMA and MDA, and no samples showed the presence of amphetamine alone.

Figure 4 shows the chromatogram obtained from the analysis of the oral fluid of a subject admitting to daily methamphetamine use. The amphetamine concentration was 108 ng/mL and methamphetamine level was 233 ng/mL. His urine sample was also positive, and his hair tested positively at concentrations of 2.4 ng/mg and 0.33 ng/mg for methamphetamine and amphetamine respectively.

**Acknowledgements**

We are grateful to Ms. Toby Evans for facilitating subject access at the Drug and Alcohol Recovery Team (DART) and to Russell Munford (Immunalysis Corporation) for the collection of all the specimens. Many thanks are due to Michelle Nguyen and Erma Abolencia for all the immunoassay testing of oral fluid and hair samples at Immunalysis Corporation.

**Table III : Results of hair specimens from subjects having greater than 5000 ng/mL of methamphetamine in their oral fluid samples.**

<table>
<thead>
<tr>
<th>Sample ID</th>
<th>Drug Use</th>
<th>Oral Fluid (ng/ml)</th>
<th>Hair (ng/mg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>57</td>
<td>1 x month</td>
<td>&gt;5000</td>
<td>Methamphetamine</td>
</tr>
<tr>
<td>18</td>
<td>3 x week</td>
<td>&gt;5000</td>
<td>Not detected</td>
</tr>
<tr>
<td>33</td>
<td>2 x week</td>
<td>&gt;5000</td>
<td>32.6</td>
</tr>
<tr>
<td>40</td>
<td>2 x day</td>
<td>&gt;5000</td>
<td>3.9</td>
</tr>
</tbody>
</table>

**Figure 4 : Oral fluid specimen from subject ID #32**

a. D5-amphetamine
b. Amphetamine (108 ng/mL)
c. D6-methamphetamine
d. Methamphetamine (233 ng/mL).
References


