A crossroad between Criminalistics and Forensic Toxicology

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Citation

Abstract
Biological stains found at crime scene, are basically intended for DNA identification. Toxicology, however, can usually supply additional information. For example, the possible use of toxic materials to force the victim of some crime to yield. Some of the difficulties of analysing stains from a crime scene, involve differentiating between older or more recent stains, varying environmental conditions, and support material. This article presents the preliminary results of using Quick Kits for detecting toxic materials in urine stains. The results show that it is feasible to use analysis kits, to detect both old and more recent urine stains, made on porous surfaces. Ongoing studies should be carried out, with the same procedure being applied to stains from distinct biological fluids, both old and new, on different supports. This would help to create a crossroads between toxicology and criminalistics, and provide new tools to help forensic investigators solve criminal cases.

INTRODUCTION
As a Forensic Science, Toxicology is often of use in detecting toxic substances in samples of food or beverages found at a crime scene. However, similar tests can also be performed to detect drugs in biological fluid samples - mostly blood and urine- taken from either, victims or suspects [1] [2] [3] [4].

The examination of urine stains in criminalistics, is mainly for identification purposes, using DNA analysis [5] [6] [7]. However, in other situations of legal conflict, urine stains can be used to establish other circumstances that may have occurred. One of these is the possible use of toxic materials to force the victim of some crime to yield. In this case, detection of these products, on various surfaces, could explain the existence and duration of that fact.

To take just five examples:

1.- Cases of mistreated children or elderly people:
The detection of urine stains on sheets, blankets or mattresses, together with determining the toxic substances present, could be of great usefulness.
The same may be said of such detection on nappies, sanitary towels or other hygienic materials.

2.- In "drug facilitated sexual assault" (DFSA), following the discovery of the scene of the crime, the detection of a drug in urine stains found could provide all-important evidence of a crime.

Traces could be found on surfaces such as sheets or vehicle seats, among others. Finding them on these surfaces is of great importance due to the short average lifespan of the toxic substances that are usually employed.

3.- In kidnapping cases, the use of these substances to weaken the resistance of the victim could also be determined.

4.- In the case of missing persons, and their discovery some time after, when it is no longer possible to undertake another type of examination, the detection of urine stains in underwear could help to clarify how the events took place.

5.- Finally, in other cases of sexual aggression with development of paraphilia behaviour - such as urophilia or undinism - the non-detection of toxic substances in the urine stains found on the victim, would be of great value. This could also apply in other crimes such as robbery, burglary, etc. to stains found at the scene of the crime.

In the first four cases, it could be determined that, at the moment of the crime, the victim was under the effect of drugs, without any possibility of defending him- or herself.

In the fifth case, it could help to eliminate a possible
circumstance put forward by the defence in order to diminish responsibility.

As a result of the above, the crossroads between Toxicology and Criminalistics creates an interesting field of study: the toxicological analysis of biological stains taken from the scene of the crime creates a new area for study: the toxicological analysis of biological stains from a crime scene.

In previous works, some investigators have demonstrated that certain drugs, can be detected by means of the instrumental analysis method (GC-MS), using blood stains generated in the lab [8] [9] [10] [11] [12].

However, in addition to instrumental analysis methods, Quick Kits are currently available, which can be used to determine the presence of certain drugs in urine or blood. These Kits can be used to analyse drugs in liquid samples extracted from living or cadaveric tissue, provided the samples are well preserved. This method has also proven to be just as efficient using stains on paper, obtained from liquid samples [13], provided they are properly preserved.

There is, however, no current information on the evaluation of these methods, using stains derived from a crime scene. The difficulties involved are significant, bearing in mind the particular conditions of crime scenes, which may be quite old and are definitely not preserved in any way. Moreover, stains may be found on a variety of support materials, from porous to non-porous, making the process even more complicated. The bibliography on toxicological stain analysis in such settings is limited and often outdated [14]

Consequently, this study, undertaken by our investigation team, involved designing a method for evaluating the efficacy of the Quick Kits currently used for detecting the presence of drugs in recent or old drug biological fluid stains, found in varying environmental conditions and on different types of support materials, in simulation of samples derived from an actual crime scene. This article contains data obtained from our preliminary experiments*, which were carried out using the procedure described below.

**MATERIAL AND METHOD**

**MATERIAL**
Urine stains support material (filter paper).

**REAGENTS**
Quick Kit for drug abuse determination: Tox/SeeTM Multi-

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Drug Screen Panel, from Bio-Rad.

**Kit characteristics:**
Qualitative immunoassay test for detecting phencyclidine, barbiturates, amphetamine, cocaine and benzoylecgonine, methamphetamine, tricyclic antidepressants, morphine, cannabinoids and benzodiazepine in urine.

Cut-off levels are as follows:
- phencyclidine: 25 ng/ml
- barbiturates (secobarbital): 300 ng/ml
- amphetamine: 1000 ng/ml
- methamphetamine: 1000 ng/ml
- benzoylecgonine: 300 ng/ml
- tricyclic antidepressants (amitriptyline): 1000 ng/ml
- morphine: 300 ng/ml
- cannabinoids: 50 ng/ml
- benzodiazepines (secobarbital): 300 ng/ml

**METHOD**
So that this preliminary study could evaluate the feasibility of the kits described above in determining drugs in biological fluid stains derived from a crime scene, a simple method was designed, focusing on urine samples.

For the purposes of this study, urine samples submitted to the Forensic Toxicological laboratory of the Legal Medicine Institute of Alicante, Spain, were used. In total, 14 samples were analysed.

The samples were collected by the investigators from the Legal Medicine Institute, and stored in suitable containers. During transport a suitably low temperature was maintained. No preservatives were added.

The method can be outlined as follows:

a. Initial determination of drugs in each of the samples received from the lab: this was undertaken immediately on receiving the sample. A drug abuse determination Quick Kit was used: Tox/SeeTM Multi-Drug Screen Panel, from Bio-Rad.

b. From each of the 14 samples received, urine stains were created on filter paper. Each stain contained five drops of
urine. The stains were left to dry and kept on a lab table, at room temperature without protection.

For the negative control, stains were formed from urine samples that provided no positive results following analysis using the Tox/SeeTM Multi-Drug Screen Panel test.

c. At intervals of 7, 15 and 30 days, the stains were tested using the Quick Kit (Tox/SeeTM Multi-Drug Screen Panel, from Bio-Rad) to determine possible drug abuse. At each interval—to confirm results—10 stains from each of the samples were analysed. Before obtaining readouts from the Kit, a few drops of saline solution were applied over the stains.

The same procedure was followed with the negative control stains.

RESULTS
The results are show in table 1:

<table>
<thead>
<tr>
<th>Drug</th>
<th>Time (days)</th>
<th>Results</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>0</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>7</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>15</td>
<td>+</td>
</tr>
<tr>
<td></td>
<td>30</td>
<td>+</td>
</tr>
</tbody>
</table>

The positive result of the test for a certain substance has been marked with the sign “+”, a negative result with “-”.

Time 0 (zero) refers to the results of the analysis described in point “a” of the method section (i.e. the tests that are carried out on the sample when received).

The shaded boxes show cases in which the initial test (at zero time) gave a negative result. Although it is logical that the same negative result would later be obtained from the stains on these samples, this has been checked by employing the test.

In all cases, the necessary negative controls were undertaken.

DISCUSSION
The following points can be made from the results obtained:

- On analysis for Benzodiazepines:

With the exception of sample 5, in all cases where the zero test gave a positive result, the presence of benzodiazepines could be detected in 15- and/or 30- day old stains.

Sample 3 provided negative results for 7 and 15 days, whereas a positive result was obtained at 30 days. This may be due to false negatives resulting from some error in analysis or stain preparation.

- On analysis for Cocaine:

With the exception of sample 8 (for which positive results were obtained only for the first 7 days), in all samples where the test at zero time gave a positive result, it was possible to detect the presence of cocaine in 30-day old stains.

As in the benzodiazepines study, there was one sample – no. 2- that provided negative results for 7 and 15 days, whereas a positive one was obtained at 30 days. Once again, this can be considered the result of false negatives resulting from the same causes explained above.

- On analysis for Methadone:

In all the samples where the test at zero time gave a positive result, it was possible to detect the presence of methadone in 30-day old stains.

- On analysis for Cannabis:

Here, the results show that the Kit is not effective in determining this substance in most prepared stains. In only two samples (of the eleven in which cannabis was detected at zero time) was a positive result obtained for 7-15 day old stains.

- On analysis for Opiates:

The results for opiates can be qualified as irregular, although in all cases it was possible to detect the substance in stains of a greater or lesser age.

In conclusion, the results of this preliminary work indicate that it is feasible to use toxic material analysis kits, on either
older or more recent biological fluid stains. The study should consequently be continued, with the same procedure being applied to stains formed by various biological fluids, both old and new, and on varying supports, both porous and non-porous.

This would help to form a crossroads between toxicology and criminalistics, and possibly provide a new tool for the use of forensic investigators, in the solving of criminal cases.

References
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