Analysis of opiates in fly larvae sampled on a putrefied cadaver

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Toxicological analyses on a putrefied cadaver are sometimes difficult to achieve, because of the absence of blood and/or urine. Drugs present in a decomposing corpse may be identified through analysis of maggots feeding off it. In this study, morphine and codeine were simultaneously identified and assayed in blood and bile of a putrefied cadaver and in the fly larvae of Calliphoridae found on the corpse.

Des analyses toxicologiques sur des cadavres putréfiés sont parfois difficiles à cause de l'absence de sang et/ou d'urine. Les drogues présentes dans un corps en décomposition peuvent être identifiées par l'analyse des vers qui s'en nourrissent. Dans cette étude la morphine et la codéine ont été identifiées simultanément et mesurées dans le sang et la bile d'un cadavre putréfié et dans la larve d'une mouche Calliphoridae dans le cadavre.

Key Words: Drugs of abuse; Larvae; Putrefaction; Opiates: Entomology.
Introduction
The use of insect larvae is an aid in the establishment of the post-mortem interval and therefore is a well-recognized procedure in the performance of forensic autopsies [1]. In decomposing bodies, toxicological investigations are sometimes difficult to achieve owing to the absence of body fluids or tissues, or their severe contamination by putrefactive compounds. In 1980, Bayer et al. proposed the identification of drug poisoning in decomposing corpses through the analysis of maggots feeding on the body [2]. They commented that the drug present in larvae could only have originated from the tissues upon which they were feeding, as there was no valid data on any metabolic mechanisms of the larvae. Since this first report, there have only been a few others involving drug analysis in maggots and these are insufficient to establish a data base [3–10]. It was suggested by Pounder in 1991 [11] that systematic data of cases involving entomo-toxicology should be collected, and the present report is presented for this purpose.

Case report
On 12 June 1992, the cadaver of a man known to be a chronic heroin abuser was found lying in his kitchen, with a spoon and a syringe containing traces of heroin, on the table. The body was completely putrefied, with large amounts of exudative body fluids on the floor. The corpse was covered with identical larvae of flies belonging to the Calliphoridae family.

The subject had been dead for 10 days. At the autopsy, radiography did not disclose the presence of bullets. Post-mortem specimens taken included 1.6 ml femoral blood and 4.8 ml bile. No urine could be taken, since the bladder was totally empty. Samples of living fly larvae were removed from different places on the corpse and pooled.

Method
Larvae (101.3 mg/larvae, n = 20), about 2 cm long, were copiously washed with deionized water, to prevent contamination by human fluids (exudation and transudation liquids), dried and stored at 4°C. Before toxicological analysis, they were washed again and dried with a filter paper and then homogenized in a 9% saline solution using a Potter homogenizer. No other pre-treatment, such as digestion, was necessary.

The larvae homogenate was extracted and analyzed as for human samples, using a technique for opiates which had been described previously by Kintz et al. [12]. Briefly after enzymatic hydrolysis (β-glucuronidase), samples were extracted with 5 ml of chloroform-isopropanol-n-heptane (50:17:23, v/v) mixture in the presence of 1 ml phosphate buffer (pH 9-2) and levallorphan (10 mg/l, internal standard), extracted into an acidic solution and back extracted into an organic solvent. After evaporation of the solvent, and derivatization with BSTFA (bis-(trimethylsilyl)trifluoroacetamide) containing 1% TMCS (trimethyl-chlorosilane), the drugs were separated on a 12 m BP 5 capillary column, identified by mass spectrometry, using a Perkin Elmer ion trap detector, and quantified by selected ion monitoring.

### TABLE 1 Concentrations of morphine and codeine in the autopsy samples

<table>
<thead>
<tr>
<th>Samples</th>
<th>Morphine (µg/l or µg/kg)</th>
<th>Codeine (µg/l or µg/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Femoral blood</td>
<td>168</td>
<td>37</td>
</tr>
<tr>
<td>Bile</td>
<td>357</td>
<td>88</td>
</tr>
<tr>
<td>Larvae</td>
<td>90</td>
<td>12</td>
</tr>
</tbody>
</table>

Results and discussion
The results of the opiate analysis of the autopsy specimens are presented in Table 1. Total codeine and morphine were detected in the larvae, indicating that even with a decomposing cadaver, toxicological investigations are possible, given the presence of larvae. At autopsy, larvae sampling is easy, and their subsequent preparation and extraction are the same as with any human tissue.

References


