## **CASE REPORT**

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# Fly Larvae: A New Toxicological Method of Investigation in Forensic Medicine

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**ABSTRACT:** Toxicological analyses on a putrefied cadaver are sometimes difficult to achieve because of the absence of blood and urine. In this study, maggots, living material, are proposed as a new medium of investigation in forensic medicine. Five drugs (triazolam, oxazepam, phenobarbital, alimemazine, and clomipramine) were identified and assayed in some tissues of a putrefied cadaver and in the maggots found on and in the body.

KEYWORDS: toxicology, larvae, putrefaction

When a cadaver undergoes putrefaction in the open air, it rapidly becomes the target of necrophage insects. Flies represent the earliest arrivals of the decomposer community colonizing corpses. They are usually called "first wave."

Quite soon after death (one to two days), depending on the outdoor temperature, the eggs of flies are present, especially on the head.

Calliphorid larvae are the most dominant component of this community and are usually present in large amounts, facilitating their sampling [1].

These larvae were used in forensic medicine many years ago for evaluating the time of death and the geographical place of death.

The taxonomy of larvae [2], their growth rate [3,4], and their metamorphic development [5] are typical parameters used to determine how long ago the death occurred. Moreover, Nuorteva and Hasanen have discussed the possibility of using emerging flies to determine the pollutant content, such as mercury, of a corpse, as blowflies have the ability to accumulate such pollutants and are, therefore, more sensitive to such a test [6].

At the autopsy of a putrefied cadaver, it is not always possible to obtain blood and urine samples. This problem has to be taken into account when drug poisoning is suspected.

Therefore, we present a case in which five drugs were simultaneously identified in

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human organs and in larvae. The present study deals with the usefulness of the toxicological investigations in maggots in an unusual case.

The examination of insect larvae from decomposed bodies for qualitative identification of drugs was first proposed by Beyer et al. in 1980 [7]. These authors have determinated phenobarbital in Calliphoridae larvae.

## **Case History**

On 23 Dec. 1987, the cadaver of a man was found lying in his home. He had been dead since 17 Oct. (=67 days).

At the autopsy, we identified a 49-year-old white male, 1.94 m tall, who was completely putrefied. The entire corpse was covered with hundreds of identical Calliphorid fly larvae. Organs were identifiable in spite of their partial decomposition. Brain and testicles were not found.

Postmortem specimens taken included heart, liver, lung, spleen, kidney, and bile. Samples of living maggots were removed from different places of the corpse and pooled in unique sampling.

## Larvae Preparation

Larvae (73.6 mg/larva, n = 100), about 2 cm long, were copiously washed with deionized water, dried, then stored at 4°C.

Before the toxicological analyses, they were again washed and dried with filter paper. Then, they were homogenized in a 9% saline solution by a Potter-Elvehjem homogenizer.

The multiple-washing treatment was necessary to prevent contamination by human fluids (exsudation and transudation liquids).

Larvae samples were analyzed in the same manner as human tissues.

## **Toxicological Analysis**

An initial screening, performed on an Abbott TDx®, indicated the presence of benzodiazepines, barbiturates, phenothiazines, and antidepressants.

Therefore, bile and tissue samples, including larvae preparations, were analyzed with a previously described high performance liquid chromatographic (HPLC) procedure for triazolam [8], oxazepam [9], phenobarbital [10], clomipramine [11], and alimemazine [12] on a Waters automatic system.

### **Results and Discussion**

The results of the five-drug analysis of the autopsy specimens are presented in Table 1. In all cases, drugs were found in the larvae. No amphetamines, cocaine, opiates, ethanol, or salicylates were detected.

To avoid the possible contamination of the larvae extractions by the other extractions, the former samples were injected, twice, at the beginning and at the end of the automatic session. In all cases, the results did not differ significantly from one injection to the other.

The use of particularly sensitive methods has permitted the assay of chemical compounds in larvae. The preparation and extraction from larvae are the same as those from any other human tissue. At autopsy, sampling is easy. Moreover, during the extraction step no emulsion was noted, which is not always the case with human tissue. Chromatograms obtained after larvae extraction have presented less endogenous peaks than the other organs. This is particularly useful in case of putrefactive materials.

Since one substance out of the four classes of psychotropes, that is, barbiturates,

Tissue	Triazolam	Oxazepam	Phenobarbital	Alimemazine	Clomipramine
Bile	894 μg/L	809 µg/L	2447 μg/L	24 μg/L	59 μg/L
Heart	398 ng/g	1317 ng/g	1391 ng/g	318 ng/g	2479 ng/g
Liver	490 ng/g	403 ng/g	3630 ng/g	368 ng/g	433 ng/g
Lung	173 ng/g	1641 ng/g	1233 ng/g	344 ng/g	455 ng/g
Spleen		777 ng/g	3641 ng/g	210 ng/g	58 ng/g
Kidney	, a	286 ng/g	1439 ng/g	66 ng/g	327 ng/g
Larvae	204 ng/g	153 ng/g	761 ng/g	22 ng/g	28 ng/g

TABLE 1—Concentrations of the five drugs identified in the autopsy samples.

benzodiazepines, neuroleptics, and antidepressants (which are the most commonly used to commit suicide) has been identified in the larvae preparations, one can reasonably suppose that other drugs from these classes could be screened in other larvae. Thus, the applications of the toxicological investigations in maggots will surely increase.

It was not possible to establish a correlation between the drug concentrations in the larvae and the human tissue. For example, the ratio varied from 1 to 5 in the case of bile and from 2.4 to 16.7 in the case of liver (Fig. 1). This could be explained by the different tropisms of drugs with respect to target organs (clomipramine has a significant cardiac tropism).

#### Conclusion

This study has indicated that even in the absence of blood and urine, toxicological investigations are possible, given the presence of fly larvae. The usefulness of this new method is evident in cases involving putrefactive materials. Moreover, these analyses can be performed on living materials, which are always more suitable for toxicological screenings.

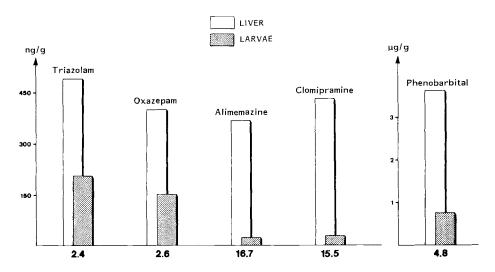


FIG. 1—Liver/larvae ratios for the five drugs identified.

aInterferences in the assay.

#### References

- [1] Payne, J. A., "A Summer Carrion Survey of the Baby Pig sus scrofa Linnaeus," Ecology, Vol. 46, No. 5, 1965, pp. 592–602.
- [2] Mégnin, P., "La faune des cadavres. Application de l'entomologie à la Médecine Légale," Encyclopédie des Sciences-Aide-Mémoire, Paris, 1894.
- [3] Williams, H., "A Model for the Aging of Fly Larvae in Forensic Entomology," Forensic Science International, Vol. 25, No. 3, 1984, pp. 191-199.
- [4] Reiter, C., "Growth Behavior of the Blue Blowfly Calliphora Vicina Maggots," Zeitschrift für Rechtsmedizin, Vol. 91, 1984, pp. 295-308.
- [5] Erzinçlioglu, Y., "The Application of Entomology to Forensic Medicine," Medicine, Science and the Law, Vol. 23, No. 1, 1983, pp. 57-63.
- [6] Nuorteva, P. and Hasanen, E., "Transfer of Mercury from Fishes to Sarcosaprophagous Flies," Annals Zoologici Fennici, Vol. 9, No. 1, 1972, pp. 23-27.
  [7] Beyer, J. C., Enos, W. F., and Stajic, M., "Drug Identification Through Analysis of Maggots," Journal of Forensic Sciences, Vol. 25, No. 2, April 1980, pp. 411-412.
- [8] Adams, W., "Specific and Sensitive High Performance Liquid Chromatography Determination of Alprazolam or Triazolam," Analytical Letters, Vol. 12, No. 6, 1979, pp. 657-671.
- [9] Brodie, R., Chausseaud, L., and Taylor, J., "High Performance Liquid Chromatography Determination of Benzodiazepines in Human Plasma," Journal of Chromatography, Vol. 150, No. 2, 1978, pp. 361-366.
- [10] Mangin, P., Lugnier, A. A., and Chaumont, A. J., "A Polyvalent Method Using HPLC for Screening and Quantification of 12 Common Barbiturates in Various Biological Materials," Journal of Analytical Toxicology, Vol. 11, No. 1, 1987, pp. 27-30.
- [11] Tricyclic antidepressants from serum, Water Clinical Application, Nov. 1981.
- [12] Midha, K., Cooper, J., McGilveray, I., Butterfield, A., and Hubbard, J., "High Performance Liquid Chromatographic Assay for Nanogram Determination of Chlorpromazine and Its Comparison with a Radioimmunoassay," Journal of Pharmaceutical Science, Vol. 70, No. 9, 1981, pp. 1043-1046.

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