

TECHNICAL NOTE

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Analysis of *N,N*-Diethyl-*m*-Toluamide (DEET) in Human Postmortem Specimens

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ABSTRACT: *N,N*-diethyl-*m*-toluamide (DEET) levels in postmortem specimens of stomach and contents, blood, liver, and urine are reported following ingestion of the compound. DEET was analyzed by gas chromatography with an OV-101 column and a nitrogen phosphorus detector. The presence of the compound in the four postmortem specimens was confirmed by mass spectrometry.

KEYWORDS: toxicology, *N,N*-diethyl-*m*-toluamide (DEET), tissues (biology), chemical analysis, gas chromatography, mass spectrometry

N,N-diethyl-*m*-toluamide, commonly known as DEET, is a widely used insect repellent. The acute oral LD₅₀ for rats has been found to be 2000 mg/kg [1], and its ingestion can cause central nervous system disturbances [2]. The study of the pharmacology and toxicology of DEET has been mainly restricted to animal subjects [1,3,4], but in 1979, Wu et al reported on the characterization of the urinary metabolites of DEET in a human volunteer after topical application [5]. There have been at least six cases in the last five years of brain disorders associated with the use of DEET [6-8]. Two of these cases were fatalities in which the subjects were said to have suffered from a "toxic encephalopathy" associated with the use of DEET [6,7].

This laboratory recently received postmortem specimens from the body of an 18-year-old woman who was killed by an oncoming train. Screening tests on the liver specimen revealed the presence of DEET (Kovat's Index 1540). Further analysis showed that the deceased had ingested the compound and that no other poisons or drugs were found.

DEET has been reported to produce toxic effects, but there is a lack of data in the literature on human tissue levels following ingestion. The levels of DEET found in the postmortem specimens of the abovementioned case are therefore reported here.

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Experimental Procedures

Materials

All chemicals and solvents were of analytical grade. DEET was obtained from Sterling Pharmaceuticals, Sydney, Australia. The purity was found to be 98.6% using a British Pharmacopoeia (BP) method [9]. A stock standard of 1 mg/mL in hexane was diluted with hexane to give a 0.1-mg/mL quantitative solution.

The phosphate buffer (pH 9) was 14.2 g of disodium hydrogen phosphate in 100 mL of distilled water.

Instrumentation

The gas chromatograph (GC) was a Hewlett-Packard 5730A equipped with an auto sampler and coupled to a nitrogen-phosphorus detector, Model 18789A. The GC column used was a 2.0-m by 1-mm inside diameter glass column packed with 3% OV-101 on Chromosorb-WHP, 100–120 mesh. The carrier gas was nitrogen at a flow of 30 mL/min. The temperature settings of the GC were: injector 250°C, detector 300°C, and oven 160°C.

The gas chromatograph/mass spectrometer (GC/MS) was a Hewlett-Packard 5985A fitted with a 25-m by 0.31-mm fused silica capillary column coated with a cross-linked methyl silicon phase. The carrier gas was helium at a flow of 2 mL/min. The oven temperature was 100°C initially for 1 min, with a 16°C/min rise to 270°C. The electron impact mode was used.

Extraction Procedure

Tissues

Ten grams of minced tissue were homogenized with thirty millilitres of distilled water. Five millilitres of 10M hydrochloric acid were added, mixed, and filtered. The filtrate was made alkaline by the addition of 5 mL of 60% potassium hydroxide and then transferred to a 50-mL screw cap glass centrifuge tube. Five millilitres of hexane were added and contents gently shaken for two minutes. The mixture was centrifuged for 5 min at 2000 rpm. An aliquot of the hexane was then directly injected into the GC.

Blood and Urine

To a 5-mL specimen, in a 15-mL screw capped glass centrifuge tube, 1 mL of pH 9 buffer was added. The solution was vortexed and 1 mL of hexane added. The contents were extracted by gently shaking for 2 min. The mixture was centrifuged for 5 min at 2000 rpm. An aliquot of the hexane was then directly injected into the GC.

Results and Discussion

Quantitation of the specimens was by peak height relative to the working standard. The experimental results are shown in Table 1. Recovery experiments were performed on demineralized water and blank postmortem tissue specimens by spiking them with known amounts of DEET. The recovery levels and the corrected experimental results for the case are also shown in Table 1.

Although these recovery levels were not high, when DEET was extracted from demineralized water (pH 13) into hexane a recovery of 90% was obtained. Hexane was chosen as the extraction solvent as it gave clean dry extracts which were amenable to direct injection using a nitrogen phosphorus detector.

TABLE 1—Recoveries and levels of *N,N*-diethyl-*m*-toluamide (DEET) found in postmortem specimens.

Specimen	DEET (Found)	% Recovery	DEET Levels Corrected for Recovery
Stomach and contents	460 mg	60	767 mg
Liver	23 mg/kg	45	51 mg/kg
Blood	2.7 mg/L	40	6.8 mg/L
Urine	0.3 mg/L	70	0.4 mg/L

A mass spectrometric analysis confirmed the presence of DEET (molecular ion of $M^+ 191$) in all four postmortem extracts.

References

- [1] Ambrose, A. M., "Pharmacologic and Toxicologic Studies on *N,N*-Diethyltoluamide," *Toxicology*, Vol. 1, 1959, pp. 97-115.
- [2] *The Merck Index*, 10th ed., Merck and Co., Inc., Rahway, NJ, 1983, pp. 412:2838.
- [3] Blomquist, L., Stroman, L., and Thorsell, W., "Distribution and Fate of the Insect Repellent ^{14}C -*N,N*-Diethyl-*m*-Toluamide in the Animal Body. I. Distribution and Excretion after Injection into Mice," *Acta Pharmacologica Et Toxicologica*, Vol. 37, 1975, pp. 121-133.
- [4] Blomquist, L. and Thorsell, L., "Distribution and Fate of the Insect Repellent ^{14}C -*N,N*-Diethyl-*m*-Toluamide in the Animal Body. II. Distribution and Excretion after Cutaneous Application," *Acta Pharmacologica Et Toxicologica*, Vol. 41, 1977, pp. 235-243.
- [5] Wu, A., Pearson, M. L., Shekoski, D. L., Soto, R. J., and Stewart, R. D., "High Resolution Gas Chromatography/Mass Spectrometric Characterization of Urinary Metabolites of *N,N*-Diethyl-*m*-Toluamide (DEET) in Man," *Journal of High Resolution Chromatography and Chromatography Communications*, Vol. 2, Sept. 1979, pp. 558-562.
- [6] Zadikoff, C. M., "Toxic Encephalopathy Associated with the Use of Insect Repellent," *The Journal of Pediatrics*, Vol. 95, No. 1, July 1979, pp. 140-142.
- [7] Heick, H. M. C., Shipman, R. T., Norman, M. G., and James, W., "Reye-Like Syndrome Associated with Use of Insect Repellent in a Presumed Heterozygote for Ornithine Carbamoyl Transferase Deficiency," *The Journal of Pediatrics*, Vol. 97, No. 3, Sept. 1980, pp. 471-473.
- [8] Tenenbein, M., "Toxic Encephalopathy Due to Insect Repellent Ingestion," *Veterinary and Human Toxicology*, Vol. 23, No. 5, 1981, p. 363.
- [9] *British Pharmacopoeia*, Vol. II, Her Majesty's Stationary Office, London, 1980, Method I, Appendix V III H, p. A93.

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