Available online at www.sciencedirect.com
SCIENCE DIRECT®
Forensic Science International 149 (2005) 167–170
Forensic Science International
www.elsevier.com/locate/forsciint

A fatal poisoning caused by methomyl and nicotine

Fumio Moriya*, Yoshiaki Hashimoto

Department of Legal Medicine, Kochi Medical School, Kochi University, Kohasu, Oko-cho, Nankoku City, Kochi 783-8505, Japan

Received 24 March 2004; received in revised form 8 June 2004; accepted 9 June 2004
Available online 25 August 2004

Abstract

A 35-year-old male was found lying in a prone position in his room. He was in cardiopulmonary arrest on arrival to hospital and was pronounced dead. There was no attempt at resuscitation. No miosis was observed on admission. At post-mortem his stomach contained 170 g greenish liquid with a small amount of shredded tobacco leaves. The serum cholinesterase activities were 47–90 IU (normal range for male: 200–440 IU). GC and GC–MS analyses showed nicotine (21.8 mg), methomyl (304 mg), and triazolam (1.69 mg) in his stomach. He had consumed tobacco leaves, Lannate™ containing water soluble methomyl (45%), and Halcion™ tablets containing 0.25 mg triazolam. Methomyl concentrations in blood were 3–8 ng/ml. Substantial amounts of methomyl (2260–2680 ng/ml) were detected in cerebrospinal fluid and vitreous humor. Nicotine concentrations in blood ranged from 222 to 733 ng/ml. A small amount of triazolam was detected only in bile (176 ng/ml) and liver (23 ng/g). The cause of death was respiratory paralysis produced by the additive effects of methomyl and nicotine shortly after consumption.

Keywords: Forensic toxicology; Fatal poisoning; Methomyl; Nicotine; Pharmacological interaction

1. Introduction

About 50–100 fatalities a year occur in Japan from carbamate pesticides and most involve methomyl [1]. Carbamate and organophosphate pesticides inhibit acetylcholinesterase and cause an accumulation of acetylcholine at synapses that stimulates parasympathetic autonomic postganglionic nerve fibers (muscarinic receptors) and somatic motor nerve fibers (nicotinic receptors). Symptoms of poisoning include excessive salivation, accelerated excretion in the respiratory tract, seizures, and even death due to paralysis of the respiratory muscles [2]. In general, carbamate pesticides are less toxic than organophosphate pesticides because they do not bind as strongly to acetylcholinesterase [2]. In addition, carbamates are metabolized rapidly and they do not accumulate in organs and produce damage [3].

Nicotine is a water-soluble alkaloid that is contained in tobacco leaves, at concentrations of 0.5–8.0%, and causes severe dependence [4]. A cigarette contains 7–24 mg nicotine, which represents a lethal dose for pre-school children [5,6]. The Japan Poison Information Center received 36,578 queries on drug and chemical poisonings in 2002 and 3790 (10.4%) were for accidental ingestion of cigarettes, cigarette ends or their liquid extracts. A large proportion of those patients were children $\leq$5 years of age [7]. Suicide has been committed by adults who ingest tobacco leaves or their liquid extracts, although the number was relatively small [8,9]. Since nicotine is slowly absorbed and stimulates emesis, fatalities rarely occur after ingestion [10]. However, if 40–60 mg of nicotine is absorbed, then excessive salivation, increased respiratory secretions, seizures, and even death caused by paralysis of respiratory muscles can occur [4].

In this report, we describe a fatal poisoning caused by an additive interaction between methomyl and nicotine.

* Corresponding author. Tel.: +81 88 880 2418; fax: +81 88 880 2420.
E-mail address: moriyaf@med.kochi-u.ac.jp (F. Moriya).

0379-0738/$ – see front matter
doi:10.1016/j.forsciint.2004.06.013

2. Case history

A 35-year-old male (170 cm tall and weighing 63 kg), who was an outpatient with psychiatric disorders, was found lying in a prone position in his room by his father. Packages of medicine and a cup containing a bluish liquid found near the body suggested suicide. A white froth was observed in and about the mouth but no vomitus was found. He was in cardiopulmonary arrest on arrival at a hospital and officially pronounced dead without any resuscitation attempts. No miosis was observed in hospital, and the diameters of the pupils were 3 mm. An autopsy, which was conducted about 40 h after death, showed no petechiae on the conjunctivae of the eyes and froth tainted with blood in the trachea. The heart weighed 310 g and contained a moderate volume of liquid blood. The lungs were heavily edematous and weighed 580 g (left) and 750 g (right). The stomach contained 170 g greenish liquid with a small amount of shredded tobacco leaves, and the gastric mucosa was tainted green and congested with a large number of petechial hemorrhages. The bladder contained no urine. No marked changes, other than congestion, were observed in other organs.

3. Toxicological analysis

3.1. Sample collection

Small samples (10–20 g) of organs were obtained after the surface was quickly washed with running water and wiped dry with a clean towel. Fluid samples (5–10 ml) were collected using disposable clean syringes. Right femoral venous blood was drawn by needle puncture after the right iliac vein was cross-clamped. Cardiac blood samples were collected using clean syringes after the walls of the heart were incised. Whole stomach contents were mixed well in a beaker and a portion (approximately 50 g) was collected.

3.2. Drug screen and confirmation

Clear supernatant of the stomach contents was screened using Triage™ Drugs of Abuse (Biosite Diagnostic Inc., San Diego, CA). The stomach contents were also tested for general acidic, neutral, and basic drugs and chemicals by gas chromatography (GC) and gas chromatography–mass spectrometry (GC–MS), as described previously [11,12].

3.3. Measurement of methomyl

An analytical method described previously [12] was modified to measure methomyl in various body fluids and tissues. Briefly, 1 ml of body fluid, 1 g of stomach contents homogenate (stomach contents: H_2O, 1:99, w/w) or 1 g of tissue homogenate (tissue:H_2O, 1:3, w/w) was mixed with 50 μl 26.6 μg/ml dichlorvos (internal standard) in methanol and 1 ml 0.5 M phosphate buffer (pH 5.5). The mixture was extracted with 4 ml dichloromethane for 20 min on a mechanical shaker. To the lower organic layer separated was added isoamyl alcohol (20 μl) and dichloromethane was evaporated under a gentle stream of nitrogen at 35 °C. The residue was dissolved in 400 μl acetonitrile and partitioned with 400 μl *n*-hexane for 30 s on a vortex mixer. The upper hexane phase was aspirated and the partition step was repeated. Remaining acetonitrile phase was reduced to 100–200 μl under a gentle stream of nitrogen at 35 °C. A 1 μl aliquot of the reduced acetonitrile phase was injected into a GC–MS.

The GC–MS system comprised a Shimadzu GC-14A (Kyoto, Japan) equipped with a TC-1 capillary column [dimethyl silicone, 15 m by 0.53 mm i.d., 1.5 μm film thickness (GL Sciences Inc., Tokyo, Japan)], and a Shimadzu QP 1100EX (Kyoto, Japan). An initial column temperature of 80 °C was maintained for 2 min and then increased to 300 °C at a rate of 10 °C/min. The final temperature was maintained for 3 min. The temperatures of the injection port and interface were 300 and 250 °C, respectively, the electron impact ionization energy was 70 eV and the carrier gas was helium at a flow pressure of 15 kPa. Selected ion monitoring was performed for methomyl at *m/z* 105 and the internal standard at *m/z* 109. The lower limits of quantitation at S/N = 3 were approximately 2 ng/ml in body fluids and 8 ng/g in tissues.

3.4. Measurement of nicotine, cotinine and triazolam

Nicotine, cotinine and triazolam were quantified in various body fluids and tissues by GC, as described previously [13,14].

4. Results

Triage™ Drugs of Abuse screening was positive for benzodiazepines. The GC and GC–MS analyses confirmed the existence of methomyl, nicotine, cotinine and triazolam in the stomach contents. No alcohols and other volatiles were detected. Methomyl came from Lannate™, a water soluble powder that contains 45% methomyl, and triazolam came from Halcion™ tablets that contained 0.25 mg triazolam. Total ion and mass chromatograms of extracts of drugs and chemicals screened in the stomach contents are shown in Fig. 1.

While 304 mg of methomyl remained in the stomach, blood concentrations were 3–8 ng/ml and the pericardial fluid, brain, lungs and femoral muscle contained 40–551 ng/ml or ng/g (Table 1). Although bile, myocardium, liver and kidney contained no methomyl, cerebrospinal fluid and vitreous humor contained substantial amounts (2260–2680 ng/ml). Nicotine and cotinine concentrations in the blood were in the ranges of 222–733 ng/ml and 1180–2240 ng/ml, respectively. Other fluid and tissue samples contained nicotine and cotinine at 76–1160 ng/ml or ng/g