# TRAGIC FLYING ATTEMPT UNDER THE INFLUENCE OF "MAGIC MUSHROOMS"

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ABSTRACT: In this study a fatal case caused by the ingestion of magic mushrooms is reported. After a trip to the Netherlands a young girl had ingested a handful of psilocybe mushrooms with a soft drink. Soon later, she was trying to fly from the window and died in the street while falling from a room situated on the second floor of a house. The victim was not known by the law enforcement authorities to be a chronic drug addict. The autopsy revealed a traumatic cause of death whereas post-mortem toxicological analysis outlined consumption of psilocybin and cannabis. Psilocybin/psilocin content in the mushroom was determined by capillary electrophoresis. The general toxicological screening for drugs was negative except for psilocin and cannabinoids in blood and urine. Quantification of psilocin was done by EI-GC/MS operating in the SIM mode after derivatization with MSTFA and with bufotenin (a constitutional isomer) as internal standard. The concentrations were 0.06 mg/l for heart blood and 0.22 mg/l for femoral venous blood. Moreover, blood levels for cannabinoids were the following: 0.03 mg/l (THC), 0.008 mg/l (11-OH-THC) and 0.09 mg/l (THC-COOH). No psilocin could be detected in hair. This is the first case to our knowledge that psilocybin consumption was indirectly related to a fatal outcome.

**KEY WORDS**: Hallucinogenic substances; Psylocin; Cannabinoids; IE-GC/MS.

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## INTRODUCTION

## Case history

A 23 year old female (individual number 1) made a trip to the Netherlands with friends and her 19 year old sister (individual number 2). They were not known to the police as drug users. Individual number 1 ingested with a glass of soft drink a handful of "magic mushrooms" corresponding approximately to 5 g containing about 90 mg of psilocybin and smoked a cannabis joint. After a few hours she jumped over a vehicle in the street (9 m) out-of a window from the 2nd floor (6 m high). At autopsy the direct cause of death was found to be a traumatic fracture of the skull.

## MATERIAL AND METHODS

#### Chemicals

All reagents were of analytical grade. Psilocin and psilocybin were purchased from Radian Corporation (Austin, TX). N-Methyl-N-(trimethylsilyl)trifluoroacetamide (MSTFA) provided from Macherey-Nagel (Düren, Germany).

## Instrumentation

The capillary electrophoresis system was a P/ACE  $5510^{TM}$  from Beckman (Fullerton, USA). The capillary zone electrophoresis method used to separate psilocin and psilocybin is a modified method described by Pedersen-Bjergaard et al. [4]. Separation was performed using a eCAP<sup>TM</sup> fused-silica capillary (Beckman) with an internal diameter of 75  $\mu$ m and a length of 40 cm. The sample introduction was accomplished by hydrodynamic injection with pressure during 5 s. The system was run at 25°C with an applied voltage of 25 kV. The compounds were detected at 190 nm. Running buffer was a phosphate buffer (10 mM) at pH 7.2. Nalorphine was used as the internal standard.

The GC/MS instrument was from Hewlett-Packard: 7673A automatic sampler, 6890 series II gas chromatograph, 5973 mass selective detector. The gas chromatograph was equipped with a HP-Ultra 2 capillary column (12 m x 0.2 mm x 0.33  $\mu m$  film thickness, temperature-programmed from 70°C (2 min hold) to 220°C at 25°C/min, to 255°C at 5°C/min and to 300°C (7 min hold). The injector temperature was 260°C, the GC/MS interface temperature 280°C; the helium carrier gas flow rate was 2 ml/min. The mass spectrometer was operated in El mode with electron energy of 70 eV.

## Extraction of hallucinogenic substances from mushrooms

After drying, mushrooms were pulverized in a ball mill (Retsch, Germany) and extracted in an ultrasonic bath with methanol during 30 min at room temperature. After centrifugation, the supernatant was evaporated and dissolved in 10 µl of methanol. For injection 30 µl of the extract was evaporated and taken up in 30 µl of running buffer.

## **Extraction of psilocin from post-mortem specimens**

A solid phase extraction was done with Chem Elut at pH 8.5 with CH<sub>2</sub>Cl<sub>2</sub>: iPrOH 85:15. The residue obtained was derivatized with MSTFA.

## Extraction of psilocin from hair specimens

To reduce external contaminants the hair was washed with warm water (5 min) and two times with acetone (1 min). After drying with warm air, hair was cut into segments of 3 cm and the different segments were pulverized in a ball mill (Retsch, Germany).

About 30 mg of pulverized hair was incubated during 2 hours at  $60^{\circ}$ C in a urea (8 M)/thioglycate solution (0.2 M) at pH 3. After adjusting pH to 8, a solid phase extraction was done with Chromabond C18EC columns (Macherey-Nagel) as described in an earlier study [1].

The eluant was evaporated to dryness under a stream of nitrogen at 40 EC. The residue obtained was derivatized with MSTFA (20  $\mu$ l MSTA and 20  $\mu$ l pyridine).

## GC/MS

 $2~\mu l$  of the extracts were injected into the GC/MS system which was operating in selected ion monitoring (SIM mode) and programmed for detection of psilocin (m/z 290 for quantification and m/z 348 for qualifier). Bufotenin, a constitutional isomer of psilocin, was used an internal standard.

## **RESULTS**

Psilocybin and psilocin content in the mushroom was determined by capillary zone electrophoresis. Separation was done in 7 minutes (Figure 1). Concentrations of

Fig. 1. Electropherogram of an extract of psilocybe semilanceata.

psilocybin and psilocin found were respectively 1.1 g/100 g and 0.5 g/100 g of dry fungi.

The general post-mortem toxicological screening for drugs was negative except for psilocin and cannabinoids.

For individual number 1 specimens from the police surgeon labelled as post-mortem heart blood as well as autopsy material was used. Concentrations in heart blood were reported in Table I.

TABLE I. POST-MORTEM HEART BLOOD RESULTS (DAY ONE) FOR INDIVIDUAL NUMBER 1

Substance	Concentration [mg/l]
Free psilocin	0.03
Total psilocin	0.09
THC	0.03
11-OH-THC	0.008
THC-COOH	0.09

From autopsy (3 days later) the following results were obtained for individual number 1 (Table II).

The toxicological findings of blood and urine of individual number 2 is reported in Table III. There was not enough specimen for the THC assay.

TABLE II. AUTOPSY POST-MORTEM RESULTS (THREE DAYS LATER) FOR SUBJECT NUMBER 1

Material	Free psilocin	Total psilocin
Heart blood	0.06 mg/l	0.17 mg/l
Femoral venous blood	0.21 mg/l	4.60 mg/l
Urine	0.03 mg/l	1.95 mg/l
Bile	1.40 mg/l	6.65 mg/l
Liver	0.65 mg/kg	0.95 mg/kg
Kidney	0.55 mg/kg	0.60 mg/kg
Lung	0.15 mg/kg	0.10 mg/kg

In hair analysis of specimens from individual number 1, the amounts of amphetamines, benzodiazepines (diazepam, nordazepam and 7-aminoflunitrazepam), cocaine, cannabinoids and opiates were below cut-offs used. Furthermore, no psilocin was detectable in these specimens with a LOD < 150 pg/mg hair.

TABLE III. TOXICOLOGICAL FINDINGS FOR SUBJECT NUMBER 2

Material	Free psilocin [mg/l]	Total psilocin [mg/l]
Blood	0.007	0.091
Urine	2.3	5.4

TABLE IV. CONSUMPTION OF PSILOCIN AND EFFECTS [3]

Consumption	Dose	Effects
5 sp* a 0.2 g	1 g resp. 5–10 mg	Minimum for a trip
10 sp a 0.2 g	2 g resp. 10–20 mg	Small trip/good trip
20–40 sp a 0.2 g	4–8 g resp. 20–80 mg	Big trip
60 sp a 0.2 g	12 g resp. 60–120 mg	Bad trip

<sup>\*</sup> sp - specimens.

## DISCUSSION AND CONCLUSIONS

Capillary electrophoresis revealed to be a suitable method to quantitate rapidly both major hallucinogenic substances in psilocybe mushrooms. Concentrations found were similar to those found in the literature in other psilocybe fungi.

As hair analysis results were negative for all the drugs tested, this confirms that the subject was not a chronic user of drugs.

Whereas during autopsy the direct cause of death was found to be a traumatic fracture of the skull, post-mortem toxicological analysis outlined consumption of psilocybin and cannabis prior to death. In a volunteer study by Brenneisen et al. [3] the mean psilocin level in blood was found to be 0.008 mg/l after a consumption of 10–20 mg psilocybin. In our study the blood concentration of psilocin was much higher and a distinction was made between free and bound drug. To have an idea about the toxicity of psilocin, it may be interesting to consider the proposal of toxicity ranking evaluated by Gerault et al [2] (Table IV) in an OD-case.

To the best of our knowledge this is the first detailed description of an indirect fatal outcome after psilocybin containing mushrooms consumption and comprehensive post-mortem toxicological investigation.

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