Fatal poisoning by Hemlock water dropwort roots (*Oenanthe Crocata*)

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Introduction

This report describes the history and investigation of a suspected plant poisoning event in Portugal, involving the death of two adult males, who have mistakenly ingested Hemlock water dropwort roots (*Oenanthe crocata*) during a 36h resistance path in May 2017, in Santarém, Portugal. Six hours after beginning the path, they have called to emergency services for assistance because they suddenly become unwell, reporting plants ingestion and describing that they felt nauseated, vomits, seizures and disorientation. Both were found dead a few hours later in a rural area. In their backpacks, in addition to food supplies, they had fennel and spearmint that were probably picked up on the route. Further investigations by police authorities found that during that morning they shared several photos in social media, including screenshots of wild parsnips (*Pastinaca sativa*).

Methods

During standard autopsy procedures, both peripheral and cardiac blood were collected, as well as stomach contents. Systematic toxicological analysis was carried out on the post-mortem samples for the detection of pesticides, drugs, therapeutic agents and alcohol, with negative results. Methodologies utilized in the analyses include an enzyme-linked immunosorbent assay (ELISA) screen for classical abuse drugs, a gas chromatography–mass spectrometry screen (GC-MS) for therapeutic agents and pesticides, and a headspace-gas chromatography with flame ionization detection (GC–FID) screen for volatile compounds.

For hemlock water dropwort sample control, 10 mg of the dry roots were washed, cuted into small segments, mixed with methanol in a ratio of 1:9 (m/v) and stirred overnight at room temperature. Further, the methodology consists of grinding and homogenizing the dry roots and the gastric content samples with a ball mills using Precellys®24 (Bertin Technologies), followed by solid-phase extraction (SPE) with Oasis® HLB cartridge's and then identification and confirmation by GC–MS in the electron ionization mode (see Figure 1).

Chromatographic Conditions

- Injection volume: 2µL
- *Split* ratio 10:1
- Helium flow: 1,1 mL/min
- Capilar Column HP-5MS (30m x 250µm x 0,25µm)
- Acquisition mode: SCAN and SIM

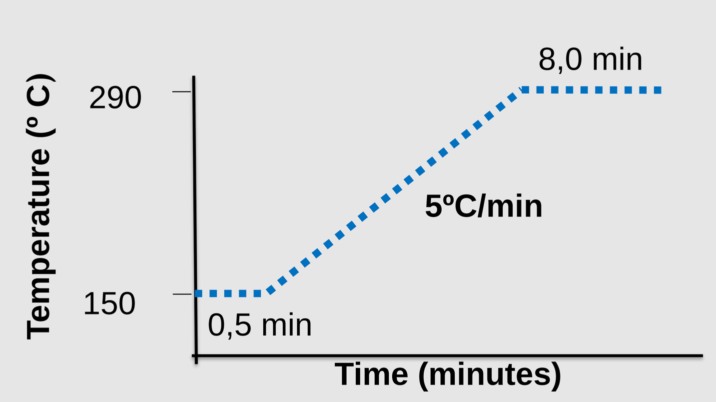


Fig 1. Flow chart of the analytical procedure.

Results and discussion

Plant fragments recovered from gastric contents were examined. A close inspection allowed to identify fennel and white vegetable fibres with orange particles (Figure 2). A review of current literature with search strings like "wild parsnips", made us suspect that they had actually ingested another plant: *Oenanthe crocata* (Figure 3), which is widely distributed in Portugal, grows along waterways and its roots (rich in *Oenanthotoxin*) are highly toxic to humans and domesticated animals (the ingestion of amounts as low as 1g/Kg can be lethal if immediate treatment is not administered).

Since no analytical standards were available for identification, the specific toxin (*Oenanthotoxin*) was isolated from the roots of a plant specimen, collected close to the death scene and positively identified as *Oenanthe crocata* by the Department of Plant Biology of the Faculty of Sciences of the University of Lisbon.

Fig 2. Fennel and white fibres recovered from gastric contents samples.

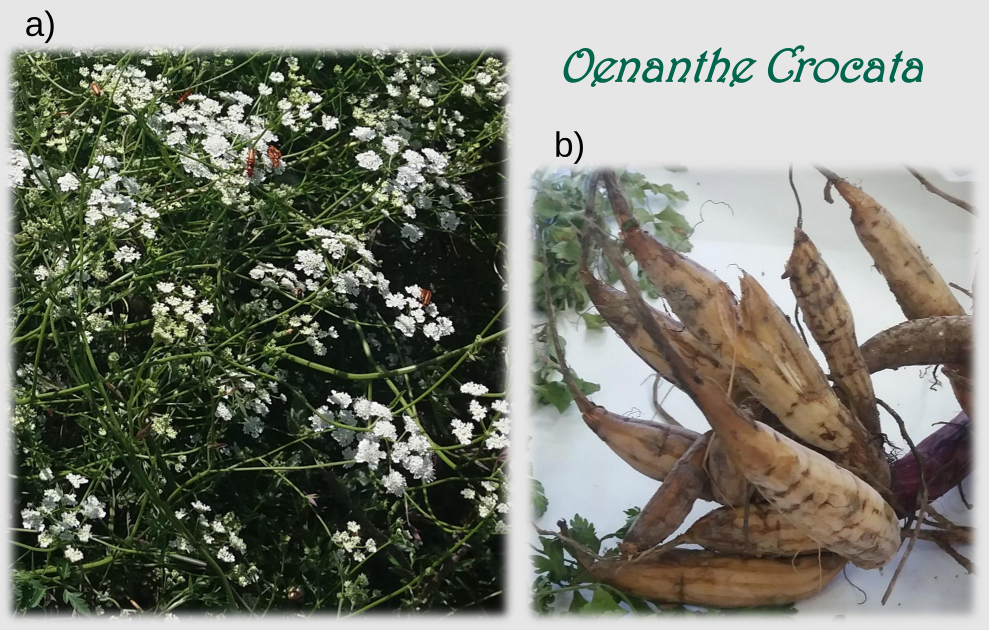


Fig 3. Oenanthe crocata, hemlock water dropwort : (a) white flowers ; (b) roots

The identification of *Oenanthotoxin* was made by GC-MS. The full mass spectrum (50-500 m/z) is shown in Figure 4. Stomach content samples were than analysed in SIM mode (*m/z* 128,153,258), and compared with a control prepared with the *Oenanthotoxin* isolated from the roots. All the criteria listed in Figure 5 for identification of *Oenanthotoxin* were fulfilled.

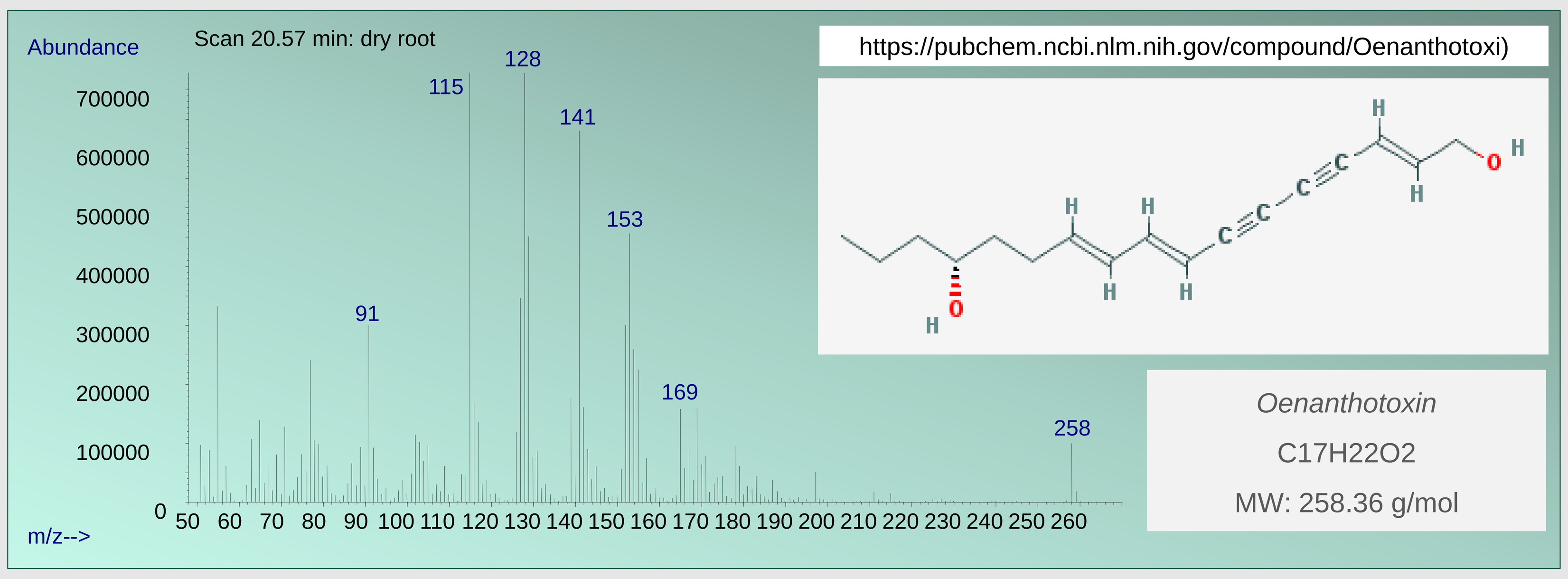
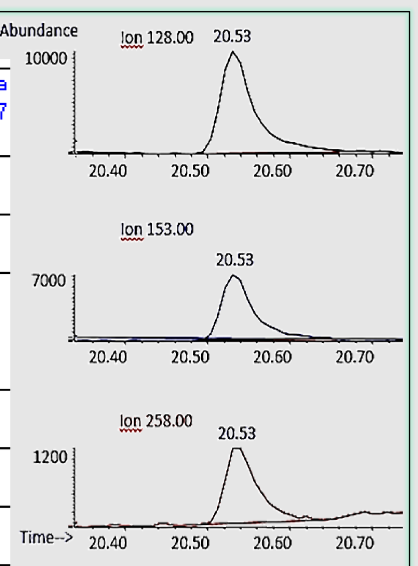


Fig 4. GC-MS mass spectrum of Oenanthotoxin, showing the structure of the compound found in gastric contents.

Sample:	0,35g of gastric content	Internal Standard:	promazine		Operator:	sfonseca
Substance:	**oenanthotoxin**	Control:	0,5g dry root		Date:	28/06/2017
Preparation:	20180723-1					

Control: 0,5g dry root	Ion	Area	relative Area	Tolerance	lower limit	upper limit
1st ion	128	13173466	100,0%			
2nd ion	153	8446716	64,1%	10 abs.	54,1%	74,1%
3rd ion	258	1488302	11,3%	5 abs.	6,3%	16,3%
oenanthotoxin (RT) =	20,55			0,1 abs.	20,450	20,650
promazine (RT) =	18,74	t.r.r. =	1,097	1% rel.	1,086	1,108

Amostra: 0,35g of gastric cor	Ion	Area	relative Area	acceptance:
1st ion	128	295690	100,0%	OK
2nd ion	153	182835	61,8%	OK
3rd ion	258	28135	9,5%	OK
oenanthotoxin (RT) =	20,53			OK
promazine (RT) =	18,73	relative RT=	1,096	OK



Identification Criteria:

- ✓ Ions relative intensities
- ✓ RRT< 2% or ± 0,1 min
- ✓ S/N ratio ≥3

Fig 5. Identification criteria of *Oenanthotoxin* in gastric contents.

Conclusions

Oenanthotoxin was successfully identified in the gastric contents of the deceased. Based on the known circumstances of death and the toxicological findings, the cause of death was attributed to accidental *Oenanthe crocata* poisoning.

This is the first reported case of confirmed toxicity and fatalities associated to the ingestion of *Oenanthe crocata* in Portugal.

This case highlights the difficulties usually observed in diagnosing plant poisoning cases and the importance of strict collaboration between forensic toxicologists and police authorities.

References

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King LA, Lewis MJ, Parry D, Twitchett PJ, Kilner EA. Identification of oenanthotoxin and related compounds in hemlock water dropwort poisoning. Hum Toxicol 1985 Jul;4(4):355-64.