Cypermethrin insecticide in viscera of suspected snake bite case: A Forensic case study

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Abstract:

The present paper reports the use of analytical techniques in identifying the reason of the death of a suspect. After the death of an individual, a post-mortem examination was conducted. As part of the investigation, the viscera (I) and (II) were sent to the Mini Forensic Science Laboratory in Solapur. The Medical Officer, in their report, mentioned that no injuries or defensive marks were found on the body, except for a small prick mark or bite mark with clotted blood observed on the right foot, between the small toe and adjacent fingers. Upon cleaning the area, it was noticed that one spot was more prominent, while the other spot was faint. Since there were no eyewitnesses present at the crime scene, the police recorded in their inquest that the death might be a result of a snake bite. No formal complaint was registered at that time. However, the viscera samples were preserved for further examination and a final opinion. In the analysis of the viscera, techniques such as Thin-Layer Chromatography (TLC) and Gas Chromatography-Mass Spectrometry (GC-MS) were employed. The results of the analysis revealed that the cause of death was cypermethrin poisoning, as indicated by the presence of cypermethrin in the viscera samples

Keywords- Snake bite, Cypermethrin, poisoning case, TLC, GC-MS

1.0 Introduction

Analytical techniques play a crucial role in forensic studies by providing valuable information and evidence that can be used to investigate crimes, identify suspects, and support legal proceedings. These techniques help in identifying and characterizing various types of evidence collected from crime scenes, such as fingerprints, DNA, fibers, drugs, poisons, gunshot residue, etc. and enable forensic scientists to determine the nature, composition, and origin of the evidence, which can link suspects to the crime or provide valuable information for the investigation. The present paper demonstrate use of GC-MS and TLC techniques in identifying the cause of the death of a victim

Systematic analysis procedure of analysis of viscera is very important and Forensic laboratory Maharashtra is having facility to analyze unknown poisons by using different analytical techniques including modern instrumentation[1]. In poisoning cases, determination of pesticide, insecticide and drugs is common practice in forensic science laboratory but in this case there is unknown poisoning and from the history it appears to be natural death.

Chromatographic techniques are discussed in literature analyzed for poisons in food[2], food products, biological samples…etc, Detection of pesticide of 11 pesticide residue samples from 70 postmortem cases is discussed[3] by Shailesh Kumar Rai *etal*. Thin layer chromatography is commonly used in organophosphorus pesticide analysis and tremendous work is done by using this technique[4-6]. The review of Mass spectrometry in application of toxicology revealed that the technique is really useful in forensic toxicology[7]

It is possible to analyse inorganic poisons as well by using techniques like ICP-MS, whereas LC-MS and GC- MS are used for analysis of organic poisons[7-8]. The separation of cypermethrin and deltamethrin is also important as they are from synthetic pyrethroid category[9]. Pesticides of organochloro, organophosphorus, carbamate group are generally used in suicidal, homicidal and even accidental cases; hence it was necessary in this case to check all types of poisons.

The case study was carried out on a 46-year-old male who was found to be dead in a prone position under a mango tree. According to the medical officer's report, the body exhibited the following observations: there were no injuries or defensive marks present on the body and the estimated time since death was 24 to 48 hours. Additionally, two small prick marks with clotted blood were observed on the right foot, specifically between the small toe and adjacent finger. Upon cleaning the area, one mark appeared more prominent while the other was faint. Hence,

he was suspected to have a snakebite The suspected case was analyzed in forensic laboratory, Solapur

2.0 Experimental

2.1 Chemical and reagents

Chemicals used for the analysis were potassium iodide, sulphuric acid, hexane, diethyl ether, acetone, methanol, chloroform, Ammonia, benzene and acetonitrile. All chemicals were used of A. R grade and are of Merck. The spray of chemicals used as Mercuric nitrate- Potassium Ferro cyanide, Mercuric nitrate- Diphenylamine, potassium iodate-starch, Alkaline resorcinol, Sodium carbonate - chloronil in acetone, O- Tolidine thereafter kept under UV, NaOH-Nickel amine, Copper acetate, O-tolidine, Dragendorff's and sodium hydroxide- ferrous sulphate, Standards like cypermethrin, Dichlorvos, Dimethoate, Malathion, glyphosate and parquet.

2.2 Procedure

Viscera samples were initially tested for volatile poisons like ethanol, methanol…etc, thereafter it was tested for inorganic poisons and organic poisons. Alkaline extraction of viscera I and II in chloroform was taken for further analysis, Acidic and neutral extraction of viscera I and II in ether was taken for further analysis.

2.3 Thin layer chromatography

Above extract was spotted along with standard poisons like cypermethrin, Dichlorvos, Dimethoate, Malathion, glyphosate and parquet on thin Layer chromatography of Silica gel and run in solvent system - Hexane:Acetone (8:2), Standard poisons and thereafter the plate was developed by using reagent mentioned in 2.1 for screening of all types of organic poisons.

2.4 Gas chromatography-Mass spectrometer

The Neutral ether extract was injected to GC-MS (Agillent technologies). HP- 5ms capillary column 25m x 0.25mm x 0.1µm was used, the method was set as inlet temperature 150^0C, MS source temperature-230^0C, QP temperature 150^0C, initial temperature of oven was kept 70^0C and hold for 5, Ramp was kept 5^0C per min heated up to 280^0C, helium was used as carrier gas with flow rate 1ml/min.

3 Results and Discussion

Volatile poisons like methanol, ethanol were not found and Inorganic poisons like arsenic, Mercury were absent in viscera samples. It was further screened for organic poisons like orgaochloro, organophosphorus, herbicides, cypermethrin and drugs by using TLC. Presence of cypermethrin was revealed in viscera sample (Fig.1). 10 TLC plates were run in solvent

system- Hexane:Acetone (8:2) and separately treated by using above reagents. The observations are given in Table 1.

The neutral ether extract of viscera I & II was further injected to GC-MS which showed presence of cypermethrin. Mass spectra m/z- 39, 51, 77, 91, 115, 127, 141, 152,163, 181, 191, 197, 209 tallies with cypermethrin.

Conclusion

In summary, although no visible injuries were found on the body, the examination of the viscera using analytical methods (GC-MS, TLC) revealed that the individual's death was a result of cypermethrin poisoning and not due to the snakebite. This finding provides important evidence for the investigation into the circumstances surrounding the death.

Figure:

1 2 3

1-Standard cypermethrin, 2-Viscera II sample, 3- Viscera I sample

Fig.-1 Thin layer chromatography of viscera samples

Table:

Table 1 Observation of viscera sample treated with spray reagent

Sr. No.	Reagent	Observations
1	Mercuric nitrate- Potassium ferrocyanide,	No spot
2	Mercuric nitrate- Diphenylamine	No spot
3	Potassium iodate-starch	No spot
4	Alkaline resorcinol	No spot
5	Sodium carbonate- chloronil in acetone	No spot
6	O- Tolidine thereafter kept under UV	No spot
7	NaOH-Nickel amine	No spot
8	Copper acetate, O-tolidine	No spot
9	Sodium hydroxide- ferrous sulphate thereafter plate was heated in oven there after hydrochloric acid	Bluish green spot at Rf -0.67 it tallied with cypermethrin
10	Dragendorff's	No spot

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