

Death is Due to Poisoning but Viscera Report is Negative

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Abstract

The conclusion of cause of death in a case where the death is due to poisoning but viscera report give negative result, pose a confusion to law and public. The intricacy of failure to find poison in viscera of the individual whose death is due to poisoning is a routine problem in India and the reasons of it are delay in examination of the viscera, improper preservation of the viscera, use of wrong analytical technique, early disintegration of poisons, complete metabolism of poisons in the body, the amount of poison in the viscera being negligible, lack of suitable chemical test for certain poisons, tempering of preserved viscera, and the biggest issue is only common poisons are screened, residual analysis of poisons, which are limited to common poisons available in the area. The other major poisons/chemicals like insulin, KCl, Adrenaline can't be detected. The salient points covering various aspects of viscera analysis in various Forensic Science Laboratories have been discussed with especial reference to the false positive or false negative results and interpretation of viscera report when it is negative in truly positive cases and vice-versa.

Keywords: Toxicology; Poisoning; Viscera; Autopsy; Collection; Preservation; Analysis; False negative Report.

Introduction

Poisoning is a significant contributor to mortality and morbidity throughout the world. More than three million poisoning cases have been reported out of which, 99% of fatal poisoning occur in developing countries, predominantly among farmers due to poisoning; including poisonous toxins from natural products are handled [1]. Suicidal self poisoning is the commonest form of poisoning in adults and accounts for at least 95% of all poisoning admission to hospital. In India majority of death due to fatal poisoning are of married males of 10–35 years of age belong to low socioeconomic status from a rural area

[2–4].

Incidences are common where no trace of poison was detected on chemical analysis of viscera, while from history and other circumstantial evidences it is almost certain or quite certain that poison was the cause of death. It may be due to many reasons, but commonly observed in hospitalized patients in which either the poison has been removed from the stomach and intestines, or detoxified, conjugated and eliminated by the kidneys and other channels or the quantity present is below the detection limits which makes its detection difficult or even impossible by the present methods of chemical analysis. In present study we tried to find out the causes of false negative in routine viscera analysis [5–9].

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Common terms used

Toxicology

It deals with properties, action, toxicity, fatal dose, detection, estimation or interpretation of the result of chemical analysis and management of Poisons.

Clinical Toxicology

It deals with human diseases caused by or

associated with abnormal exposure to chemical substances.

Acute toxicity

Acute toxicity is that property of a substance which causes adverse effect in an organism through exposure to the substance in a single short term dose.

Chronic toxicity

Chronic toxicity is that property of a substance which causes adverse effects in an organism through repeated or continuous exposure to the substance.

Oral toxicity

Oral toxicity is that property of a substance which causes adverse effects in an organism by the swallowing of that substance.

Dermal toxicity

Dermal toxicity is that property of a substance which causes adverse effect in an organism, if the substance is absorbed through the intact skin of the organism.

Inhalation toxicity

It is that property of a substance which causes adverse effects in an organism by the substance being absorbed during respiration of the organism.

Toxicity

Toxicity is that property of substance which causes any adverse effect to an organism.

Hazard

Hazard is the probability of an adverse effect to occur.

Lethal Dose (LD50)

It is measure of toxicity and represents that dose of a substance which when administered kills 50% of the test population of animal.

Subacute poisoning

It shows features of both acute and chronic poisoning.

Fulminant poisoning

It is produced by a massive dose. In this death occurs rapidly, sometimes without preceding symptoms.

What is poison? [7,15]

A Poison may be generally described as any substances which when administered or taken in small quantity is capable of producing deleterious symptoms on the body.

Or

A substance may be termed a poison that has cumulative effect if administered for a length of time so that ends fatally.

Or

A substance that is capable of causing the illness or death of a living organism when introduced or absorbed.

Or

A substance that can cause people or animals to die or to become very sick if it gets into their bodies especially by being swallowed.

Or

A substance that through its chemical action usually kills, injures, or impairs an organism.

Or

A substance with an inherent property that tends to destroy life or impair health.

Or

Poisons are substances that cause disturbances to organisms, usually by chemical reaction or other activity on the molecular scale, when a small quantity is absorbed by an organism.

Or

Any substance that can cause severe organ damage or death if ingested, breathed in, or absorbed through the skin.

Sources of poisons [7,8,14]

1. Domestic or household sources

In domestic environment poisoning may more commonly occur from detergents, disinfectants, cleaning agents, antiseptics, insecticides, rodenticides etc.

2. Agricultural and horticultural sources

Different insecticides, pesticides, fungicides and weed killers.

3. Industrial sources

In factories, where poisons are manufactured or poisons are produced as by products.

4. Commercial sources

From store-houses, distribution centers and selling shops.

5 Uses as drugs and medicines

Due to wrong medication, overmedication and abuse of drugs.

6. Food and drink

Contamination in way of use of preservatives of food grains or other food material, additives like coloring and odouring agents or other ways of accidental contamination of food and drink.

7. Miscellaneous sources

Snake bite poisoning, city smoke, sewer gas poisoning etc.

8. Animal poisons

Snake, scorpion and insect bite etc.

Restricted list of common poisons [9]

Poisons may be classified in many ways. However, As per Forensic sera laboratory in India, list of common poisons include the following¹⁰⁻¹⁴.

1. Gaseous poisons: Carbon monoxide, phosphine, cyanide etc.
2. Volatile poisons: Ethanol, methanol, ethylene dibromide, chloral hydrate, ethylene glycol etc.
3. Non-volatile poisons: Drugs such as Barbiturates, Benzodiazepines, Phenothiazines salicylates, Amitryptaline, Opiates and Narcotics, Amphetamines etc.
4. Metallic poisons: Lead, mercury, arsenic, antimony etc.
5. Pesticides: Organochloro, Organophosphorus,

Carbamates & pyrethroids etc.

6. Anions: Bromides, chlorates, fluorides, nitrates etc.
7. Miscellaneous: Kaner, Dhatura, fertilizers, etc.

What is viscera test?

The analytical toxicology compels the determination of the poisonous substances found in the body and its excreta. Methods vary to enable the toxicologist to choose the procedure that best fit to the particular laboratory facilities, personal preferences and the conditions of that moment.

Concepts of viscera test

Forensic toxicology concern analytical checking of statements made by witnesses during the course of police inquiry. So it needs a high degree of perfection right from the collection and preservation of the test samples to the analysis, result and interpretation with due consideration to the artifacts which may develop during this course due to one or the other reasons like improper collection, preservation, handling, decomposition and interpretation of results.

Collection, preservation and forwarding biological materials for toxicological analysis [7-9, 14]

1. The quantity of viscera/body fluids to be preserved in all cases of death due to poisoning or suspected poisoning
 - a. Stomach: Whole with its full content
 - b. Liver: Half or 500 gms
 - c. Small intestine: 30 cms
 - d. Kidney: One half of each Kidney
2. In all cases of suspected poisoning including carbolic acid, saturated solution of common salt should be used as preservative for viscera.
3. Blood is preserved in all cases and quantity of blood should be about 100 ml.
4. Blood is preserved in Sodium fluoride, oxalate, EDTA, mercury chloride, gold chloride citrate etc.
5. Some recommend preservation of spleen as a

- routine measure. But, when other organs are preserved, preservation of spleen does not add much to the advantage.
6. In cases where poisoning by acids is suspected (except carbolic acid), rectified spirit should be used as preservative. Denatured alcohol or formalin should not be used while preserving the samples for toxicological analysis.
 7. For determining the alcohol levels in living persons suspected to have consumed alcohol, 5 ml of blood using sodium fluoride as preservative, and minimum of 10 ml of urine without preservative should be collected and forwarded.
 8. In case of road accidents and where the death is suspected due to influence of alcohol 5 ml of blood should be collected and similarly in case of alcohol poisoning or drunkenness cases 5 ml blood and 10 ml urine samples should be collected and preserved properly.
 9. In case of burn victims to determine the levels of carbon monoxide, 10 ml of blood sample preserved by putting a liquid paraffin layer above it should be collected in a glass bottle and forwarded for analysis.
 10. Urine should be preserved, if urine is available in the bladder. While collecting urine it should not get contaminated with blood. Urine can be syringed out or spooned out after dissecting the anterior wall of the bladder. Quantity collected 100 ml or whole amount in the bladder.
 11. No useful purpose will be served by chemical analysis of viscera in case of electric shock victims and persons known to have died due to diseases like TB, cancer, hepatitis, AIDS etc.
 12. It will be of no consequence if chemical analysis of viscera is carried out in case of natural deaths due to starvation, sunstroke, old age, lightning, extreme cold etc.
 13. In case of drowning where death is due to drowning, no additional purpose will be served by chemical examinations.
 14. Testing for diatoms in visceral organs, spleen and bone marrow may be most useful in cases of drowning. In such case control sample of the water in which body was recovered should be collected.
 15. In case of snake bite or other insect bites, samples of skin bites of affected area should only be collected.
 16. In cases of deaths due to administering injections, the sites of injections, subcutaneous tissues of skin (along with the needle tract) weighing about 100 gms should be collected.
 17. In case of inhalation of gaseous inhaled poisons like carbon monoxide, coal gas, hydrocyanic acid, chloroform or other anesthetic drugs, the lung tissues, brain and blood from the cavity of the heart should be preserved.
 18. Shaft of long bones, a tuft of head hair, finger and torn nails and some muscles should be preserved in case of chronic poisoning by heavy metals like mercury, arsenic, lead etc.
 19. In cases of prolonged use of drugs like barbiturates, samples like hair, nails are to be collected for chemical analysis.
 20. A piece of heart, portion of brain and spinal cord should be preserved if poisoning is by nuxvomica or strychnine.
 21. Brain and urine should be preserved in suspected cases of poisoning by barbiturates, opium or anesthetics.
 22. In highly putrefied bodies, larvae, maggots, pupa and the other entomological samples should be preserved and forwarded.
 23. In cases of embalmed bodies vitreous humour from eye balls usually remains uncontaminated by the process and may serve the purpose of analyzing urea, creatinine and ethyl alcohol, hence such samples should be collected and forwarded.
 24. Fatty tissue should be taken from abdominal walls in case of pesticide poisoning.
 25. Soil samples from above, beneath and sides of the dead body and control soil samples away from the dead body should be taken in cases of exhumed or skeletalised dead bodies.
 26. Parts of both lungs should be preserved in all cases of poisoning with volatile poisons or poisons partly excreted with the expired air.
 27. Whole of the heart is preserved in cardiotoxic poisoning.
 28. Whole of the brain is preserved in suspected cerebral poisoning.
 29. In suspected heavy metals like lead poisoning cases, blood is preserved.
 30. Hair should be preserved in case of poisoning with arsenic or copper.
 31. Hair and Nails should be preserved in case of poisoning with arsenic.

32. Uterus and vagina are preserved if abortifacient drug was used, locally or systematically, when the drug has direct action on the uterus.
33. Skin scrap from an area stained with a suspected poison.
34. Suspected stained area of the dress, suspected packet of poison, strips of tablet recovered from the pocket of the dress of deceased.

Analysis report and its meaning [9-12]

Chemical analysis reports are usually expressed in different ways by different laboratories which are discussed below:

1. Article does not contain any chemical poison

It indicates that the viscera have been examined for all the common poisons and none found positive.

2. No chemical poison could be detected

It means, if the case history is given as suspected poisoning and during analysis no positive result is found.

3. Sample is not fit for analysis

Sometimes sample received for analysis are in the putrefied decomposed and dried form. Moreover, quantity of viscera sent for analysis is very less to carry out all the possible analysis. In such condition it is reported like this.

Different causes of false negative viscera report [7, 15-21]

In some cases, the history, circumstance and the postmortem findings may clearly suggest that poisoning is the most likely cause of death but the toxicology report may be still negative which is called false negative report. The possible explanations of false negative reports are:

1. In the case where the victim/deceased was admitted to the hospital for a considerable time, the possibility of detecting poison in the viscera gets lesser. In such conditions often viscera report may be negative. In real poisoning cases and some other substances other than the poison may come positive which was taken in the hospital for therapeutic

reason like promethazine etc. The medication itself may alter the poisonous substance and make its detection difficult or even impossible. Hence, it is Important to know about the treatment records. Ask for treatment notes, PM notes since the patient is hospitalized.

2. In postmortem decomposition, many poisons present in the tissue undergo chemical changes which cannot be detected. Putrefaction of normal tissue may produce substances, which may give chemical reaction similar to those obtained from toxic compounds. Most volatile compounds are lost due to putrefaction.
3. The whole poison may disappear from the lungs, in case of volatile poison by evaporation or oxidation due to faulty preservation like dried up blood sample, organic solvent poison gets evaporated during extraction and concentration. Due to lack of preservatives the blood clots. We cannot determine volatile poisons like alcohol, acetone, aldehydes, etc. For gas poisoning the blood should be adding paraffin oil layer over blood sample.
4. The poison might have been vomited out, excreted, neutralized, metabolized, detoxified to such a quality that it cannot be detected by general chemical analysis. The poison after absorption may be detoxified and eliminated. Vomit of persons should be sent also stomach as per it should be collected by I.O. from the scene of crime even if it is dried. Before giving the treatment the M.O. should collect the stomach aspirate of the patient admitted due to poisoning.
5. It is well known that usual routine toxicological screening procedures may not detect hemoglobin like carboxyhaemoglobin, sulphamethemoglobin and methemoglobin, diuretics, solvents, radioactive compounds, antibiotics, non-steroid anti-inflammatory substances except aspirin and paracetamol, calcium channel blockers, beta blockers etc.
6. Some drugs are rapidly metabolized and there may be complete metabolism of the poison in the body. Drugs like haloperidol and oxycodone are rapidly metabolized and only metabolite can be detected. Early disintegration of poison like narcotism, the substances are rapidly metabolized and create difficulty in analysis. Some drugs have short half life and are rapidly metabolized. In delayed death cases poison may be excreted or detoxified completely.
7. Biological toxins and snake venom are protein and cannot be separated from the body tissues.

- Immunoassay method may detect these poisons but this facility is not easily available in all Forensic science laboratories.
8. Some insecticides, fungicides which are highly soluble in water and they are not extracted in diethyl ether common solvent for extraction of poison in toxicological work.
 9. Some organic poisons decompose due to improper preservation. Drugs which have been found to decompose during the storage at 40°C are clonazepam, cocaine, isoniazid, methadone, morphine and nitrazepam. Cocaine gets hydrolyzed in alkaline medium. Alkaloids and phenothiazines like L.S.D and psilocybin are photo labile. Catecholamines are easily oxidized if container is not airtight.
 10. Some drugs are in very small amount and need considerable amount of viscera and sensitive analysis procedures. For example, Amphetamines may be undetected in the blood owing to the very low concentration present, even after fatal dose. Many potent drugs like clonidine, ergot alkaloids, dioxin, digoxin, THC pose such problems.
 11. Diacetylmorphine (Heroin) is rarely detected as it gets rapidly hydrolyzed to monoacetyl morphine which is present in urine in small concentration.
 12. Anaesthetics are mostly esters and get hydrolyzed at room temperature. Many highly volatile substances like aromatic and halogenated hydrocarbons, solvents, anesthetic agents, and noxious gases as H₂S, CO, NO₂ behave in this manner.
 13. Some substances like fentanyl may have structural dissimilarity from their drug class prototype and give negative results for that particular group.
 14. Tampering of viscera during preservation and in preserved bottles with vested interests or wrong motive. Addition of strong chemicals like soap, bleach powder or glutaraldehyde alters the results in immunoassay. Sometimes the paper label of the bottle after few years is torned/falls in storage room and a wrong level is put on viscera bottles by store in-charge/ M.O.
 15. Poor laboratory quality assurance, defective analysis, as many laboratories are not upgraded with modern facilities.
 16. As a protocol, substances detected may not be reported as these are naturally occurring substances or constituents of the human body like phosphates.
 17. It has been observed by many researchers in routine practice that too polar like iron, lead and ethyl glycol, volatile like solvents, aromatic or halogenated hydrocarbons gases, non- volatile like plant or fungal alkaloids, low concentration like very potent drugs and substances, toxic anions like thiocyanate, cyanide, fluoride and nitrites and new substances like busiprone are not detected by conventional toxicological screening.
 18. In burn cases if the blood is completely charred and dried, in the cases of alcohol intoxication if the sample is not preserved properly in sodium fluoride and potassium oxalate and analyzed after some time gap. It is decomposed and results are never dependable in such cases. Blood sample should be kept in refrigerator till the time of analysis. All biological specimens received in the laboratory are stored in cold room till taken for their analysis.
 19. It is well known that usual routine toxicological the screening procedures in FSL's include limited poisons called as common poisons such as volatile poisons, non volatile poisons, pesticides (most commonly used in that region by farmers for crop protection), metallic poisons, cyanides, alcohol etc. may not be detect other than common poisons.
 20. Forensic science laboratory doing only residual analysis of chemical compounds, not metabolites. For complete screening metabolic study is also important. For that advanced technique like GC-MS should be used as in USA.
 21. Sending insufficient biological material by M.O. such as tissue, blood, urine etc. may also affect the result.
 22. Some vegetable alkaloids cannot be detected by chemical analysis. As they are highly water soluble and cannot be extracted in ether or other organic solvents.
 23. Use of wrong analytical techniques/methods.
 24. Non availability of sophisticated instrument like GC-MS, GC-MS/MS, LC-MS, LC- MS/MS.
 25. Lack of expertise, qualified and experienced toxicologists / scientists / experts / purity of chemicals and availability of poor control insecticide / drugs / alkaloids / planed poisons / animal toxins. A negative screen does not necessarily mean that a toxin is not present. It says only that none on the list of those that were screened has been found.

Different causes of false positive viscera reports

1. When the viscera is not properly preserved and sent for analysis after a lapse of time there is every possibility of postmortem production of ethyl alcohol, cyanide, carbon monoxide, ketones, sulfides etc, which can give false positive result.
2. If viscera is preserved in formalin or denatured spirit, it may give false positive test for methanol and ethanol poisoning.
3. False positive results in immunoassays are mainly due to cross reactivity and structural similarity.
4. False positive reaction for opiate group can be seen on consumption of poppy seeds, chlorpromazine, dexamethorphan and diphenylolate tablets.
5. There may be false positive reaction for amphetamine due to cross reactivity on account of consumption of ephedrine nasal drops, pseudoephedrine in cough syrups, chloroquine and procainamide.
6. Phencyclidine is falsely represented by dextromethorphan, diphenhydramine, doxylamine and thioridazone.
7. Faulty instruments and lack of their standardization may lead to false positive or false negative results.
8. Faulty figures of substances in reporting due to human error on viscera examination from laboratories. It is suggested that chemical examination report should not be taken as a gospel truth in each and every case.
9. Tampering of viscera during preservation and in preserved bottles with vested interests or wrong motive.

Conclusion

The salient points covering various aspects of viscera analysis have been discussed with special reference to the false negative result. Finally more research needs to provide better understanding in analysis of viscera due to poisoning. It is also necessary to know how we can reduce the incidences of false negative chemical report. The policy maker should take the help of Forensic experts for better results and possible substitution of methods.

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STATEMENT ABOUT OWNERSHIP AND OTHER PARTICULARS
“Indian Journal of Maternal-Fetal and Neonatal Medicine” (See Rule 8)

- | | | |
|---|---|--|
| 1. Place of Publication | : | Delhi |
| 2. Periodicity of Publication | : | Quarterly |
| 3. Printer's Name | : | Asharfi Lal |
| Nationality | : | Indian |
| Address | : | 3/258-259, Trilok Puri, Delhi-91 |
| 4. Publisher's Name | : | Asharfi Lal |
| Nationality | : | Indian |
| Address | : | 3/258-259, Trilok Puri, Delhi-91 |
| 5. Editor's Name | : | Asharfi Lal (Editor-in-Chief) |
| Nationality | : | Indian |
| Address | : | 3/258-259, Trilok Puri, Delhi-91 |
| 6. Name & Address of Individuals
who own the newspaper and particulars of
shareholders holding more than one per cent
of the total capital | : | Asharfi Lal
3/258-259, Trilok Puri, Delhi-91 |

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