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IP International Journal of Forensic Medicine and Toxicological Sciences

Journal homepage: http://www.ijfmts.com/

Review Article

Forensic significance in the cases of drowning deaths: An elaborative study

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ARTICLE INFO

Article history:
Received 27-11-2021
Accepted 14-12-2021
Available online 21-01-2022

Keywords:
Drowning
Death
Diatoms
Bone Marrow
Medico legal aspect

ABSTRACT

Drowning is a form of asphyxia which is caused by submersion/immersion of the body in water or any other fluid that occurs due to the aspiration of fluid into air passages and is mostly accidental. The main question arises in the case of a body recovered from water is whether the individual was alive at the time he entered water. If a body is found in water is does not necessarily mean, that this person has drowned. The bodily external and internal findings are necessary in medicolegal investigation of drowning deaths. Drowning is difficult to determine and are often diagnosed by eliminating other potential causes of death whereas diatom test is significant for the concluding analysis of drowning deaths. Diatoms found inside the body may serve as corroborative evidence in the diagnosis of the cause of drowning death. It can be ascertained whether the drowning is ante-mortem or post-mortem. The diatom test considered as the only tool to examine drowning cases. The present study highlights some specific points to drag conclusive results in the investigation of deaths due to drowning.

1. Introduction

Drowning is asphyxia in nature which prevents entering of air into the lungs via inhalation of fluid into air passages i.e., nose and mouth.[1] In drowning cases the death is caused by irreversible brain damage which occurs the development of irreversible cerebral anoxia and hypoxia. The several phases have been described during the drowning process, first a breath-holding phase, followed by involuntary inspiration, gasping for air and loss of consciousness. The state of consciousness is generally developed within three minutes of submersion.[2] Death of a victim found in water should not always be related to drowning so the diagnosis of drowning is one of the most difficult tasks in the field of forensic medicine. The finding out the cause of death in those cases is challenging. Only a systematic examination and complete autopsy can determine the death as presence of asphyxia symptoms of drowning.[3] In general, it is also found that the physical/external and autopsy examinations are not as specific in drowning cases as complete laboratory examination.[4] However, the majority of bodies found in water are typically determined the death from asphyxia due to drowning and in many cases ischemic cardiovascular disease, also act as a contributing factor. Many further investigations also proved that underlying natural diseases may contribute to cause death in water. In some circumstances, the cause of death is homicidal while others are natural due to some intoxication, influence of drugs and alcohol or lack of swimming ability.[5] In the recovered bodies from drowning deaths, it is still a challenge to ascertain a death due to the absence of typical post mortem findings.[6] In these circumstances the presence of diatoms is only screening method which is reliable and it signifies a microbial fingerprint of the time and place

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https://doi.org/10.18231/j.ijfmts.2021.026

of occurred drowning.[7] This diatom test is very important examination to determine the drowning was antemortem or postmortem in nature.[8] In drowning related deaths, diatoms get deposited in the internal organs i.e., Kidney, liver, lung, brain and in the bone-marrow of the bones i.e., long bones. This is due to the inhalation of water that would penetrates into the blood circulation from alveoli through the alveolar spaces of lungs enter in the alveolar spaces.[9–12] Though, in antemortem drowning the entering of any diatom in lungs with water is not possible. It would not be transported to distant body organs due to the lack of blood circulation. According to the Forensic law of comparison "Only the likes can be compared". Similarly, in drowning cases the water sample which is collected from the site of drowning and the diatom extracted from the organs and tissues could be compared to established a correlation between them.[13–15] The present study focuses on determining the drowning related deaths with medicolegal and forensic aspects.

1.1. Drowning death investigation in autopsy

1.1.1. External findings

In Medicolegal terms, the external findings in death investigations could be ligature mark, state of left and right eyes, pupils, Cornea/Conjunctiva, Natural orifices, the presence of blood, froth etc. in mouth, nose, ears.[16]

1.1.2. Internal findings

In the postmortem findings it includes;

1. Cranium & spinal Cord (Brain must be exposed in every case, Spinal cord need not to be examined except in case of injury to vertebral column/Spinal Cord), Scalp, Skull, Meninges and Vessels, Brain / Brain Weight, Vertebrae & Spinal Cord
2. Mouth, Pharynx & Oesophagus
3. Neck -Condition of neck tissues Thyroid, Hyoid bone, Larynx & Trachea
4. Thorax-Chest wall, Ribs/Sternum and Cartilage, Pleura / pleural Cavity, Lung (Rt & Lt) Weight, Pericardium, Heart / Heart Weight, Coronary Arteries & Large Blood Vessel
5. Abdomen- Peritoneum, Retro peritoneum, Stomach and its contents, Small Intestine and its content / Length Small Intestine, Large Intestine and its content / Length Large Intestine, Liver and Gall Bladder / Liver Weight, Spleen / Spleen Weight, Pancreas / Pancreas Weight, Kidney (Rt & Lt) Weight

Drowning Deaths may be Homicidal, Suicidal and Accidental (Fig. 1 to 6). If Cause of Death found due to asphyxia as a result of drowning, then samples will preserve for further diatom test. Suicide, homicide or accident investigators dilemma of drowning in the pictures given below:

Fig. 1: Partial drowning

Fig. 2: Body features skin removal

Fig. 3: Partial drowning

Fig. 4: Suicide at Sagartal

Fig. 5: Homicidal drowning

Fig. 6: Hand, foot tide and washer women hands

1.2. Diatom testing

Diatom testing is a most important test in drowning case in forensic investigation. Diatoms (Bacillariophyceae) have been classified as a group of algae which is unicellular, photosynthetic and eukaryotic microorganisms.[17] Diatoms having various types of different characteristics such as shape, size and color. They can be distinguishable based on founded species with unique silica cell walls also known as "frustules" and can be vary depending on their environment. Diatoms create flora profiles that can be analysis for forensic purpose. It is an extremely diverse microorganism comprising more than 200000 known species.[18] Their cell walls composed of transparent opaline silica and are adorned by intricate and striking patterns of silica. Diatoms live in water, or even in moist environments or soils.[19] Some diatoms appear as free-floating cells in the plankton of ponds, lakes, oceans and can be found in rivers, springs marine waters, fresh water etc.[20] Traditionally diatoms are divided in two distinct shapes: centric diatoms (Centrales), which are radially symmetric, and pennate diatoms, which are bilaterally symmetric (Pennales) but these are further classified into three classes: centric diatoms (Coscinodiscophyceae), pennate diatoms without a raphe (Fragilariophyceae), and pennate diatoms with a raphe (Bacillariophyceae).[21] The diatoms are of unicellular organization, but some form colonies. All microscopic diatoms cells range in size from 5micron μm to 3mm millimeter that can be viewed and observed with the help of microscope. Generally, the use of light microscope and Scanning electron microscope to view and detect the diatoms. It was firstly observed in 1703 with the help of a simple microscope.[22] To check the presence of diatoms in tissues and organs is an effective method that can help to solve drowning cases after a required thorough examination to affirm any results.[23] The diatom test is very significant in medicolegal aspects and a valuable tool in forensic science to identify the drowning deaths with clear interpretation of results.

1.3. Extraction of diatoms

To the detection of diatoms, a comparative study of the species of diatom in the water sample and organ sample should be completed and accordingly the results would be correlate. Various type of developed techniques available for the extraction of diatoms. Acid digestion method which used nitric acid (HNO_3) and Hydrogen peroxide (H_2O_2) for the extraction of diatoms.[24,25] Enzymatic digestion method using proteinase K, ATL Buffer and 5N HCl by Qiagen which is effective and simplify for diatom extraction from suspected drowning cases. This method is less time-consuming and less laborious.[26] A new method called Microwave Digestion-Vacuum Filtration Automated Scanning Electron Microscopy (MD-VF Auto SEM) with microwave digestion and vacuum filtration we also developed for diatom testing.[26,27] Fluorimetry is a technique which is also used for the isolation of diatoms from tissue samples on the basis of luminescent properties. In this method the specific fluorescent tags can differentiate diatoms from the collected water samples at the scene of incidence and from the body tissues in drowning dead bodies.[27] Molecular biology introduces molecular methods called gene sequencing and PCR (Polymerase Chain Reaction) based method of diatom testing[14–16] i.e., 16s/18s rRNA subunits of ribosomal RNA for the detection of planktonic DNA from human tissues samples in drowning cases.[28,29] Traditional methods are widely considered in the laboratories called Acid/Chemical Digestion Method.

1.4. Acid digestion method of diatom extraction

In this method different acids were used for the digestion of diatom cell i.e., Nitric Acid (HNO_3), Sulfuric acid (H_2SO_4), Hydrochloric acid (HCl) and Hydrogen peroxide (H_2O_2). In Acid Digestion Method take acid into the conical flask with sample then heat on water bath. Centrifuge the sample and wash the pellet with double distilled water. Now dissolve pellet in water then take microscopic slide for smear preparation and the diatoms can be seen with the help of compound microscope[30]

1.5. Extraction of diatoms from water samples

Thoroughly shake the water bottle of sample and leave it for some time to let it settle down then carefully discard the upper water. Take about 50 ml of water sample with settled material and transferred it into a sterilized glass beaker of 100 ml. Add concentrated nitric acid (10-20 ml) and kept the samples for two hours. After that the centrifugation were performed at 5000 rpm for 10 minutes in micro-centrifuge tubes. The supernatant was discarded and leaving a pellet that containing diatom. Washing twice the pellet with double distilled water centrifuge it in order to remove the traces of acid.[31]

1.6. Extraction of diatoms from tissue and body organs

Kidney, liver, lung, brain and bone marrow were taken in one-two gram quantity and placed in a sterilized glass beaker. Add 50 ml nitric acid and samples should be left overnight and after that boiling should be performed for half an hour. A fat layer found deposited at the top and then discard that layer. The centrifugation should be performed of remaining samples at 4000 rpm for 10 minutes. The supernatant should be discarded and washing of pellet were performed thrice with distilled water. Then prepare the microscopic slides, and they should be dried on hot plate. Use compound microscope for study or view the diatoms.

A comparative analysis was also performed in which sulfuric and nitric acid digestion at 90° C for overnight with 30% diluted H_2SO_4 maintaining overnight at room temperature. In the results of this study the first procedure of digestion (classic method) with sulfuric and nitric acid found aggressive with high destroyed diatom fragments whereas the other digestion procedure (with H_2SO_4) is less aggressive with no precipitates was observed.[28]

In the acid digestion method, there are some limitations and drawbacks due to the strong treatment of acids as the structure of diatoms may be destruct meanwhile an improved method were also developed that results good recovery and the ratio was defined as 3:1 of nitric acid to hydrochloric for the extraction of diatoms.[29–32]

2. Conclusion

The determination for the cause of death is difficult in drowning cases. Whereas forensic autopsy is necessary to find out the external and internal findings during investigation. Therefore, these findings can't conclude drowning as a cause of death without the diatom confirmatory test whether it is present or not and if present then the find out of species is also important. A forensic medical officer will collect samples (e.g., tissues and bones) for further forensic analysis in drowning related cases and the composing results of all examination can be accomplished and the diatom test is significant in drowning cases. In future some advanced methods and technology such as molecular biological techniques are incorporating and can be used for the recognition of diatoms.

3. Source of Funding

None.

4. Conflict of Interest

None.

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Cite this article: Chaudhary R, Dhingra V. Forensic significance in the cases of drowning deaths: An elaborative study. *IP Int J Forensic Med Toxicol Sci* 2021;6(4):122-126.