

Role of Diatom Test in Forensic Science for Examination of Drowning Cases

¹Ajay Kumar, ²Manoj Malik and ³Anita Kadian

^{1,2,3}Forensic Science laboratory, Madhuban, Karnal-132001, Haryana (India).

¹Department of Zoology, Kurukshetra University, Kurukshetra-136119, Haryana (India).

e-mail* - ajayindorakuk@yahoo.com

Abstract: Diatom analysis is a valuable tool in forensic science and it is useful in diagnosis of drowning cases. The basic principal of the “diatom test” in drowning is based on correlation between diatoms are present in the medium where the possible drowning took place and inhalation of water causes penetration of diatoms into the alveolar system and blood stream. These diatoms are deposit into the brain, kidneys and other organs. For solving of drowning cases, hard bones (sternum and clavical) as well as soft tissues (lungs and liver) of drowned bodies and sample of water in which possible drowning take place are usually sent to the Forensic Science Laboratories for detection of diatom. In present study, 7 drowning human cases are examined. Acid digestion test method on laboratory rats were used on the basic for evaluation of diatoms methods as supporting in forensic of drowning. Result revealed from examined of 7 human cases are suspected for drowning, four cases are positive (death due to drowning) while three cases are negative (death other than drowning).

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1. Introduction:

Gregory Popp (1904), became the first scientist to solve a court case where the geological make up of soils was used to secure a criminal conviction (Ruffell and McKinley, 2005). Nevertheless, it was not until 1975 that the first textbook on forensic geology was published (Murray and Tedrow, 1975) and there is still only a very limited dedicated literature in this field (Saferstein, 2001; Murray, 2004; Pye and Croft, 2004; Ruffell and McKinley, 2005; Ruffell, 2006). Many general texts on crime scene investigations make little or no mention of forensic geology (Saferstein, 2001) and there is a lack of knowledge in the legal profession and police forces of its potential (Pye and Croft, 2004; White, 2004). However, one geologically-based forensic technique that has become established in forensic science is i.e., “diatom test” for drowning cases (Peabody and Burgess, 1984; Pollanen, 1998; Cameron, 2004; Horton et al., 2006).

Drowning can be defined as death due to full or partial submersion in a fluid (Timperman, 1972; Krstic et al., 2002). The fundamental principal of the diatom test in investigation of drowning is based on the postulation that diatoms are present in the fluid where possible drowning took place and the inhalation of the fluid causes penetration of diatoms into the alveolar system and blood stream, and leading to their deposition into brain, kidneys and other organs (Krstic et al., 2002). If the victim was dead before the body was submerged, the transport of diatom cells to various organs is prevented because lack of circulation and water sample.

Diatom analysis can be of further use in Forensic Science through identifying the provenance of individuals, clothing or materials from sites of investigation (Peabody, 1980, 1999; Siver et al., 1994; Pollanen et al., 1997; Krstic et al., 2002; Cameron 2004). Where materials have been submerged or there has been contact with littoral or riparian sediment or vegetation, diatom analysis of sediments or other diatomaceous traces present on clothing or footwear can be used to identify the type of habitat (Cameron, 2004). Suspense among drowning cases increases day by day, hence the present study revealed that how “diatom test” is a tools for examination of drowning cases.

2. Materials and Methods:

Water body sample, in which possible drowning took place and bone samples i.e., sternum, clavical and femur was sent in the Forensic Science Laboratory, Madhuban, Karnal, Haryana, India for detection of diatoms. Acid digestion method (Digincamillus et al., 2011) was used for digestion the hard bone sample i.e., sternum, clavical and femur and water sample in which possible drowning took place.

For detection of diatoms in water sample, it is centrifugation in three times on centrifuge (Research centrifuge-REMI) with 4000 rpm for 8 minutes in 100 ml of centrifuge tubes. The supernatant discarded and residue put on slides. The slide put on warm hot plate. Similarly way, detection of diatoms in bone sample i.e., sternum, clavical and femur, it is digested in const. HNO₃. Solution is

heated until its colored turned and finally cleared. Now samples put for cooling in whole night. It is also centrifugation on three times on centrifuge (Research centrifuge-REMI) at 4000 rpm for 10 minutes in 100 ml of centrifuge tubes. It is also again centrifuge with distilled water (use for washing of resultant suspension of silica diatoms) at 4000 rpm for 10 minutes in 100 ml of centrifuge tubes. The supernatant discarded and residue put on slide. The slide put on warm hot plate. Finally the slides mount with DPX and observe under the optimum 100X of microscope (LEICA-DMLB, Oil electron microscope). After detection the diatoms in both samples, a correlation of diatoms between the water sample in which possible drowning took place and bone samples i.e., sternum, clavical and femur was observed and examination of drowning cases clarify on the diatom species of both samples.

3. Results:

In case study I, a died body of adult female was found floating in a sewerage of a river. For examined the drowning case, the water sample and bone sample i.e., clavical and sternum was sent in the laboratory. After examination the diatoms i.e., *Coscinodiscus centrales*, *Anomoeoneis aphaerophora* was detected in water sample and *Coscinodiscus centrales* in bone sample. Hence the result will be positive and death due to drowning. Similarly way, in case study II, a died male body was found in a pond. Post mortem report examined that found death to be suspension. For examined the case, the water sample and bone sample i.e., clavical and sternum was sent in the laboratory. After examination found that diatoms i.e., *Coscinodiscus centrales*, *Naviculla caspidata* was detected in water sample and *Naviculla caspicuda* in bone sample. Hence death due to drowning. Also in case study III, the body of adult male was found bank of a Yamuna river. The died body sign shows that due to poison. Post mortem report examined that found death to be suspension. For examined the case, the water sample and bone sample i.e., clavical and sternum was sent in the laboratory. After examination the diatoms i.e., *Naviculla radiosa*, *Ghomphonema clavatoides* was detected in water sample. However, diatoms could not be detected in bone sample. Hence the result clarify that death other than drowning (Table 1).

Also in case study IV, an adult male died body was found floating in the canal of a river. For examined the case, the water sample and bone sample i.e., clavical and sternum was sent in the laboratory. After examination the diatoms i.e., *Pinularia viridis*, *Tubularia fasciculata*, *Naviculla caspidata* was detected in water sample and diatoms *Pinularia viridis* could be detected in bone sample. Hence the

result will be positive and death due to drowning. Similarly way, in case study V, a died body of new born baby was found near a dirty drain. Like above for examined the case, the water sample and bone sample i.e., clavical and sternum was sent in the laboratory. After examination the diatoms i.e., *Tubularia fasciculata* was detected in water sample, however, diatoms could not be detected in bone sample. Hence the result will be negative and death other than drowning (Table 1).

Also in case study VI, a unknown body part (from pelvic to knee) was floating in the water canal. Post mortem report examined that found death to be suspension. For examined the case, the water sample and bone sample i.e., femur was sent in the laboratory. After examination the diatoms i.e., *Cymbella turgid*, *Pinularia viridis* was detected in water sample, however, diatoms could not be detected in bone sample. Hence the result clarify that death other than drowning. Finally, in case study VII, a adult male died body was found in the drain. For examined the case, the water sample and bone sample i.e., clavical and sternum was sent in the laboratory. After examination found that the diatoms i.e., *Navigulla frigless*, *Naviculla caspicuda*, *Cymbella tumeda* was detected in water sample and *Cymbella tumeda* in bone sample. Hence the result clarify that death due to drowning (Table 1).

4. Discussion:

All the controversies the reliable methods in examination of drowning, represent the main cause of accidental death in USA, the scientific on validity of the diatom test begin (Guy, 1861). Tyagi (1985) collected water samples from various water bodies like lakes, ponds, wells and drains in Delhi. Water samples were treated with conc. HCl acid and supernatant was discarded. Then addition of conc. H₂SO₄ turned it blackish (conc. H₂SO₄ charred present organic material). The supernatant was cooled and added with solid NaNO₃. Suspension was reheated until its color turned brown and finally cleared. Distilled water was used for the washing of the resultant suspension of silica diatom cells and residue was re-suspended in acetone. Pollanen (1998) also extracted diatoms from putative water samples using acid digestion method.

Thomas et al. (1961) and Timperman (1962) dissolved sternum bone marrow in a Kjeldahl flask containing 50 cc of nitric acid. After half an hour the yellow fluid turned transparent. This solution was allowed to cool and then centrifuged. The residue was put on the slide and examined under a microscope. But Pollanen et al. (1997) made a slight modification in this method by removing bone marrow (50 gm) from femur bone and put into a

boiling flask. Approximately, 50 ml. of concentrated nitric acid (analytical grade) was added and marrow - acid suspension was simmered on a hot plate for approximately 48 hours in a fume hood. After cooling suspension at room temperature it was centrifuged (200-300 g/30 min). The residue so obtained was then added in distilled water and again centrifuged. Then final supernatant was discarded and pellet containing nitric acid-resistant material was aspirated using a Pasteur pipette and put on a clean microscopic slide for examination.

Horton (2007) employed the "Diatom test" in two case study of drowning i.e., the body of a woman was found floating in the river and the died body of a boy was found face down in the pond. He examined in the drowning cases, the best correlation in water sample as well as bone sample. Horton et al., (2006) also examined the drowning cases on the bases of diatom test. In his study, a body was found face down in the pond and it was suggested that drowning took place in the pond. He collected samples for diatom analysis from four transects around the circumferences of the artificial pond, two samples of sediments from the center of pond to act as a control in the examination of diatom assemblages associated

with three living tissue samples. After the diatom test, it was examined that diatom was detected in all body samples. Hence the result will be positive and death due to drowning.

In the present, 7 numbers of human cases was examined. Among seven cases of drowning four was positive and three was negative. In positive human cases diatoms, *Coscinodiscus centrales*, *Anomoeoneis aphaerophora* in water sample and *Coscinodiscus centrales* in bone sample of case I, *Coscinodiscus centrales*, *Naviculla caspidata* in water sample and *Naviculla caspidata* in bone sample of case II, *Pinularia viridis*, *Tubularia fasciculate*, *Naviculla caspidata* in water sample and *Pinularia viridis* in bone sample of case IV and *Navigulla frigless*, *Naviculla caspicuda*, *Cymbella tumeda* in bone sample and *Navigulla frigless* in bone sample was detected. Hence it clarify that death due to drowning. Also diatoms, *Naviculla radiosa*, *Ghomphonema clavatooides* in water sample of case III, *Tubularia fasciculate* in water sample of case V and *Cymbella turgid*, *Punilaria viridis* of case VI was detected. But diatoms could not be detected in bone sample of cases III, IV and VI. It shows that death other than drowning.

Table 1. Diatoms in both samples for examination of drowning cases.

Case No.	Water sample	Bone sample	Diatom in water sample	Diatom in bone sample	Examination results
1	Water sample	Sternum/ clavical	<i>Coscinodiscus centrales</i> , <i>Anoboeoneis aphaerophora</i>	<i>Coscinodiscus centrales</i>	Positive
2	Water sample	Sternum/ Clavical	<i>Navicula caspidata</i> , <i>Coscinodiscus centrales</i>	<i>Navicula caspidata</i>	Positive
3	Water sample	Sternum/ Clavical	<i>Navicula radiosa</i> , <i>Gomphonema clavatooides</i>	Absent	Negative
4	Water sample	Sternum/ Clavical	<i>Pinularia viridis</i> , <i>Tubularia fasciculate</i> , <i>Navicula caspidata</i>	<i>Pinularia viridis</i>	Positive
5	Water sample	Sternum/ Clavical	<i>Tubularia fasciculate</i>	Absent	Negative
6	Water sample	Femur	<i>Cymbella turgid</i> , <i>Punilaria viridis</i>	Absent	Negative
7	Water sample	Sternum/ Clavical	<i>Navigula frigles</i> , <i>Navicula caspidata</i> , <i>Cymbella tumda</i>	<i>Cymbella tumda</i>	Positive

5. Conclusion:

The “diatom test” for drowning is one of the most often applied and studied application of diatom analysis in forensic investigations and become an established forensic technique because diatoms have many attributes that are applicable to forensic science. Diatom test also provide a record of environmental conditions via, diatoms relationship to water quality and aquatic habitat. The basic principal of the “diatom test” in drowning is based on inference that diatoms are present in the medium where the possible drowning took place and that the inhalation of water causes penetration of diatoms into the alveolar system and blood stream, and thus, their deposition into the brain, kidneys and other organs. The transfer function approach presented in this paper offers a quantitative method to provide an informal assessment of “reliability” of the “diatom test” through correlations between control samples and samples from organs and clothing. In our present cases study, a correlation is established between water sample in which present drowning took place and bone sample i.e., sternum, clavical and femur. It is also study that diatom test has large numbers of advantages in Forensic Science.

Correspondence author:

*Ajay Kumar

Department of Zoology, Kurukshetra University,
Kurukshetra-136119, Haryana (India).

Mobile no. 09813077684

e-mail* - ajayindorakuk@yahoo.com

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