DIATOMOLOGICAL STUDIES FROM THREE WATER BODIES OF JAIPUR

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ABSTRACT

In suspected drowning cases forensic pathologists are often asked whether the drowning was anti-mortem or post-mortem, and along with this an equally important question to be answered here is that if the death really took place at the same site from where the body was recovered. Here question arises about the exact site of drowning and in forensic investigations; it imparts necessity for the precise localization of site of drowning. The proposed study deals with the algological analysis that, in turn has recorded diatom type in three water bodies of Jaipur (Rajasthan) region. On the basis of this database, diatomological maps have been designed, which are supposed to be of immense help in diagnosis of suspected drowning cases occurring in this region.

Keywords: - diatoms, drowning, water body

INTRODUCTION

In forensic investigation, while solving drowning cases, diatoms can easily be detected in viscera of drowned body. Further matching of diatoms from both, bone marrow and drowning site can strengthen this supportive evidence and a positive conclusion can be drawn whether the person was living or not when drowning took place. Diatoms are abundant and diverse in aquatic habitats and this uniqueness makes them of forensically important in cases of suspected drowning. In the absence of other evidence, diatoms detected in the body tissues are the most reliable indicator of a drowning *(Kobayashi et al., 1993].* Even when there are only skeletal remains, diatoms can be detected in the bone marrow of drowned victims. In cases of suspected drownings, 20 diatoms per 100 microlitres of pellet from a 10 gm lung sample, or five complete diatoms from other organs, are normally required for a positive conclusion *(Ludes et al., 1994*).

Diatoms are divided into two groups based on overall symmetry of the cell walls; radially symmetrical forms are called "centric" diatoms, while bilaterally symmetrical forms are referred to as "pennate" diatoms. One remarkable aspect of these organisms is that they have cell walls made of glass (silicon dioxide), *(Round et al., 1990).* Diatoms can differ from one water body to another with response to local factors via mineral contents, temperature, water stratification and pH, *(Pollanen, 1996 and Chapman, 1973)*. As the qualitative and quantitative diatom analysis agrees well with aquatic conditions prevailing at certain site of drowning thus diatoms can also provide information about the general aquatic conditions at the site of drowning*, (Antti, 1991; Ludes et al., 1996; Pollanen, 1997; and Hurlimann et al., 2000). Burton et al. (1990)* reported significantly fewer taxa in the high alkaline sites. According to *Kwandrans (1993)* small numbers of species with lower diversity are found in the streams with low pH, and the streams closer to circumneutral pH have a higher diversity and greater number of species.

Patrick (1970) described that toxic pollutants may inhibit diatom reproduction, although they may not be killed immediately. His experiments show that when the pH of a stream is lowered from 7 to 5.5, the various diatom species are not killed, but fail to reproduce. Other factors like sensitivity to water quality changes *(Dixit et al., 1992),* impacts of pollution *(Patrick ,1968),* nutrient enrichment i.e. silicon *(Whitmore, 1989),* acidity/pH *(Whitmore, 1989; John, 1988)* sewage outflows *(Anderson et al., 1990; Yoshitake and Fukushima, 1990),* Salinity of water *(Pollanen, 1996)* and temperature *(Pollanen, 1996)* also effect growth of diatoms.

Sometimes problem regarding putative drowning site can arise when drowning is suspected somewhere else. Another case where drowned body is found on land and reference water samples taken at a site of submersion are not available. As the qualitative and quantitative diatom analysis agrees well with aquatic conditions prevailing at certain site of drowning. The algal community, particularly the diatoms, is especially useful in linking suspects and victims to crime scenes in or around fresh water. Diatom species are easily identified from the characteristic shape of the cells and unique refractive pattern caused by the silica in the cell wall *(Miller Coyle et al., 2001).* Thus diatoms can also provide information about the general aquatic conditions at the site of drowning and in these critical situations 'continuous water monitoring' of number and genera of diatoms at a site can be helpful in making a link between the drowned body and possible site of drowning, *Tyagi (1985); Ludes, et al. (1996 and 1999)*. More over a comparison between diatoms found in the tissue and the alga of water site also allows excluding the possibility of laboratory contaminated and air-inhaled diatoms before death.

A more elaborated and outstanding work related to associating victims with particular aquatic sites in suspected drowning cases was done by *Ludes et al* (1996) in France. Three aquatic sites (rivers) with high probability of drowning cases were chosen and monthly data base profiles of various diatom species were prepared. In 1999*, Ludes et al* utilized previously made database for

the 20 suspected drowning cases where site of drowning was unknown and 20 other cases where drowning sites were known. They opined that correlation of relative abundance of each taxon recovered in the lung samples with those found in the water samples may thus be a reliable method to indicate the site of drowning. ***Tyagi (1985)*** also conducted water monitoring system, and prepared database profiles of various diatoms of twenty-five water bodies of Delhi. Seasonal variations were also observed as diatoms experienced two-population maxima, one in spring and other in autumn but winter and summer seasons were having mostly fewer number of diatom flora. In another effort ***AGO et al. (2004)*** suggested that analysis of the putative drowning medium was essential for an accurate diatom test for drowning in Island like Yoronjima located in the open sea. There was considerable variation owing to the difference of the location, distance from the shore, depth and tide of sea in the winter season only. *Amphora, Cocconeis, Cymbella, Diploneis, Navicula* and *Nitzschia* were among the most common species and *Chaetoceros* and *Skeltonema* were restricted to mainland only. ***Natasha and Aleksej (2005)*** reported a criterion for differential determination of manner of death of dead bodies recovered from water. In cases where drowning site is not available or suspected, using diatomological maps of water bodies (including rare types of diatoms) possible site of drowning could be layout. ***Trent (2004)*** also supported affect of seasons on diatom population. Earlier, ***Pollanen et al. (1997) and Pollanen (1997)*** had studied seasonal number (quantitative) and species (qualitative) variations. ***Antti (1991); Pachar and Cameron (1992); Pollanen (1997); Hurlimann, et al. (2000)*** also favoured this study for the diagnosis of drowning cases where site is unknown. Observations taken during the summer season of year 2005, we have made an attempt to investigate weather these diatom species can really characterize a particular water body. A diatomologic database of the selected water bodies in the Jaipur is worked out and some rare types or site-specific diatoms have been identified and located.

MATERIAL AND METHOD

Collection and analysis of water samples:

The diatom samples were collected from the following three different water bodies of Jaipur:

1. *Jall Mahal (Lake)*
2. *Mavaath (small pond in front of Jaigarh fort)*
3. *Galta ji (a religious water bath)*

Table-1 showing Site descriptions

Name of the site	Type of site	Type of area	Time of sample collection
1. *JALL MAHAL*	*Pond*	Dry Hills of Rajasthan	July 2005
2. *MAVAATH*	*Pond*	Dry Hills	July 2005
3. *GALTA JI*	*Water Fall*	Dry Hills	July 2005

Since diatoms are very small in size, they cannot be collected individually; instead, collection from their natural habitat involves filtering of water samples or else collecting some of the substratum on which they are attached. Water samples along with some stones and vegetation were collected from all sides of the selected water bodies. These samples were collected in sterilized one liter plastic bottles. Formalin solution was added to these samples in order to avoid further growth of diatoms. These formalin-preserved samples were mixed well to break up attached diatoms and to suspend the algae.

Approximately 200 ml of water sample was transferred into an acid washed 250 ml glass beaker. Samples were added with 40-45 ml of concentrated nitric acid (HNO_3) and a pinch of Potassium dichromate $K_2Cr_2O_7$. Then samples were then allowed to stand undisturbed for two hours. These samples were transferred to properly marked plastic centrifuge tubes. Samples were then

centrifuged at 3000 rpm for 10 minutes. Each time supernatants were pipette out and only a pellet of diatom frustules remained at the bottom. This pellet was re-suspended in deionized water and again centrifuged at the previously set pattern. This process was repeated twice to ensure all traces of the acid were removed. Five permanent slides (for each side) were prepared using the 'cleaned' diatom frustule material. D.P.X. was used as a mounting medium. Other used chemicals were also tested for the presence of diatoms. Observed were taken under Light microscope at 1500X oil immersion and analyses were based on count of 50 or above diatoms per slide. Photographs of diatoms were captured using computerized software (Q-Win Leica). Studies made by ***Round et al. (1990)*** and ***Chapman and Chapman (1973)*** were consulted for the identification of diatoms. For this purpose online supporting material was also consulted (see references).

RESULTS AND DISCUSSION

It has been observed that diatom species vary in both number and type from one site to another. Galta ji is a water fall and water flows from one side to another. Large type of *Synedra and Melosira* along with some other varieties of diatoms i.e. *Rhoicosphenia and Cyclotella* were also reported. Diatoms were found in abundance in this water body. But in Jall mahal, water is stagnant and highly poluted with the industrial and city severage. Fewer types and number of diatoms were reported and *Cyclotella* was reported as the only dominating diatom. Mavath is a seasonal water pond, where elephants are brought to take bath. A great diversity of diatoms i.e. *Nitzschia, Diatoma, Navicula, Geissleria, Achcanthidium* were observed in this water body. Results also reveal that a few diatoms like *Melosira, Synedra, Rhoicosphenia, Geissleria* were restricted to a particular water body, while *Navicula, Cyclotella* were found in all three sites.

Table-2 showing some site specific diatoms

WATER BODY	SITE SPECIFICF DIATOMS
1. *GALTA JI*	*Melosira, Synedra, Rhoicosphenia, Cyclotella, Navicula*
2. *JALL MAHAL*	*Cyclotella, Navicula*
3. *MAVAATH*	*Nitzschia, Diatoma, Navicula, Geissleria, Achnanthidium, Cyclotella*

PICTURES OF SOME DIATOMS

Pic-1 Synedra and Melosira

Pic-2 Nitzschia

Pic-3 Navicula

Pic-4 Diatoma

CONCLUSIONS

Diatom microflora can play a crucial role in search of possible drowning site, in those cases where drowning site is unknown. This makes diatoms as an indicator of a particular drowning site. But results pertaining to such studies should be properly interpreted in context to post-mortem findings and the police investigations. The studies related to the water monitoring of diatoms should also be extended to other water bodies of Jaipur in order to complete the whole network of water bodies, so that more extensive reference database in case of suspected drownings can be provided. During the present investigation the data has been collected only in summer season. For the completion of this study, it is necessary to investigate the variations in diatoms diversity in other seasons too.

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