Identification for Death by Drowning from Teeth and Bones

I. Identification for Death by Drowning from Teeth

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From very long before, macroscopical findings in cadaver or histological, bacteriological, and chemical studies have been made on the identification for death by drowning, while we have frequently encountered the difficulty on it only by those means, because most of the drowned cadavers are floated on water surface in the advanced state of putrefaction, followed by discovery. On the other hand, by means of disorganization, an attempt available for the identification of death by drowning through detecting diatoms from lung and digestive tracts has been also established by Revenstorf[1](1904), Kasparek[2](1937), Buhtz et al.[3](1938), Incze et al.[4](1955), Mueller et al.[5](1949), and others.[6]

The merit of this method consists in the possibility of the identification for death by drowning irrespective of a fresh or old cadaver, for diatoms are not nearly affected by acids and bases.

Tomonaga[7][8] (1938) is the first in Japan who applied it to the identification for death by drowning. Tomonaga and his co-workers[9-16] noticed the excellence of the disorganization method (Diatom method) and have improved it. In present, it has been recommended as the most suitable and reliable method for the identification of dead cause and widely used in the fields of criminal investigation and medico-legal judgement.

The author has been as yet reported the method of identification for death by drowning from teeth and bones. In present paper, it, especially from teeth is dealed with.

MATERIALS AND METHODS

In this study, disorganization was carried out on a sand dish according to Tomonaga's technique.[9] Tooth was well washed with distilled water and decalcified with diluted hydrochloric acid for 5–10 hours. Subsequently, after washed again with distilled water or resected on one's surface with a surgeon's knife, one was transferred into a Kjeldahl flask and 2–4 ml of huming nitric acid was added. The Kjeldahl flask was heated on a sand dish until tooth had been completely

destroyed and solution cleared up. After cooled at room temperature, solution transferred into 50 ml centrifuged tube and centrifuged out. Supernatant was removed through a suction pump and sediment centrifugally washed with distilled water. Washing procedure was repeated several times so as to remove acid. Subsequently, this solution was transferred into a spitz glass and centrifuged out, and then, sediment was put on a slide glass and observed microscopically. Of course, the used flask, test tube, and distilled water etc. were checked being free from diatoms.

RESULTS

1) Detection of diatoms from teeth of non-drowned cadaver

A cadaver (38-yr-old woman died of acute pneumonia) was subjected to the study on whether diatoms are detected from teeth of non-drowned cadaver or not. First of all, with the object of knowing how many diatoms are contained in teeth of non-drowned cadaver, tooth was immediately decalcified and disorganized with acids without any pretreatment after its extraction. As indicated in Table 1, in 3 out of 8 cases, diatoms were not found at all. There were a few diatoms in each of 5 cases and only 4 diatoms in the greatest. Subsequently, after tooth was decalcified with diluted hydrochloric acid for 5 hrs, it was taken out, washed well with distilled water, and then allowed to disorganize. At the same time, decalcified solution was disorganized separate from tooth. As the result is given in Table 2, diatoms were not found in the least in 6 out of 10 cases. The numerals enclosed with parentheses in the table show the number of diatoms detected in decalcified solution.

In the following study, after decalcification, tooth was resected on its surface with a surgeon's knife and then, disorganization was carried out. As seen in Table 3, there were no diatoms in the least in 8 among 10 cases. Furthermore, these results were almost in agreement with those of other non-drowned cadavers.

In order to know "how many will diatoms adhered be able to remove" by repeating of washing with distilled water after tooth is contaminated in water containing numerous diatoms, the following studies were performed. After plunged into water containing 32,440 diatoms per 1 ml for three days, tooth was taken out, washed well with distilled water, and subjected to decalcification and disorganization. In the same way, another tooth was treated in water containing 53,820 diatoms per 1 ml for three days. In these decalcified solution, diatoms found 2 in the former and 5 in the latter, respectively, but not detected in both disorganized teeth.

Moreover, the interpretation on whether diatoms that are occationally found from teeth of non-drowned cadaver, exist in their dental pulps from the first or enter them through aberration under the routine process will be in detail described in

Table 1. Detection of Diatoms from Teeth of Non-drowned Cadaver (Tooth was immediately decalcified and disorganized without any pretreatment after extraction)

	⌊1	⌊2	⌊4	⌊5	6⌋	⌈2	⌈1	⌈6
Weight (g)	1.1	0.8	1.1	1.1	2.2	0.7	0.6	1.8
Fragilaria	2	1				2		
Mel. distans						2		1
Navicula		1						1
Epithemia				1				
Total	2	2	0	1	0	4	0	2

Table 2. Detection of Diatoms from Teeth of Non-drowned Cadaver (Tooth was washed with distilled water after decalcification)

	⌊6	⌊7	⌊1	2⌋	3⌋	4⌋	8⌋	⌈1	⌈7	2⌋
Weight (g)	2.2	2.0	1.0	0.8	1.2	1.2	1.4	0.5	2.0	0.7
Fragilaria		2	1	(3)						
Mel. distans		(1)	1							
Navicula			1							1
Coscinodiscus		1							1	
Total	0	3(1)	3	0(3)	0	0	0	0	1	1

( ) : in decalcified solution

Table 3. Detectiom of Diatoms from Teeth of Non-drowned Cadaver (Tooth was resected with knife after washing and decalcification)

	⌊3	⌊8	5⌋	7⌋	⌈3	⌈5	⌈6	⌈8	3⌋	4⌋
Weight (g)	1.2	1.8	1.1	2.1	1.2	1.2	2.1	2.1	1.0	1.0
Fragilaria								1		
Navicula							1			
Coscinodiscus							1			
Total	0	0	0	0	0	0	2	1	0	0

the discussion latter. However, diatoms detected from teeth of non-drowned cadaver all may be thought to enter them through aberration during disorganization and preparation for microscopy, because they can not be in general detected from non-drowned cadaver. Therefore, in the case that only a few diatoms are found from each of teeth of non-drowned cadaver, there will be within the bounds of possibility into which they are introduced through aberration.

2) Detection of diatoms from teeth of drowned cadaver

Whether or not what number of diatoms can be detected from each of teeth of drowned cadaver was investigated and the result was compared with those obtained from other tissues such as lung, liver, and kidney of the identical cadaver.

Case 1 is a drowned cadaver who was discovered in the harbor near a wharf and a great number of diatoms, the majority in the species of *Fragilaria* and *Navicula* were found in his lung. From digestive tracts, liver, and kidney, the species of diatoms, *Fragilaria* and *Navicula* were detected. Accordingly, the dead cause was identified to be death by drowning. In addition, as shown in Table 4, the same species of diatoms as those detected in lung were found in considerable numbers from teeth. There were 22.5 to 28.5 diatoms per 1 g of left incisor and 88.5 to 90 diatoms per 1 g of right one. Furthermore, the number of diatoms per 1 g of each of tissues such as lung, liver, kidney, and stomach and duodenum contents is illustrated in Table 5. Diatoms were very great in lung, and stomach and duodenum contents, but very few in liver and kidney.

Table 4. Detection of Diatoms from Teeth of Drowned Cadaver
(1st case: a 36-yr-old male)

	$\overline{1}$	$\overline{2}$	$\underline{1}$	$\underline{2}$
Weight (g)	1.2	0.8	1.3	1.0
Fragilaria	20	12	69	46
Navicula	3	6	25	15
Mel. distans	2	5	9	10
Coscinodiscus	1		4	5
Gomphonema	1		6	5
Mel. varians				6
Skeletonema			2	3
Total	27	23	115	90
per 1 g	22.5	28.8	88.5	90.0

Table 5. Detection of Diatoms from Various Organs in 1st Case

Material	The number of diatoms
lung	165.4
liver	0.87
l-kidney	0.86
r-kidney	0.78
stomach	1047.3
duodenum	928.0

(Showing the number of diatoms per 1 g)

Case 2 and 3 both are drowned cadavers who fell into water on account of drunkness and died. In case 2, the number of 18.2 to 32.5 diatoms per 1 g of each of teeth was found (Table 6). As the result obtained from other tissues of the identical cadaver is given in Table 7, diatoms were very great in lung and stomach contents, but very few in liver, kidney, bile, and urine. In case 3, diatoms were within the range of 11.0 to 19.0 per 1 g of each of teeth and the variance in the number of diatoms among teeth was very slight as indicated Table 8. Further, as shown in Table 9, lots of diatoms were found in lung, and stomach and duodenum contents, but a few diatoms in another tissues. Judging from the

results above mentioned, diatoms were always detected from each of teeth of drowned cadavers and diatoms found in tooth were very great compared with those from organs of the great circulatory system such as liver and kidney etc. Moreover, even in teeth of the identical cadaver, the number of diatoms varied considerably among teeth.

Table 6. Detection of Diatoms from Teeth of Drowned Cadaver (2nd case: a 57-yr-old male)

	⌈2	⌈6
Weight (g)	0.4	1.1
Mel. distans	5	5
Fragilaria	1	5
Navicula	3	3
Coscinodiscus	4	2
Nitzschia		2
Cymbella		2
Skeletonema		1
Total	13	20
per 1 g	32.5	18.2

Table 7. Detection of Diatoms from Various Organs in 2nd Case

Material	The number of diatoms
lung	122.6
liver	0.6
l-kidney	1.03
r-kidney	0.39
urine	0.2
bile	0.57
stomach	89.5
duodenum	7.0

(Showing the number of diatoms per 1 g)

Table 8. Detection of Diatoms from Teeth of Drowned Cadaver (3rd case: a 31-yr-old male)

	⌈3	⌈4	⌈5	3⌋	4⌋
Weight (g)	1.0	0.8	0.8	1.0	1.0
Mel. distans	5	6	6	8	7
Fragilaria	4	3	3	1	4
Navicula	5	2	3	1	5
Coscinodiscus	1	1	1	1	2
Cyclotella					1
Total	15	12	13	11	19
per 1 g	15	15	16.3	11	19

Table 9. Detection of Diatoms from Various Organs in 3rd Case

Material	The number of diatoms
lung	100.7
liver	0.42
l-kidney	0.34
r-kidney	0.14
stomach	506.8
duodenum	85.0

(Showing the number of diatoms per 1 g)

3) Detection of diatoms from teeth of skeletonized cadaver.

The following three cases are cadavers that were found in water in the completely skeletonized state and their dead causes were assumed to be death by drowning by means of the findings through disorganization method.

Case 4 is a male cadaver who was discovered as being skeletonized in his car

under water after lapse of one year since his missing. According to the report of police investigators, he called at the house of his friend on his way to home from his office, drunk there, and has been missing since he left the house at about 9 p.m. in defiance of his friend's remonstrance. When he was discovered, body had been already skeletonized completely and soft tissues were nothing. A lot of diatoms, mainly in the species of those lived in sea water were found from premolar and molar tooth, and the same species of diatoms as those from teeth were detected from ribs. Therefore, the dead cause was assumed to be death by drowning (Table 10).

Table 10. Detection of Diatoms from Teeth of Skeletonized Cadaver (4th Case)

	⌊4	⌊6
Weight (g)	1.3	2.2
Nitzschia	23	75
Coscinodiscus	11	23
Chaetoceros	5	14
Rhizosolenia	2	12
Cyclotella	5	14
Fragilaria	3	6
Thalassiothrix		8
Skeletonema		5
Bidulphia		5
Diploneis		4
other diatoms	1	24
Total	50	190
per 1 g	38.5	86.4

Taking various findings and report into consideration, because he drunk sake, fell into sea water from a quay-wall through mistake in the course of operation of his car, and sank under sea bottom of 10 meters in depth, he was assumed not to be able to escape from his car and to be death by drowning.

Case 5 is a cadaver who was discovered in an irrigation pond. Body was utterly skeletonized and bones were kept separate one from another. Ten teeth were disorganized and the result is shown in Table 11. A great number of diatoms were found from teeth of ⌊6, 6⌋ and 7⌋, in which tooth of 7⌋ had 2,753 diatoms per 1 g. But,diatoms in tooth of 2⌋ were relatively few compared with those from the formers and calculated as 56 per 1 g. More, the composition of diatoms detected from teeth was nearly in agreement one another and in the species, *Navicula* was the greatest, followed by *Fragilaria*. From ribs, the same species of diatoms as those from teeth were found. In the case that lots of diatoms are

Table 11. Detection of Diatoms from Teeth of Skeletonized Cadaver (5th Case)

	⌊2	⌊4	⌊6	⌊7	2⌋	4⌋	6⌋	7⌋	⌈6	⌈6⌉
Weight (g)	1.0	1.1	2.2	2.2	1.1	1.3	2.2	2.2	2.3	2.3
Navicula	100	873	2068	1027	24	112	1723	2493	548	259
Fragilaria	48	43	976	629	17	69	864	1714	338	170
Pinnularia	22	22	324	77	13	25	289	440	114	39
Nitzschia	6	3	308	67	1	6	113	342	28	10
Gomphonema	2	1	100	139	3	7	98	264	43	22
Cymbella	7	8	135	60		5	90	301	29	22
Cyclotella	15	1	50	60	1	15	82	98	46	33
Mel. distans	1	5	54	37		13	35	122	38	37
other diatoms	7	9	228	80	3	6	113	283	39	28
Total	208	965	4243	2176	62	258	3407	6057	1223	620
per 1 g	208	877	1929	989	56	198	1549	2753	532	270

found from various organs as in an instance of this cadaver, a great number of diatoms must be contained without fail in drowning water. Then the author drew water from several places near the scene and carried out the investigation for diatoms in it. There were 2,390 diatoms per 1 ml of water that was drawn in October, the same month as this cadaver was discovered, but in June, 4,470 to 15,000 diatoms per 1 ml. On the other hand, the composition of diatoms was as follows usually: the species of *Navicula* was the greatest, followed by that of *Fragilaria*.

Case 6 is a skeletonized skull that was caught in her net when a fishing boat had been in the course of her operation in East Chaina Sea. Five teeth remained were subjected to disorganization and the result is indicated in Table 12. There were 7.2 to 11.1 diatoms per 1 g of each of teeth and the variance in the number of diatoms detected was very slight among teeth.

Table 12. Detection of Diatoms from Teeth of Skeletonized Cadaver (6th Case)

	⌊5	⌊6	4⌋	5⌋	6⌋
Weight (g)	1.8	2.0	1.2	1.8	2.0
Nitzschia	6	8	6	11	7
Thalassiothrix	5	8	6	9	5
Rhizosolenia	2				4
Skeletonema		3			
Diploneis					1
Total	13	19	12	20	17
per 1 g	7.2	9.5	10.0	11.1	8.5

According to the results through disorganization about teeth of skeletonized cadaver, their dead causes each was assumed to be death by drowning. Even in teeth of the identical cadaver, there were a tooth containing lots of diatoms and on the contrary, one having only a few diatoms. The variation was demonstrated not to be due to the distinction of tooth, such as incisor or molar tooth, tooth on upper jaw or one on lower jaw, and left tooth or right one.

DISCUSSION

On the identification for dead causes of cadavers discovered in water, the author has always experienced that the number of diatoms detected from organs of the great circulatory system is very few as compared with those numbers from lung and digestive tracts. When cadavers would be thrown into water, water maight easily enter lungs and stomach, of course, in case of drowned cadaver and even in non-drowned cadaver by slight water-pressure.[10] In addition, diatoms derived from diet, mainly from drinking water are always contained in digestive tracts before death. Accordingly, from thus reasons above mentioned, it is impossible to identify the dead cause as death by drowning only on the basis of a fact that diatoms were found from lung and digestive tracts by disorganization. For deciding the dead cause as death by drowning, diatoms must be verified not only in lung and digestive tracts, but also in organs of the great circulatory system. But, diatoms detected from liver and kidney area few even in drowned cadaver. In the case that postmortem time lapsed very long and lots of injuries are given by fishes and tiny marine animals after death, because varions organs had been already disintegrated and vanished away, it is impossible for soft tissues to carry out disorganization. In such a case, there is nothing of other methods except testing for hard tissues such as bones and teeth, assuming whether or not the dead cause is death by drowning.

The identification for death by drowning from bones out of hard tissues has been already reported by Mikami et al.[17] (1959), Tamáska[18] (1961), Thomas et al.[19] (1961), Okuyama[20] (1961), Timperman[21] (1962), Furuno et al.[11] (1963) and Koseki[22] (1969), but the report from teeth not yet seen. Teeth may be considered to be contaminated with diatoms derived from diet, mainly from drinking water. Contrary to expectation, the experimental result showed that such diatoms were a few in the average, 4 in the greatest, and not contained in certain tooth at all. Diatoms adhered to tooth are usually removed by washing with distilled water, but if the surface of tooth is resected with a surgeon's knife, the further good result will be obtained. Even in tooth resected with a surgeon's knife, in 2 out of 10 cases, 1–2 diatoms were found. In such a case, it is quite impossible to distinguish whether diatoms exist in dental pulp from the first or enter it through aberration in the course of laboratory work. Since diatoms are contained "of

course" in tap water and in air, the chance which when disorganization is carried out, diatoms enter teeth due to aberration may be considered as follows: namely, being led from water at the time apparatus for testing is washed, from distilled water and reagents under the routine process of disorganization, and from air when washing procedure is centrifugally repeated several times in order to remove the remaining acid. Judging from the reasons above mentioned and author's experiences[11–15][23–25] concerned in the identification for drowned cadaver for long time, diatoms which were detected even in tooth that had been washed or resected on its surface after decalcification were assumed, not because they originally exist in tooth, but because they entered it through aberration (shown in Table 2 and 3).

In recent year, some investigators[26–30] have entertained a question about whether disorganization is suitable for the identification of death by drowning or not, because diatoms are detected in non-drowned cadavers. Spitz[26–28] has emphasized that diatoms derived from diet pass through intestine and attain organs of the great circulatory system, and in the further investigation for rabbits administrated with diatom earth orally, 92 % of the treated and 49 % of the untreated free from diatom earth, had diatoms in their organs of the great circulatory system. In contrast to that, Mueller[31] has reported that diatoms detected from non-drowned cadaver are likely to enter body through aberration, for diatoms were been detected only from one case having symptom for cor pulmonale, out of 30 cases of non-drowned cadavers. Therefore, he pressumed that spitz would take diatoms introduced through aberration for original ones. Reh[30] has studied diatoms in livers, kidney, and bone marrow of non-drowned cadavers with Kämper and reported that diatoms were detected in 18 among 20 cases, whereas diatoms were approximately 3 in the greatest. Janitzki[32] has detected 0.01–0.03 diatoms per 1 g of kidney of one case and lungs of 2 cases among 8 cases of non-drowned cadavers. Timperman[33] has also reported as follow: there were nothing of diatoms in 13 among 17 cases of non-drowned cadavers and possitive about diatoms in 4 cases, in which 2 cases were late exhumations of bodies recovered from wooden offins; and there were negative about diatoms in livers of 11 out of 12 cases of non-drowned cadavers and possitive in one case, which was again an exhumation. Yamamoto[34] has stated not to be able to deny the possibility that diatoms may enter organs through aberration, because diatoms could not be detected from another tissues of other cases except muscle and bone marrow of each one among 40 cases of non-drowned animals. Suyama[35] has reported that he studied diatoms on 10 cases of non-drowned cadavers and detected 4–17 diatoms or their fragments per 5 g of lung ; and these diatoms were inhaled together with dirt and spat out outwards body within relatively short time. Yamashita[16] has pointed out that through animal subjects, it is impossible for diatoms derived from diet, to enter organs of the great circulatory system after absorption and further, diatoms detected from livers of non-drowned animals be due to the aberration of diatoms in the

course of routine process of disorganization. Mueller has said that if apparatus is used after washing well, it would be impossible completely to remove diatoms introduced through aberration. Tomonaga has adviced to have to pay attention lest apparatus for disorganization should be contaminated with diatoms. Yamashita has stated the similar following views : when organs having lots of diatoms are subjected to disorganization, diatoms are always remained in the flask and these diatoms are further detected in it in spite of repeating of washing. And, if apparatus is washed as well as possible, it would be impossible utterly to deprive it of diatoms. Suyama has reported that on lack or mastery in skill, the findings concerning diatoms are clearly different between both and such a discrepancy is due to imcomplete washing and contamination at the time specimens are taken. Koseki[36] looked the direct cause leading diatoms to non-drowned cadaver upon contamination in air, reagents, and fixing solutions and the lack of skill in the routine process of disorganization.

According to disorganization of 3 cases of drowned cadavers, lots of diatoms were found in each of 3 cases. Moreover, over all these cases, the number of diatoms detected from tooth was compared with those from other tissues of the identical cadaver. Diatoms in each of teeth were more fewer than those from lung and digestive tracts, but greater than those from liver and kidney, and far great as compared with those from tooth of non-drowned cadaver, which are introduced through aberration.

In 3 cases of skeletonized cadavers, the results from their teeth were well in agreement with those from drowned cadaver. Consequently, their dead causes were assumed to be death by drowning.

So far as teeth are decalcified with diluted hydrochloric acid and huming nitric acid added, heated strongly, they are completely disorganized. Teeth are a most suitable material for detecting diatoms, because of extracting them from cadavers easily and nothing of putrefaction.

SUMMARY

For teeth of drowned and non-drowned cadavers, disorganization was carried out and the results were as follow ;

Diatoms were detected from all their teeth in the drowned cases and the number of diatoms per 1 g of each of the teeth was greater than those from organs of the great circulatory system of the identical cadaver. But, in teeth of non-drowned cadaver, diatoms could not be detected. We must take account of diatoms introduced through aberration for assuming whether the dead cause is death by drowning or not, especially in the case that only few diatoms are detected and the greatest possible many teeth should be used for disorganization.

In 3 cases of skeletonized cadavers that were discovered in water, a great number

of diatoms were found from their teeth in all cases. Therefore, their dead cause were assumed to be death by drowning.

If teeth are treated huming nitric acid after decalcification with diluted hydrochloric acid, followed by strong heating, they are disorganized without difficulty.

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