

## The Distribution of Diatoms in Yoronjima and Application of the Diatom Test for the Diagnosis of Death by Drowning in Open Sea Islands

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### Abstract

The distribution of diatoms in Yoronjima was investigated as a model study to evaluate the application of diatom testing for the diagnosis of death by drowning off islands located in the open sea. Sea water samples were collected at 10 sites off Yoronjima. The numbers of diatom varied owing to the difference of location, the distance from shore, the depth of sea and the tide. These results suggest that analysis of the putative drowning medium is essential for an accurate diatom test for drowning in islands like Yoronjima located in the open sea.

**Key words:** Legal medicine, Drowning, Diatom test, Open sea island, Yoronjima

### Introduction

Diatom testing is the most reliable method to diagnose death by drowning. The diatom test for drowning is based on inhalation of diatom-laden water in the alveolus and embolization to internal organs<sup>1)</sup>. If diatoms are detected from organs such as the liver, spleen and kidneys of cadavers, the diagnosis of drowning is confirmed. Diatoms have silica-based frustules which are resistant to acid digestion so that the recovery of diatoms from a cadaver is possible. Diatoms proliferate broadly and abundantly distribute all the year in rivers, lakes, ponds and the sea, but their numbers decrease rapidly in the open sea<sup>2)</sup>. Therefore, in the case of drowning at sea, there is the possibility of getting a false negative result by diatom testing. In this study we investigated the distribution of diatoms in Yoronjima as a model study to evaluate the application of the diatom test for the diagnosis of drowning off islands located in the open sea.

### Materials and methods

As shown in Fig. 1, Yoronjima is situated in the most southwestern part of Kagoshima Prefecture. About 6,000 people live in Yoronjima. The shoreline is 23.65 km and the area of the island is 20.49 km<sup>2</sup>. Sea water samples

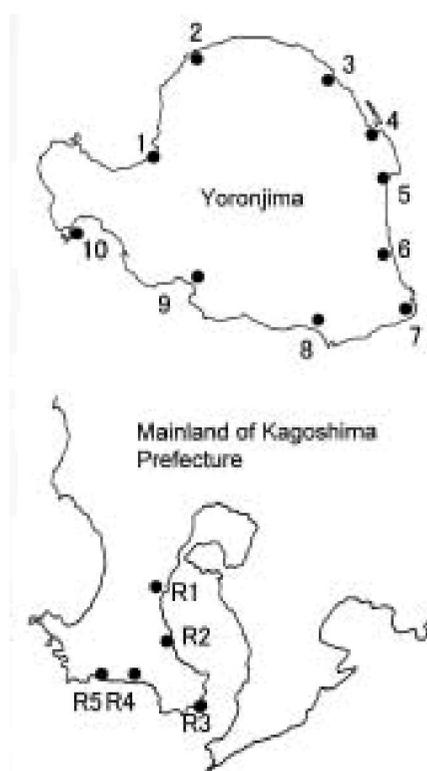


Fig. 1. Map of the sites collected sea-water samples in Yoronjima and the mainland of Kagoshima Prefecture. 1: Tyabana (Harbor), 2: Ukachi (Beach), 3: Kurohana (Beach), 4: Minata (Beach), 5: Funakura (Beach), 6: Oganeku (Beach), 7: Akasaki (Harbor), 8: Maehama (Beach), 9: Hakibina (Beach), 10: Yoron/Tomori (Harbor), R1: Taniyama (Harbor), R2: Nukumi (Beach), R3: Yamagawa (Harbor), R4: Ei (Harbor), R5: Makurazaki (Harbor).

were collected in winter at 10 sites off Yoronjima and 5 sites of the mainland of Kagoshima Prefecture. The sea water of 200 mL was collected from the surface and the bottom of the shore and from the surface several ten meters off-shore at every site. Sea water was also collected from the surface several hundred meters off-shore at four sites, Tyabana (1), Oganeku (6), Hakibina (9) and Yoron, originally called Tomori (10). In Minata (4) and Funakura (5), the samples of sea water were collected at high tide and at ebb tide in spring tide. The portions (1 mL, 10 mL and/or 30 mL) of the collected sea water were concentrated to 0.2 mL by centrifugation using a table-top centrifuge at 3,200 rpm for 10 minutes with a 10 mL centrifugation tube. The concentrated sea water was incubated with about 2 mL of fuming nitric acid for 30 minutes in boiling water. The sea water treated with acid was washed twice with pure water and twice with pure ethanol by centrifugation at 3,200 rpm for 10 minutes. The supernatant was transferred on a cover glass heated on a hot plate. The residue was completely dried and was attached with Mount Media (Wako Pure Chemicals) on a slide glass. Diatoms were examined with an optical microscope at a magnification of 400-fold. The numbers of diatoms on all fields of the prepared specimen were counted and the density of diatoms was calculated as the number of diatoms/1 mL of sea water. Each piece of decomposed diatom was counted as one. The first hundred diatoms observed on a prepared specimen were classified and the classified numbers were used to represent the percentage of diatom species. Pieces of fragmented diatoms or diatoms unclassified because of being extremely small and/or lying on its girdle were classified as 'others'.

## Results and discussion

### *Numbers of diatoms*

As shown in Table 1, the densities of diatom ranged from 3.3 to 2395 diatoms/mL in Yoronjima and from 16.5 to 753.0 diatoms/mL in the mainland of Kagoshima Prefecture. The densities of diatom in sea water from the surface of shore (Surface in Table 1) at Ukachi (2), Akasaki (7) and Yoron (10) in Yoronjima were lower than the minimum value in the mainland of Kagoshima Prefecture, 17.3 diatoms/mL at Makurazaki (R5). In the case of the bottom of shore (Bottom in Table 1), those values at Ukachi, Kurohana (3) and Yoron were

lower than the minimum value in the mainland of Kagoshima Prefecture, 16.5 diatoms/mL at Yamagawa (R3). The values on the surface tens of meters off-shore (Offshore in Table 1) at Ukachi, Minata (4), Akasaki and Yoron were lower than the minimum value in the mainland of Kagoshima Prefecture, 20.5 diatoms/mL at Ei (R4). Ukachi and Yoron being the low densities of diatom locate on the northernmost and on the westernmost part of in Yoronjima, respectively (Fig. 1).

The densities in sea water from surface several hundred meters off-shore in four sites tested were extremely low, Tyabana (1): 1.5, Oganeku (6): 0.8, Hakibina (9): 0.5 and Yoron (10): 2.9 diatoms/mL.

The densities of diatom in Minata (4) were higher on ebb tide than on high tide. On the other hand, the densities in Funakura (5) were higher in the surface and the bottom at the shore on high tide.

The numbers of diatom varied considerably owing to the difference of location, the distance from shore, the depth of sea and the tide. These results suggest that a careful analysis of the putative drowning medium is essential for an accurate diatom test for drowning off islands like Yoronjima in the open sea.

### *Species of diatoms*

Species of diatoms in the samples are shown in Table 1. The 10 sites of Yoronjima were similar to one another. *Amphora*, *Cocconeis*, *Cymbella*, *Diploneis*, *Navicula* and *Nitzschia* were usually observed in Yoronjima samples. In contrast, *Chaetoceros*, *Navicula*, *Nitzschia* and *Skeletonema* were usually found in samples off the mainland of Kagoshima. *Chaetoceros* and *Skeletonema* mainly existed in the mainland and were seldom detected in Yoronjima samples.

The 'others' in the Table 1 shared above 50% in every site except Taniyama. The 'others' contains small diatoms (Fig. 2). Small sized diatoms penetrate the alveolus-capillary barrier more easily<sup>3-5</sup>, so a high percentage of 'others' is advantageous for diatom testing to confirm drowning.

Seasonal variations in the number (quantitative) and species (qualitative) of diatom were reported previously<sup>3,4,6</sup>. Yoronjima in the subtropical zone has roughly two seasons, a long summer and a short winter. This study was carried out on sea water samples

Table 1. Density and species of diatoms

Samples	Place names*	Parts	Density (Nos/ml)	Diatoms																			
				Species** (%)																			
				Amp	Ast	Bac	Cam	Cha	Coc	Cos	Cym	Dic	Dip	Fra	Gom	Gra	Mel	Nav	Nit	Ske	Rare****	Others****	
Tyabana (1)	Surface		35.6	6					6	1	3		1	2			7	4	7				63
	Bottom		43.2	2	2				5		2		3	1	3				2	6			74
	Offshore		28.0	5	1				5		4		3	1					3	5			73
Ukachi (2)	Surface		6.4	4		1			5		2		2	2	1	1			7	2			73
	Bottom		10.2	5					6		3		11						1	12			62
	Offshore		3.5	1	2				2		4		2	4					6	5		1	73
Kurohana (3)	Surface		107.6	2					23		1		2						15	2			55
	Bottom		19.2						17		2		2						12	8			59
	Offshore		80.3	3					12		2		4			1			9	4			65
Minata (4) (high tide)	Surface		38.0	2	2				4		2		5	1	1				2	6		1	74
	Bottom		21.1	7	3		1		12		4		1	2		3			6	4			57
	Offshore		12.8	3	1		2		15		4	1	3	1	1	1			5	3		2	58
Minata (4) (ebb tide)	Surface		62.7	2	1	1			8		2	1	1			9			1	6			68
	Bottom		45.9	3	6				5		2		1	2		1			3	11			66
	Offshore		52.0	4	2				9		2	1				4			2	4			72
Funakura (5) (High tide)	Surface		193.9	6	4		1		14		2		2	2	4				1	1		1	62
	Bottom		836.0	5					13		7		3		3				1	8			60
	Offshore		175.4	6			1		11		3		2	3					1	3			70
Funakura (5) (Ebb tide)	Surface		151.6	5	5				13		2		4			1			1	7			62
	Bottom		106.0	10	2				17		3		3	2					5	2			56
	Offshore		237.9	8					7		5		1						2	2		1	74
Oganekuni (6)	Surface		54.0	8	6				7		1		3	5		3			1	6			60
	Bottom		46.8	6					7	1	2		7	6					2	6			63
	Offshore		37.5	5					7		4		12	3						2		1	66
Akasaki (7)	Surface		14.9	6	5				9		2		4						1	4			69
	Bottom		2395.0	3	3				5				1						2	3			83
	Offshore		7.7	3	3				6		2		2	2		3			1	4			74
Maehama (8)	Surface		38.6	3	3				8		1		2	3					1	1			78
	Bottom		213.0	1	2				10				4						6	8			69
	Offshore		62.5	4	3				5				3						4	8			73
Hakibina (9)	Surface		68.7	2					3		1		1	2			1	4	6			1	79
	Bottom		119.0	2					5				1						11	2			79
	Offshore		58.7	2	3				11		1		2						3	6			72
Yoron (10)	Surface		14.7	1	2				6		1	2		1	2	1			7	2			75
	Bottom		3.3	3		1		1	7	1			1	1					2	6			77
	Offshore		4.1	4				10	8		1	2		1		1			2	5			66
Taniyama (R1)	Surface		412.0					1	2		1		1						1		75		19
	Bottom		239.0																3		81		15
	Offshore		753.0							1									1	1	84		13
Nukumi (R2)	Surface		203.6	4					4					2					5	4	5	1	75
	Bottom		185.7	1					4				1						4	2	8	1	75
	Offshore		100.6						4	3		5		2	1	1			5	3	4	1	71
Yamagawa (R3) (Outside of a breakwater)	Surface		30.2	2				24	1	1				3					1		15		53
	Bottom		16.5					6		2	1									1	5		85
	Offshore		35.4		1			20		6						1			1		21		50
Yamagawa (R3) (Inside of a breakwater)	Surface		31.8					26		1										1	14		58
	Bottom		47.1					34	1	2										1	7		55
	Offshore		36.4					36		1											12		51
Ei (R4)	Surface		27.5	1	4	2		2	1				2	2								1	85
	Bottom		365.0		12				5										2	5		1	75
	Offshore		20.5	2	3			2	6											4		1	82
Makurazaki (R5)	Surface		17.2	3		1	2	17	1					2					1	10		1	62
	Bottom		23.8	2	2	3		18						1						9	1	1	63
	Offshore		59.8			2		30						1					1	5		1	60

\* Numbers in parentheses correspond with those in Figure 1.

\*\* Amp: *Amphora*, Ast: *Asterionella*, Bac: *Bacteriastrum*, Cam: *Camptylodiscus*, Cha: *Chaetoceros*, Coc: *Cocconeis*, Cos: *Coscinodiscus*, Cym: *Cymbella*,

Dic: *Dictyocha* (not diatom), Dip: *Diploneis*, Fra: *Fragilaria*, Gom: *Gomphonema*, Gra: *Grammatophora*, Mel: *Melosira*, Mes: *Mestogloria*, Nav: *Navicula*, Nit: *Nitzschia* and Ske: *Skeletonema*.

\*\*\* Rare: containing *Biddulphia*, *Corethron*, *Mestogloria*, *Pleurosigma* and *Suriella*

\*\*\*\* Others: diatoms extremely decomposed and unclassified because of being too small and/or lying on girdle.



Fig. 2. Observed diatoms. Arrow indicates a small diatom unclassified because of being extremely small. At a magnification of 400-fold.

collected in the winter season. Data from the summer season is necessary to complement this study.

This study was presented orally at the progress report of 2003 survey of the research project "An Interdisciplinary Study for Endogenous Development of the Isolated Small Island -A Representative Case Study for Yoron Island-". The abstract in Japanese will be published in Kagoshima University Research Center for the Pacific Islands Occasional Papers No. 41 (2004).

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## 与論島をモデルとした外海島嶼における溺死診断のための プランクトン検査の有用性の検討

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### 要 旨

水中死体の溺死診断のために行うプランクトン（主に珪藻）検査の外海島嶼における有用性を評価するために与論島における珪藻の分布調査を行った。島内10箇所（茶花港, 宇勝海岸, 黒花海岸, 皆田海岸, 船倉海岸, 大金久海岸, 赤崎漁港, 前浜海岸, ハキビナ海岸, 与論/供利港）より試料を採集した。また, 県本土5箇所（谷山港, 生見海岸, 山川新港, 額娃漁港, 枕崎港）からも試料採取を行い対照とした。海水は発煙硝酸により壊機後, その遠心沈渣のプレパラート標本を作製し, 鏡検により珪藻を観察した。海岸の海面, 海底, 沖合数10m沖合の海面での珪藻数は, 宇勝海岸や与論港などで県本土に比してやや少ないところもみられたが概ね県本土の最低値（16.5個/mL）よりも高い値を示した。一方, 沖合数100mの海水では珪藻数は激減した（0.5～2.9個/mL）。珪藻数は, 採取場所, 海面と海底, 海岸からの距離, 潮の干満などにより変化がみられた。この様な成績は, 与論島のような外海島嶼における水中死体に対して行なうプランクトン検査では, 推定される入水地点の海水の検査が不可欠であることを示唆した。