Dinoflagellate Cysts Records from Core Samples of Modern Marine Sediment at the Luoyuan Bay Mouth

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Abstract: Thirty species of dinoflagellate cysts in 15 genus are identified from KMZK5 Core samples of recent marine sediment at the Luoyuan Bay mouth, Fujian. All of these dinoflagellate cysts are first recorded in the Luyuan Bay, 12 species of them are not distributing in the near sea area such as Sansha Bay and Minjiang estuary, including 6 kinds of toxic species, such as *Alexandrium affine*, *A. minutum*, *A. tamarense*, *Gonyaulax spinifera*, *Gymnodinium catenatum* and *Scrippsiella trochoidea.* The abundance and vertical distribution characterestics of the main and the toxic dinoflgellate cysts are also studied in the paper.

Keywords: dinoflagellate cysts; vertical distribution; toxic dinoflagellate species

Introduction

The dinoflagellate resting cysts are the non-motion zygotes formed by vegetation cells sexual reproduction. They may float in waters and eventually sink on the sea floor to resist the unfavorable environmental condition for plankton cells, after recovery of the environmental condition they can germinate and re-establish motile population in the sea waters and reproduce in an agamogenesis way, so the dinoflagellate cysts may be treated as one seed source of red tide [1, 2].

Two different phragma, organic sporo-pollenin or calcareous material build up the wall of modern dinoflagellate cysts, and the organic sporo-pollenin wall is highly resistant to the chemically and biologically adverse ambient condition, which provides specific resistant features of the resting cysts and gives them a chance to be preserved perennially in marine sediments. The dinoflagellate cysts in marine sediment can reflect the taxa of vegetation cells in waters so that the dinoflagellate communities and the marine environmental changes can be reconstructed on the basis of the cyst assemblages [3].

The Luoyuan bay locates in the intersection of Luoyuan County and Lianjiang County, Fujian Province, which is surrounded by hills with a large internal sea area and a narrow mouth. With the large-scale increasing of cage aquaculture and the decreasing of laminaria culture, the frequency and the duration of red tide occurrence increase distinctly in the Luoyuan bay [4]. Although the investigation on algae assemblages including dinoflagellate communities in the water column had been carried out before [5], researches on dinoflagellate cysts in sediment have not been done in the Luoyuan bay. The depth of core sediment samples reserving the dinoflagellate cysts mainly limits to the surface of 0-20 cm,

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there is lack of the corresponding research on dinoflagellate cysts in the core sediment exceeding 20cm in depth [2, 5]. The component and the abundance of dinoflagellate cysts in the KMZK5 core, which locates in the Luoyuan bay entrance sea area, is about 100 cm-long, are studied in this paper. The results can not only provide reference for studying the change of dinoflagellate communities in waters, but also reconstruct the process of eutrophication in the Luoyuan bay.

1 Materials and methods

1.1 Sampling

The KMZK5 core sediment, which was collected in the Luoyuan Bay, 100 cm in length (26°26′51″N, 119°49′54″E; 17.3 m water depth; Fig. 1), was taken continuously every 4 cm, so a total of 25 samples were analysed.

The core sediments consist of clayish fine grain and shell fragments from 0 cm to 4 cm, gray clay from 4 cm to 100 cm. There is no evident bioturbation in the core section.

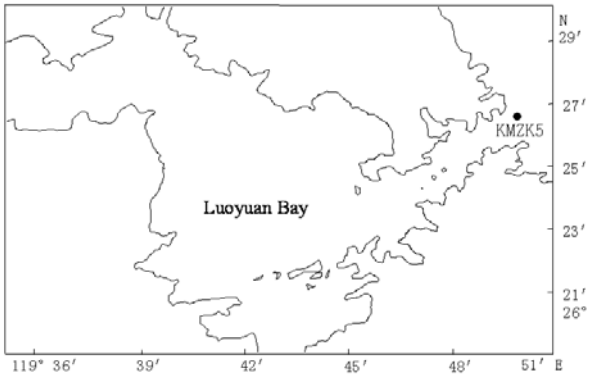

Fig.1 Location of the sampling core

1.2 Methods of analysis

The samples for analysis are placed into a drying oven under 70 °C for about 24 hours and weighed 5 gram in beaker. Add 20 mL distilled water and mix it with the sediment. Then disperse the samples by ultrasonic disintegrator at a frequency of 150 Hz for 2 min. Wash the dispersed samples with distilled water under a set of two sieves of mesh-size with 125 μm being the upper and 15 μm being the lower to remove the tiny particles of sediment. Put the residue in a 10 mL tube for centrifugation (approx. 4000 rpm, 2 min). Suck out the supernatant by a pipette till to 2 mL and shake it up, then add 1 mL solution of the tungstenic acid natrium (1.6 special gravity), suck out all of the upper layer samples carefully by a pipette and move them into a 5 mL tube to wash on the 15 μm sieves for two times, collect all residues on the 15 μm sieves into the 5 mL vial and make up the solution 2 mL with distilled water. This is the refined sample ready for the next study. The cysts identification was carried out under an bio-microscope Nikon E400 with a

photomicrographic system. The total number of identification cysts is counted until more than 100 grains.

2 Results and discussion

2.1 The components of dinoflagellate cyst species in Core KMZK5

More than 30 dinoflagellate cyst species belonging to 15 genera (Plate I) were identified from the sediment of KMZK5 core on the basis of the criterion created by Kazumi and Yasuwo[6]. The species of dinoflagellate cyst are: *Alexandrium affine*, *A. minutum*, *A. tamarense**, *Cochlodinium polykrikoides*, *Diplopsalis lenticula*, *Gonyaulax scrippsea*, *G. spinifera**, *Gymnodinium catenatum**, *Gyrodinium impudicum*, *Lingulodinium polyedrum*, *Pheopolykrikos hartmannii*, *Polykrikos kofoiddii*, *Prorocentrium reticulatum*, *Protoperidinium americanum*, *P. avellana*, *P. claudicans*, *P. compressum*, *P. conicum*, *P. conicoides*, *P. denticulatum*, *P. latissimum*, *P. leonis*, *P. minutum*, *P. oblongum*, *P. pentagonum*, *P. subinerme*, *P. thorianum*, *Pyrophacus steinnii*, *Scrippsiella trochoidea**, *Zygabikodinium lenticulatum* (*means toxic species).

A total of 17 species belonging to 6 genera of dinoflagellate vegetation cells have been identified in the water column[5], they were *Ceratium furca*, *Cer. fusus*, *Cer. inflatum*, *Cer. deflexum*, *Cer. macroceros*, *Cer. massiliense*, *Cer. trichoceros*, *Cer. breve*, *Cer. gibberum*, *Cer. tripos*, *Cer. pulchellum*, *Dinophysis caudate*, *Noctiluca scintillans*, *Prorocentrum micans*, *Protoperidinium depressum*, *P. oceanicum*, *Pyrophacus* var. *horologium*. Among them only *Pyrophacus* var. *horologium* can form the resting cyst, but the cyst of this species was not found in the sediment. 29 species of dinoflagellate were recorded in the Xinghua Bay, Fuqing Bay, Sansha Bay and Shacheng Bay according to the literature [5], among which only *Protoperidinium conicum*, *P. pentagonum* and *Pyrophacus* var. *horologium* can form the resting cyst.

The dinoflagellate cysts found in the core sediments from the Luoyuan bay have the similar characteristics to that were found in surface sediments from the adjacent Mingjiang estuary and Sansha bay[7], for example, among the 20 species in 9 genera of dinoflagellate cysts recorded in the surface sediments from the Sansha bay, there are 12 species found in the Luoyuan bay: *Alexandrium affine*, *A. minutum*, *A. tamarense*, *Gonyaulax scrippsea*, *G. spinifera*, *Lingulodinium polyedrum*, *Polykrikos kofoiddii*, *Prorocentrium reticulatum*, *Protoperidinium americanum*, *P. conicum*, *P. minutum* and *Scrippsiella trochoidea*. Among the 17 species in 7 genera of dinoflagellate cyst recorded in the surface sediments from the Mingjiang estuary [1], there are more than 14 species found in the Luoyuan bay, such as *Alexandrium affine*, *A. tamarense*, *Gonyaulax scrippsea*, *G. spinifera*, *Polykrikos kofoiddii*, *Prorocentrium reticulatum*, *Protoperidinium avellana*, *P. conicum*, *P. conicoides*, *P. denticulatum*, *P. leonis*, *P. oblongum*, *P. pentagonum* and *Scrippsiella trochoidea*. But the other 12 species of dinoflagellate cysts recorded in the Luoyuan bay are not found in the Mingjiang estuary and Sansha bay, including *Cochlodinium polykrikoides*, *Diplopsalis lenticula*, *Gymnodinium catenatum*, *Gyrodinium impudicum*, *Pheopolykrikos hartmannii*, *Protoperidinium claudicans*, *P. compressum*, *P. latissimum*, *P. subinerme*, *P. thorianum*, *Pyrophacus steinnii*, *Zygabikodinium lenticulatum*, especially *Gymnodinium catenatum*, the toxic dinoflagellate cyst has never been recorded in the Luoyuan bay before.

The dinoflagellate cysts preserved in the sediments suggest that the same vegetation cells should

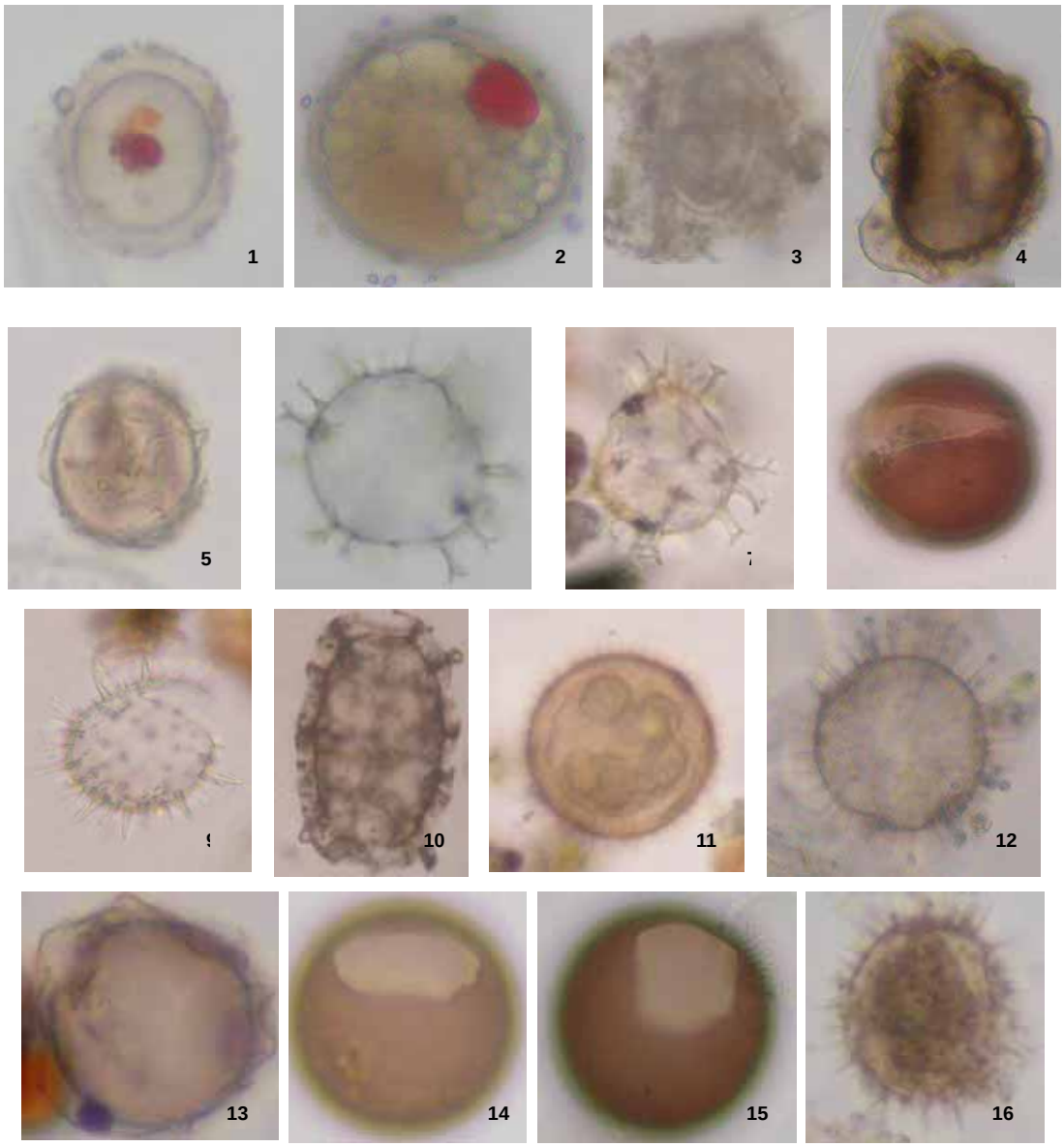

Plate Ⅰ Dinoflagellate cysts from core samples of submarine sediment at the Luoyuan Bay mouth

1. *Alexandrium affine* (×500), 2. *Alexandrium minutum* (×700)，3. *Alexandrium tamarense* (×400),
4.*Cochlodinium polykrikoides* (×600), 5.*Diplopsalis lenticula* (×600), 6.*Gonyaulax scrippsae* (×600),
7.*Gonyaulax spinifera* (×500), 8.*Gymnodinium catenatum* (×500), 9. *Lingulodinium polyedrum* (×500),
10.*Polykrikos kofoiddii* (×500), 11.*Pheopolykrikos hartmannii* (×550), 12. *Prorocerat*ium *reticulatum* (×700),
13. *Protoperidinium americanum* (×800), 14, 15. *Protoperidinium avellana* (×650),
16.*Protoperidinium conicum* (×570),

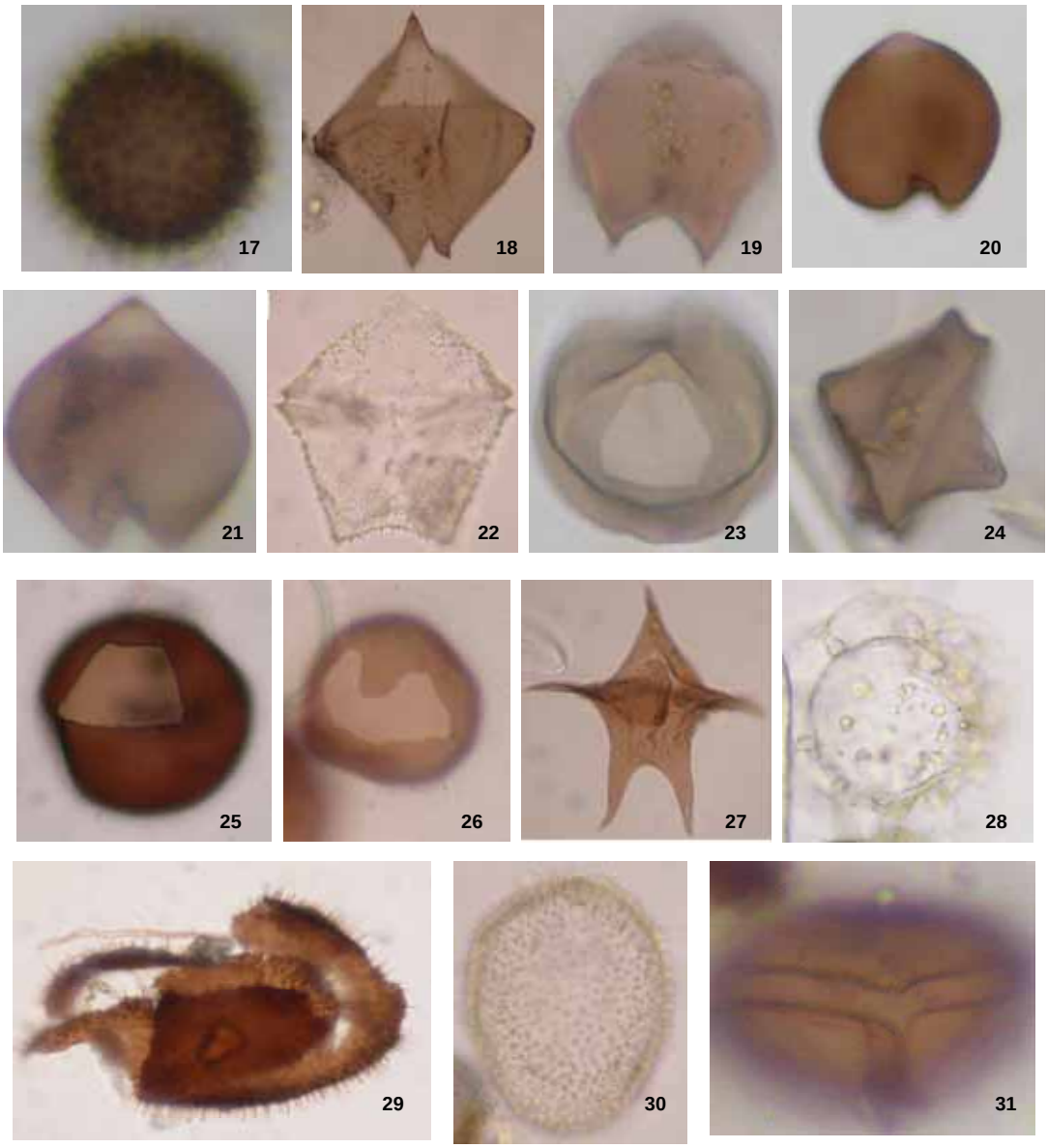

Plate Ⅰ (continue) Dinoflagellate cysts from core samples of submarine sediment at the Luoyuan Bay mouth

17. *Protoperidinium minutum* (×1000), 18. *Protoperidinium latissimum* (×350),
19. *Protoperidinium leonis* (×380), 20, 21. *Protoperidinium oblongum* (×450),
22. *Protoperidinium pentagonum* (×400), 23, 24. *Protoperidinium subinerme* (×600),
25. *Protoperidinium thorianum* (×600), 26. *Protoperidinium denticulatum* (×400),
27. *Protoperidinium compressum* (×500), 28. *Pyrophacus steinnii* (×400),
29, 30. *Scippsiella trochoidea* (×400), 31. *Zygabikodinium lenticulatum* (×800)

exist in water. But there has obvious distinction between dinoflagellate cysts found in the sediment samples and the dinoflagellate vegetation reported by the literature [5], this fact also exists in the Mingjiang estuary [1], Sansha bay [7] and Xiamen bay [8]. Besides the fact that most dinoflagellate cells recorded in the past investigation could not produce resting cysts, it may be caused by the reasons as follows. Firstly, the criterion mesh-size tow net is 77 μm according to the Ocean Investigation Criterion- Halobios Investigation (GB/T 12763. 6-1991), but the sizes of many kinds of dinoflagellate cells range from 40 μm to 60 μm, those cells with sizes smaller than 77μm can leak from the standard mesh-size tow net. Secondly, some dinoflagellates can decompose at once as dying, it is very difficult to be preserved by fixed liquor, such as *Polykrikos kofoiddii* can be observed alive in the spot test of sea water, but it can never be observed in the fixed samples. Thirdly, some newly discovered dinoflagellates species, which can produce resting cysts, may probably be introduced into the study sea area by some approaches, such as by ballast water discharging, by introduction of new species for aquatic-breeding. A detailed study need be carried out to solve this problem.

All the identified dinoflagellate cysts are preserved in the core sediment, some kinds of dinoflagellate cysts may be alive according to the fresh protoplasm in surface sediments, the other dinoflagellate cysts in core sediments cannot reproduce or have just reproduced to leave empty cysts. More precise dinoflagellate cysts identification by cyst breeding can be carried out after collecting modern surface sediment samples in the Luoyuan bay in the future.

2.2 Vertical distribution of dinoflagellate cysts

The vertical distribution of important dinoflagellate cyst species and their abundance, the total cysts abundance is plotted in Fig.2. *Gonyaulax scrippsea*, *Lingulodinium polyedrum*, *Polykrikos kofoiddii*, *Prorocerat ium reticulatum*, *Protoperidinium leonis* and *P. oblongum* have relatively high abundance, with the values about 20 cysts•g^{-1} (dry weight), but the other dinoflagellate cysts abundances are less than 10 cysts•g^{-1} and distributing discontinuously in the whole core section.

As shown in Fig.2, *Gonyaulax scrippsea*, *Lingulodinium polyedrum*, *Protoperidinium leonis* and *P. oblongum* generally have maximum abundance values in the upper part of the core and increase evidently at 4 - 6 cm, *L. polyedrum* and *P. leonis* reaches the maximum value of about 15 cysts•g^{-1} at 5 cm, which relatively abundance reaches 14 % of the total cysts. *Lingulodinium polyedrum* is a typical coastal species, and its high abundance (5 % - 40 % of the total cysts) is an index of eutrophic waters [9], so a eutrophication process can be deduced on the basis of the cyst abundance of *Lingulodinium polyedrum* at 5cm. High abundance of dinoflagellate cysts in surface sediments is likely to associate with the red tide occurring in the Luoyuan bay in the past two years[4]. Main kinds of dinoflagellate cysts' abundances are no more than 10 cysts•g^{-1}, such as *Lingulodinium polyedrum* is less than 5 cysts•g^{-1}, *Gonyaulax scrippsea*, *Polykrikos kofoiddii* and *P. oblongum* can not be found in some layers of the core between 20cm and 55cm. In the lower part of core (under 55cm), main dinoflagellate cysts present continuously, *Gonyaulax scrippsea* has a high abundance of 9 - 15 cysts•g^{-1} and reaches the maximum value of 15 cysts•g^{-1} at 90cm, *P. leonis* has two high value points (up to 13 cysts•g^{-1} and 14 cysts•g^{-1}) at 56cm and 86cm respectively. The cyst abundance of *Prorocerat ium reticulatum* limits to 10 cysts•g^{-1}, and has an increasing trend with depth

under the section of 50 cm in the core.

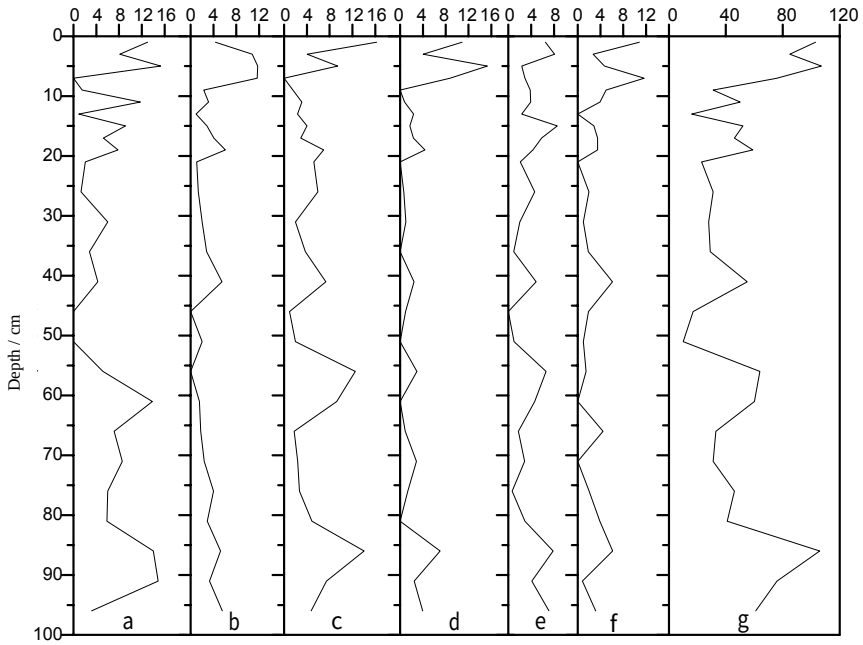

Fig. 2 Vertical distribution of main dinoflagellate and total cysts concentration

a. *Gonyaulax scrippsea*, b. *Lingulodinium polyedrum*, c. *Polykrikos kofoiddii*, d. *Prorocerati*um *reticulatum*, e. *Protoperidinium leonis* f. *P. oblongum*, g. total abundance of all dinofalgellate cysts.

The total dinoflagellate cysts abundance is from 10 cysts•g^{-1} to 107 cysts•g^{-1}, in the upper part of the core (0 - 30 cm), the total cysts abundance shows an increasing trend toward the top of the core and shows relatively high values at 5 cm, the total cysts abundance has 3 peak values (56 cysts•g^{-1} at 41 cm, 64 cysts•g^{-1} at 56 cm, 106 cysts•g^{-1} at 86 cm) between 30 cm and 100 cm, there are lower values of cysts abundance between the peak values, and the minimum value of cysts decreases to 10 cysts•g^{-1} at 86 cm.

The depositional behavior of dinoflagellate cysts is like that of clay, and their abundance increases along with the percentage of clay increasing in sediments (Dale 1983) [10]. Anderson suggests that dinoflagellate cysts mainly deposit in the low sea floor in the process of clay grain accumulation [11]. The core (0 - 4 cm) consists of clayish fine sand at the top sample (0 - 4 cm) and clay in the part of 4 - 100 cm, which may explain for the maximum dinoflagellate cysts abundance at 4 - 6 cm. Besides, the core is located at the mouth of Luoyuan bay, the high abundance of the dinoflagallete distributing in the inner bay can be transported to the outside of the bay by ebb-tide, and deposit along with the hydrodynamic condition decreasing in the process of transportation. Recently, the rapid development of aquiculture at the Luoyuan bay has led to eutrophication in the sea waters, red tides in the southwest and east of the bay happen almost every year, moreover the frequency of the red tide shows an increasing trend [4]. Thus, the

high abundance of dinoflagellate cysts in the surface sediment from the Luoyuan bay mouth is likely to indicate the red tide events occurring in the bay; high abundances of dinoflagellate cysts in the different lower sections of the core may also indicate the red tide events occurring inside of the bay in the past years. For lacking surface and core sediment samples from the inner part of the bay, the red tide records and the data of the core sediment age-dating, the reason for the high abundance of dinoflagellate cysts in the lower part of the core awaits further analysis in the future.

2.3 Distribution of toxic dinoflagellate cysts

Six toxic species have been identified from the core sediment samples shown in Fig. 3, they are *Alexandrium affine*, *A. minutum*, *A. tamarense*, *G. spinifera*, *Gymnodinium catenatum* and *Scrippsiella trochoidea*. Among them, *Alexandrium affine* only distributes in the top of core with an abundance of 1 cysts•g^{-1} and the other toxic dinoflagellate cysts distribute discontinuously. The characteristics of *A. minutum* and *A. tamarense* distribution are obviously discontinuous in the whole core section, their abundances have the similar trends at 50 - 100 cm, no more than 10 cysts•g^{-1}. *G. spinifera* distributes throughout the whole core with a maximum abundance of about 9 cysts•g^{-1} at the top of core and different peak values of abundance at 6 cm, 40 cm, 60 cm, 86 cm and the bottom of the core. *Gymnodinium catenatum* only distributes in the top of samples or in the lower part of the core (74 - 100 cm) and the abundance ranges from 0 cysts•g^{-1} to a maximum value of 5 cysts•g^{-1} at 90cm. *Scrippsiella trochoidea* first appears from the 60 cm section and distributes in the upper part of the core, its abundance is less than 5 cysts•g^{-1}.

The identified species of toxic dinoflagellate cysts in the Luoyuan bay this time are more than in other sea areas in Fujian province[1, 7, 12, 13], additionally, the frequency of the red tide had an increasing trend in last 4 years, so the probability of toxic red tide breaking out is higher in the Luoyuan bay. In fact, from April 2002 to the middle ten days of May, there experienced the worst recorded marine toxicity event which caused 3 people to death, more than 50 people were poisoned by eating *Nassarius* in Luoyuan, Ningde, Xiamen and Putian [14], and the paralytic shellfish poisoning (PSP) toxin was detected from the samples collected in these poisoned person. Among the 6 toxic species in sediment samples from the Luoyuan bay, *Alexandrium minutum*, *A. tamarense* and *Gymnodinium catenatum* can produce PSP toxins, although these three species cysts' abundance is very low, the PSP toxin produced by the species of *Alexandrium* can be concentrated to high level by mussels [8], which may cause human death very easily when those mussels were eaten by accident. The residents nearby the area should pay more attention and avoid toxicity event breaking out again in the future.

3 Conclusion

a) Thirty species (belonging to 15 genera) dinoflagellate cysts were identified in the sediment samples of 100cm long KMZK5 core at the Luoyuan bay mouth, including 6 toxic species *Alexandrium affine*, *A. minutum*, *A. tamarense*, *Gonyaula spinifera*, *Gymnodinium catenatum* and *Scrippsiella trochoidea*.

b) The maximum abundance values of main dinoflagellate cysts are less than 20 cysts•g^{-1} and the

total cysts abundance ranges from 10 to 107 cysts•g^{-1}, in the upper part of the core (0 - 30 cm), the total dinoflagellate cysts abundance shows an increasing trend toward the top of the core and reaches relatively high values at 5 cm, whereas under 30 cm its abundance shows 3 peak values.

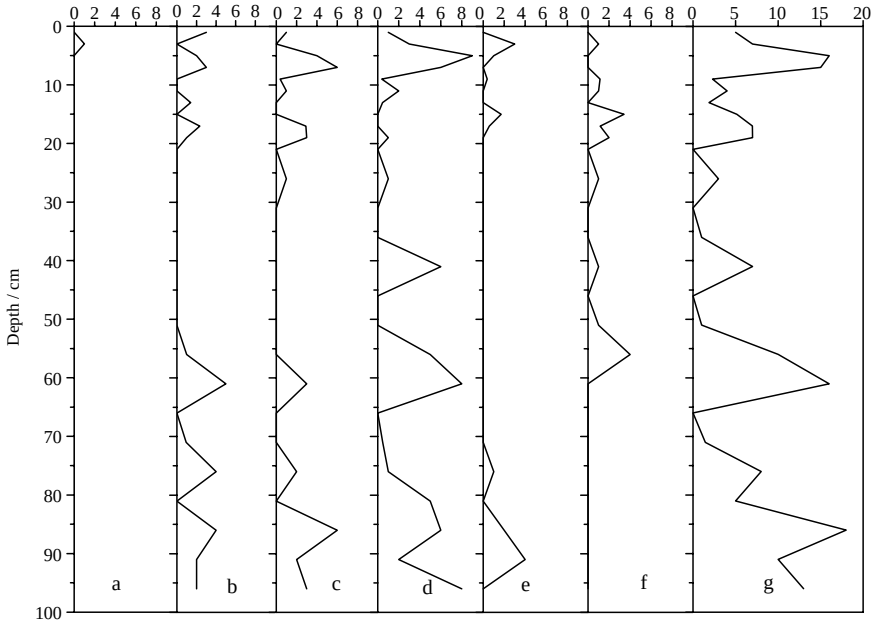

Fig. 3 Distribution of toxic dinoflagellate cysts

a: *Alexandrium affine*, b: *A. minutum*, c: *A. tamarense*, d: *G. spinifera*, e: *Gymnodinium catenatum*, f: *Scrippsiella trochoidea*, g: total abundance of toxic dinoflagellate cysts

c) The 6 toxic species are present discontinuously in low abundance values which limit to 10 cysts•g^{-1}, their percentage of the total cysts abundance changes in a fluctuating way. These toxic dinoflagellate cysts indicate that the Louyuan Bay sea area is well provided with the conditions for the toxic red tide to break out, the residents around the Luoyuan bay should pay more attention and avoid toxicity events happening in the future.

Acknowledgements

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罗源湾口柱状沉积物中的甲藻孢囊

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摘 要：通过对福建罗源湾口海域 KMZK5 柱状沉积物中甲藻孢囊的分析，共鉴定出 15 属 30 种甲藻孢囊。对比发现这 30 种甲藻孢囊是该湾以前未被记录的种类。其中 12 种是附近海域也未曾发现的种类，6 种为有毒种类：缘亚历山大藻、小型亚历山大藻、塔玛亚历山大藻、具刺膝沟藻、链状裸甲藻、锥状斯氏藻.同时对甲藻孢囊的主要属种和有毒种类的丰度、分布在垂直方向上的变化特征进行了初步研究.

关键词：甲藻孢囊；丰度；垂直分布；有毒种；罗源湾