EFFECTS OF SALINITY, TEMPERATURE AND PH ON HARMFUL ALGAL BLOOMS SPECIES IN KOTA KINABALU COASTAL WATERS

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BORNEO MARINE RESEARCH INSTITUTE

UNIVERSITI MALAYSIA SABAH

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ABSTRACT

EFFECTS OF SALINITY, TEMPERATURE AND PH ON HARMFUL ALGAL BLOOMS SPECIES IN KOTA KINABALU COASTAL WATERS.

The occurrence of harmful algal blooms (HAB) since last three decades in Sabah waters had caused poisoning incidents, which lead to the human and organism illness. Some HABs species also caused damages especially lost in the local aquaculture industries. Realizing the lack of information available regarding to the factors affecting the bloom, thus this study was carried out. The objective of this study was to determine the effects of physico-chemical properties of seawater (i.e. salinity, temperature and pH) on the HABs species viz. *Pyrodinium bahamense* var. *compressum, Cochlodinium polykrikoides* and *Gymnodinium catenatum* in the Kota Kinabalu waters from January to December 2007. At six sampling stations, physical properties of seawater were measured *in-situ* at sea surface using multi probes (Hydrolab Surveyor 4 and HANNA Instrument) and phytoplankton samples were collected at 0.5 meter depth using water sampler. The highest peak of *C. polykrikoides* was recorded on 3rd December 2007 (1.54×10^7 cells L^{-1}). During the bloom, sea surface salinity, temperature and pH recorded were 26.4 psu, 29 ºC and 8.78, respectively. Maximum concentration of *G. catenatum* was also reported in December with cell densities of 1.24×10^6 cells L^{-1}. *Pyrodinium bahamense* var. *compressum* reached a maximum concentration on August, with the cell density of 2×10^4 cells L^{-1}. During the bloom, salinity, temperature and pH were 30.9 psu, 31.3 ºC and 8.50, respectively. Salinity and pH of seawater were positively correlated ($p<0.05$) to the cell concentrations of *C. polykrikoides*, whereas temperature showed negative correlation ($p<0.05$). No correlation was observed between the physico-chemical parameters with cell density of *G. catenatum*. However, negative correlation was observed between salinity and cell density of *P. bahamense* var. *compressum* ($p<0.05$), whereas temperature showed positive correlation. This study is useful in order to understand the mechanism of HABs occurrence and this may help government or any relevant agencies in mitigate and manage HAB problems in Sabah.

ABSTRAK

Kehadiran ledakan populasi alga bahaya sejak tiga dekad yang lalu di perairan Sabah telah mengakibatkan kes keracunan, yang menjurus kepada kematian tidak kira manusia atau organisma. Sesetengah ledakan populasi alga bahaya ini turut menyebabkan kerugian terutamanya kepada industri akuakultur tempatan. Menyedari kekurangan maklumat berkaitan dengan faktor mempengaruhi ledakannya, maka kajian ini dijalankan bagi mengenalpasti kesan perubahan sifat fiziko-kimia air laut (iaitu saliniti, suhu dan pH) terhadap kelimpahan beberapa spesis alga bahaya iaitu Cochlodinium polykrikoides, Gymnodinium catenatum dan Pyrodinium bahamense var. compressum di perairan Kota Kinabalu, bermula dari bulan Januari sehingga Disember 2007. Bacaan fiziko-kimia permukaan air laut direkodkan dengan menggunakan 'multi probe' (Hydrolab Surveyor 4 dan HANNA Instrument) dan sampel fitoplankton diambil pada kedalaman 0.5 meter menggunakan 'water sampler' di enam buah stesen. Densiti sel tertinggi bagi C. polykrikoides direkodkan pada Disember 2007 (1.54 x 10^7 sel L^{-1}). Pada masa tersebut, saliniti, suhu dan pH yang dicatatkan adalah 26.4 psu, 29 ºC dan 8.78. Maksimum densiti sel G. catenatum turut direkodkan pada bulan Disember, dengan sel densiti iaitu 1.24 x 10^6 sel L^{-1}. Maksimum densiti sel P. bahamense var. compressum direkodkan pada bulan Ogos sebanyak 2 x 10^4 sel L^{-1}. Pada masa tersebut, saliniti, suhu dan pH yang dicatatkan adalah 30.9 psu, 31.3 ºC dan 8.50. Didapati wujud korelasi positif antara saliniti dan pH terhadap densiti sel C. polykrikoides ($p<0.05$), manakala korelasi negatif diperoleh antara suhu dan densiti sel C. polykrikoides. Tiada korelasi diperoleh antara densiti sel G. catenatum dan parameter fiziko-kimia. Namun, wujud korelasi negatif antara densiti sel P. bahamense var. compressum dan saliniti, sebaliknya wujud korelasi positif terhadap suhu. Diharapkan kajian ini dapat membantu dalam memahami fenomena ledakan populasi alga bahaya, seterusnya dapat membantu pihak kerajaan atau agensi-agensi yang berkaitan di dalam menangani dan pengurusan isu peningkatan populasi alga bahaya di Sabah.

TABLE OF CONTENTS

	Page
TITLE	i
DECLARATION	ii
CERTIFICATION	iii
ACKNOWLEDGEMENTS	iv
ABSTRACT	v
ABSTRAK	vi
LIST OF CONTENTS	vii
LIST OF TABLES	x
LIST OF FIGURES	xi
LIST OF ABBREVIATIONS	xii
LIST OF SYMBOLS	xiii
LIST OF APPENDICES	xiv
CHAPTER 1 : INTRODUCTION	
1.1 An Introduction	1
1.2 Objectives	4
1.3 Significance of the Study	5
CHAPTER 2 : LITERATURE REVIEW	
2.1 Harmful Algal Blooms (HABs)	
2.1.1 History of HABs Occurrences	6
2.1.2 Definition of HABs	6
2.1.3 Hazards of HABs	7
2.2 HABs Phenomenon in Sabah, Malaysia	10
2.3 Factors Triggering HABs	
2.3.1 Nutrients	13
2.3.2 Physico-chemical Properties of Seawater	15

CHAPTER 3 : METHODOLOGY

3.1 Study Area 18
3.2 Sampling Schedule 20
3.3 Sampling Procedures 21
3.4 Species Identification and Quantification 22
3.5 Data Analysis 22

CHAPTER 4 : RESULTS

4.1 Temporal Distribution of HABs Species
4.1.1 *Cochlodinium polykrikoides* 24
4.1.2 *Gymnodinium catenatum* 24
4.1.3 *Pyrodinium bahamense* var. *compressum* 28

4.2 Spatial Distribution of HABs in Kota Kinabalu Waters 28
4.3 Physico-chemical Parameters during the HABs 33
4.4 Effects of salinity, temperature and pH on HABs Species
4.4.1 *Cochlodinium polykrikoides* 40
4.4.2 *Gymnodinium catenatum* 41
4.4.3 *Pyrodinium bahamense* var. *compressum* 41

CHAPTER 5 : DISCUSSIONS

5.1 Temporal Distribution
5.1.1 *Cochlodinium polykrikoides* 43
5.1.2 *Gymnodinium catenatum* 46
5.1.3 *Pyrodinium bahamense* var. *compressum* 47

5.2 Spatial Distribution
5.2.1 *Cochlodinium polykrikoides* and *Gymnodinium catenatum* 48
5.2.2 *Pyrodinium bahamense* var. *compressum* 49

5.3 Effect of Salinity on HABs Species
5.3.1 *Cochlodinium polykrikoides* 49
5.3.2 *Gymnodinium catenatum* 50
5.3.3 *Pyrodinium bahamense* var. *compressum* 51

5.4 Effect of temperature on HABs species
5.4.1 *Cochlodinium polykrikoides* 52
5.4.2 *Gymnodinium catenatum* 53
5.4.3 *Pyrodinium bahamense* var. *compressum* 54

5.5 Effect of pH on HABs Species 55
5.6 Limitations of the Study
5.6.1 Misidentification of *Cochlodinium polykrikoides* and 56

Gymnodinium catenatum
5.6.2 Weather and Sea Condition 57
5.6.3 Availability of Equipments and Boat 57

CHAPTER 6 : CONCLUSIONS AND RECOMMENDATIONS

6.1 Conclusions 58
6.2 Recommendations 59

REFERENCES 60

APPENDICES 69

LIST OF TABLES

		Page
Table 2.1	Illnesses caused by HABs species	9
Table 2.2	Confirmed PSP cases in Sabah	11
Table 2.3	Field measurements of salinity and temperature during *Cochlodinium* sp. blooms.	17
Table 3.1	GPS coordinate of sampling stations in the study area	20
Table 3.2	Sampling schedule throughout 2007	21
Table 4.1	Salinity, temperature and pH during the highest density of *C. polykrikoides, G. catenatum* and *P. bahamense* var. *compressum*	33
Table 4.2	Non - parametric correlation between salinity, temperature, pH and cell density of *C.polykrikoides*	37
Table 4.3	Non - parametric correlation between salinity, temperature, pH and cell density of *G. catenatum*	38
Table 4.4	Non - parametric correlation between salinity, temperature, pH and cell density of *P. bahamense* var. *compressum*	49
Table 5.1	Summary of *C. polykrikoides* occurrences worldwide	45
Table 5.2	Salinity and temperature observed during *Gymnodinium catenatum* blooms	51
Table 5.3	Salinity and temperature observed during *Pyrodinium bahamense* var. *compressum* blooms in Asia region	52

LIST OF FIGURES

		Page
Figure 2.1	Factors regulating HABs	13
Figure 3.1	Location of sampling stations in Kota Kinabalu coastal waters.	19
Figure 4.1	*C. polykrikoides, G. catenatum* and *P. bahamense* var. *compressum* found in Kota Kinabalu coastal waters	24
Figure 4.2	Weekly *C. polykrikoides* abundance (average cell density) at each station in 2007	26
Figure 4.3	Weekly *G. catenatum* abundance (average cell density) at each station in 2007	27
Figure 4.4	Weekly *P. bahamense var. compressum* abundance (average cell density) at each station in 2007	29
Figure 4.5	Total HABs occurrences (based on the monthly occurrences)	30
Figure 4.6	Numbers of *C. polykrikoides, G. catenatum* and *P. bahamense* var. *compressum* cells densities at each station	32
Figure 4.7	1-year distribution of *C. polykrikoides* density, in relation with salinity and temperature	34
Figure 4.8	1-year distribution of *G. catenatum* density, in relation with salinity and temperature	35
Figure 4.9	1-year distribution of *P. bahamense var. compressum* density, in relation with salinity and temperature	36
Figure 4.10	1-year distribution of *C. polykrikoides* density, in relation with pH	37
Figure 4.11	1-year distribution of *G. catenatum* density, in relation with pH	38
Figure 4.12	1-year distribution of *P. bahamense var. compressum* density, in relation with pH	39

LIST OF ABBREVIATIONS

ASP	Amnesic shellfish poisoning
BOD	Biological oxygen demand
CO_2	Carbon dioxide
DSP	Diarrhetic shellfish poisoning
Fe	Iron
HABs	Harmful Algal Blooms
IM	Inter-monsoon
N	Nitrogen
NEM	Northeast monsoon
NSP	Neurotoxic shellfish poisoning
PSP	Paralytic shellfish poisoning
pH	hydrogen ion concentration
ppt	Part per thousand
SWM	Southwest monsoon
TEU	Twenty-foot equivalent units
UMS	Universiti Malaysia Sabah

LIST OF SYMBOLS

%	Percent
^{o} C	Degree Celsius
&	And
$	Dollar
N	Nitrogen
P	Phosphorus
NO_3^-	Nitrate
PO_4^{3-}	Phosphate
Fe	Ferum / Iron
Se (IV)	Selenium
NH_4^+	Ammonium
$\mu g\ L^{-1}$	Microgram per litre
$mg\ L^{-1}$	Miligram per litre
ml	Mililitre
ng/µL	Nano gram per microlitre

LIST OF APPENDICES

		Page
Appendix A	Sampling data at Station 1	72
Appendix B	Sampling data at Station 2	73
Appendix C	Sampling data at Station 3	74
Appendix D	Sampling data at Station 4	75
Appendix E	Sampling data at Station 5	76
Appendix F	Sampling data at Station 6	77
Appendix G	Significant different between months	78
Appendix H	Descriptive analysis	87
Appendix I	Significant different of sea surface salinity, temperature and pH, cell density of *C. polykrikoides, G. catenatum* and *P. bahamense* var. *compressum.*	88

CHAPTER 1

INTRODUCTION

1.1 An introduction

It has been three decades since Sabah of Malaysia has been facing the harmful algal blooms (HABs) problems along its coastal waters. HABs can be defined as large propagation of phytoplankton, which sometimes associated with discolouration of surface seawater. Some of the HAB species can be harmful, either by producing toxins (Sellner *et al*., 2003), cause oxygen depletion (Puslé, 1995) or can damage fish's gill tissue (Puslé, 1995). Since 1970s, many poisoning cases and even mortalities were reported all over Sabah. This has become a public concern as the HABs species are increasing from time to time in Kota Kinabalu coastal waters. To date, three dinoflagellates species have been reported to cause blooms along Kota Kinabalu coastal waters, which are *Pyrodinium bahamense* var. *compressum* (Roy, 1977), *Cochlodinium polykrikoides* (Anton *et al*., 2008) and *Gymnodinium catenatum* (Mohammad-Noor *et al*., 2008).

In Sabah, the first occurrence of HABs species was reported in Kota Kinabalu coastal waters in 1976 (Roy, 1977; Ting & Joseph, 1989). Species responsible for this bloom was identified as *P. bahamense* var. *compressum,* a toxin producer species. This species is reported to produce saxitoxin (Gedaria *et al*., 2007), which being accumulated by filter feeders (e.g. clams and oyster) and cause paralytic shellfish poisoning (PSP) in humans upon consumption of these contaminated shellfishes. During this event, two fatalities and eight illnesses were reported after consuming contaminated clams. Since then, blooms of this toxic dinoflagellate have became an annual event along Kota Kinabalu coastal waters (Ting & Joseph, 1989).

Gymnodinium catenatum, a PSP species was first found in low number in Sepanggar Bay in 2003 (personnal comm.). So far, no PSP incidence has been reported before. Later, in January 2005, large dark red patches were observed in front of Universiti Malaysia Sabah's (UMS) jetty (Anton *et al*., 2008). Species responsible was later identified as *C. polykrikoides*, a fish killer non-toxic species. Subsequent high densities of this species were observed in March 2005 and May-June 2005, but no more bloom was observed from September 2005 to April 2006, which the reason could be due to the rainy season of the northeast monsoon (Sabah Meteorological Department, 2005). *Cochlodinium polykrikoides* abundance reached a maximum concentration of 6 x 10^6 cells L^{-1} in early June 2006 and this bloom was coincided with the mortality of cultured finfish at the aquaculture cages near this area (Anton *et al*., 2008).

Reoccurrences of HABs in Kota Kinabalu coastal waters had called for detail study to understand the environmental factors, which may contribute to the occurrence of HABs. Although there were many reports on HABs in Sabah waters, the understanding of the environmental effects on these HABs species is still limited. The formations of HABs are reported to correlate with the interaction between physical and ecological factors (Wong *et al*., 2009). The analysis of the continuous field data during HABs suggests that blooms are governed by environmental factors (e.g. temperature, light extinction and nutrients), but equally important are the physical processes such as tidal flushing, wind and tidal mixing, and upwelling (Wong *et al*., 2009). However, this study only focuses some of the factors that might contribute to the occurrence of HABs, which are salinity, temperature and pH. This is to understand the basic condition of HABs occurrences and subsequently to predict the HABs occurrences.

The study area, which is located at the northern part of Kota Kinabalu waters, is influenced by freshwater load from few rivers nearby, mainly from Inanam – Likas River and Menggatal River. These rivers were reported to discharge excessive unt of amonutrient from industrial park and domestic wastewaters nearby (Mokhtar *et al*, 1994; ECD and UMS, 2002; Toha, 2008). In addition to supply excessive nutrient loads, these freshwater loading also might influence the salinity, temperature and pH of seawater. Thus, the conditions create an optimum requirement for these HABs species to bloom. For instance, occurrences of high cell density of *C. polykrikoides* along the seashores of Yeosu and Tongyeong, Korea seem to result from rainfall-initiated inflows of high levels of nitrate after a suitable physical and chemical open water environment has been created for *C.*

polykrikoides to spread initially (Lee, 2006). This suggests a wide tolerance for nutrient condition in addition to temperature and salinity. It is important to study on the effect of physico-chemical parameters of seawater for HABs species to bloom as these basic factors may influence the toxin production (e.g. Baker *et al*., 2007). Acute toxic activity of harmful algae is often highest under conditions that are sub-optimal for growth (Plumley, 1997). Previous studies have shown that reduced growth due to non-optimal temperature and salinity increased the toxicity of *Prymnesium parvum* to juvenile fish (Baker *et al*., 2007). Maximum toxicity of *P. parvum* to fish was observed at extreme (low and high) values of salinity and also for lower temperatures, while maximum growth and abundance were found at warmer temperatures and moderate salinities. Thus, it is needed to understand the basic conditions for the HABs species to bloom and/or to produce toxin. The blooms dynamics of *C. polykrikoides*, *G. catenatum* and *P. bahamense* var. *compressum* in Kota Kinabalu waters presented in this study can serve as a comparison to those previous blooms elsewhere, where different dynamics may govern them.

1.2 Objectives

There are three main objectives of this study:

1. To determine the temporal and spatial distribution of *C. polykrikoides, G. catenatum* and *P. bahamense var. compressum* in northern part of Kota Kinabalu coastal waters, Sabah.

2. To determine the sea surface salinity, temperature and pH during the occurrence of HABs.

3. To determine the effects of the salinity, temperature and pH on cell density of *C. polykrikoides*, *G. catenatum* and *P. bahamense* var. *compressum*.

1.3 Significance of the study

This research is necessary due to the lack of information available on the HABs cases in Malaysian waters, specifically in Sabah, such as factors affecting the bloom and the mechanisms of HABs occurrences. As the occurrences of HABs are observed yearly, it is important to understand the dynamic of the blooms. This includes the causal factors that contribute to the growth of these HAB species.

Intensive monitoring is required in order to update the recent status on the temporal and spatial distribution of the HABs species along Kota Kinabalu coastal waters. Besides, this study will provide a baseline data of the favourable condition for these HABs species to bloom.

CHAPTER 2

LITERATURE REVIEW

2.1 Harmful algal blooms (HABs)

2.1.1 History of HABs occurrence

Historically, algal blooms have been described in the logs of early mariners such as Captains James Cook and George Vancouver as poisonous fish and shellfish and discolouration of seawater (Prakash *et al.*, 1971). In July 1774, while a royal naval battleship HMS Resolution was off the coast of Vanuatu in the Pacific Ocean, several of Captain James Cook's crew became sick after consuming freshly caught red fish. The symptoms such as gastrointestinal disturbance, followed by some neurology and cardiovascular of ciguatera poisoning were observed (Vancouver, 1798; Doherty, 2005). Ciguatera is a form of poisoning that occurs after consuming tropical and subtropical smaller reef fish which feed on toxic dinoflagellates, *Gambierdiscus toxicus* (Achaibar *et al.*, 2007).

In 1793, fatal cases caused by shellfish poisoning had been reported by Captain George Vancouver, when he and his crew landed on Poison Island in Canada (known as British Columbia during that time). They noted that it was taboo for local Indian tribes to consume shellfish when the seawater became bioluminescent due to algal blooms (UNESCO, 2006).

These HAB occurrences in Vanuatu and Canada are examples of the earliest harmful algal blooms (HABs) cases recorded worldwide. This proved that HABs have occurred for such a long time and now become a global concern due to it damaging, losses and effects to aquaculture and food security.

2.1.2 Definition of HABs

Harmful algal blooms have become the preferred scientific term rather than red tides because these outbreaks do not have any relation to the tides, and they may or may not colour the water red (Sverdup *et al.*, 2003). Besides, some species of algae may bloom and colour the water but these blooms are not harmful to human and other organisms. Thus, HABs are defined as propagation in phytoplankton abundance, resulting in transformation of sea water surface colour. Besides discolouration of surface seawater, these algae have potential to produce toxin that can affect human or organism's health (Boesch *et al*., 1997).

Phytoplanktons are mainly unicellular plant-liked organisms and autotrophic (self-feeding), but some are heterotrophic (non-pigmented species that consume organic substrate) or mixotrophic (can do both; autotrophic and heterotrophic). The most important groups belonging to the marine phytoplankton are the diatoms, dinoflagellates and cyanobacteria. Of the 5000 species of marine phytoplankton identified, about 300 species can produce HABs and mostly are dinoflagellates (UNESCO, 2006). Dinoflagellates have red to green pigments and can exist at lower light level because they can both photosynthesize like a plant and ingest organic materials like an animal (mixotrophic) (Sverdup *et al*., 2003). Some of these dinoflagellates species are capable to discolour the seawater surface when they proliferate up to millions cells per litre and cause harm, for example *Cochlodinium polykrikoides* (Gárate-Lizárraga, 2004), *Alexandrium* spp., *Karenia brevis* and *Noctiluca* spp. (Boesch *et al*., 1997). However, there is no colour visible in other harmful species, such as the chlorophyll-free dinoflagellate *Pfiesteria piscicida* and *Dinophysis* species (Jacobson & Anderson, 1994) but both of these species can produce toxin at very low densities, generally less than 1,000 cells per litre (Burkholder & Glasgow, 1997; Smayda, 1997).

Formation of the phytoplankton bloom is actually a good process to the environment as it enhances biological productivity, by producing about 80 % of the oxygen that we breathe (Bhat *et al*., 2006). But the bloom becomes an issue when toxins are produced, which lead to fish mortality and losses to aquaculture and fisheries industries. These blooms also causes hazard to human health when humans accidentally consume contaminated finfish and shellfish and some of these blooms can also create anoxic condition (Boesch *et al*., 1997).

2.1.3 Hazards of HABs

HABs species cause damages and hazards to fishery resources, human health and marine ecosystem in many ways. Firstly, HABs species which is able to produce toxin can directly cause mass mortalities of wild and cultured shellfish and finfish (Maclean, 1979; Adnan, 1993; Kim, 1997). Secondly, it can also cause mortality when the toxin from HABs species move through food chain, affecting zooplankton, larvae, adult fish and even on birds, marine mammals and humans itself (Geraci *et al.*, 1989; Anderson & White, 1989; Shumway, 1995). Lastly, the blooms may affect on marine ecology by forming an anoxia/hypoxia condition (oxygen depletion).

The most important issues come from the HABs species that produce toxin. Hallegraeff (1993) has mentioned that about 300 species of marine phytoplankton can form bloom and out of these, 80 species tend to produce potent toxins. There are four major adverse effects caused by the toxins through shellfish i.e. paralytic shellfish poisoning (PSP), diarrhetic shellfish poisoning (DSP), neurotoxic shellfish poisoning (NSP) and amnesic shellfish poisoning (ASP). Those various illnesses caused by HABs species are summarized in Table 2.1.

Some HABs species do not produce toxins, but is able to cause harm to marine organisms by creating an anoxia/hypoxia condition (oxygen – deficient). This happens through decaying of phytoplankton resulting in a high biological oxygen demand (BOD), thus causing fish mortality. Low concentration of dissolved oxygen occurs mainly in the morning when the respiration rate of large quantities of phytoplankton are highest and decaying process are rapid (Smayda, 1979). During the *Ceratium* blooms in Canadian Lakes, school of fish were found dead through suffocation (Nicholls *et al.*, 1980). Other cases of fish mortalities due to oxygen deficiency also have been reported in the coast of Italy (Froglia, 1970) and Australia (Hallegraeff & Bolch, 1992).

Table 2.1: Illnesses caused by HABs species

Syndrome	Source	Symptoms	Causative species	Toxins produced
Paralytic shellfish Poisoning (PSP)	Clams, scallops, oysters, mussels & some coral reef gastropod	Tingling, numbness, giddiness, muscular paralysis, fever, possible respiratory arrest	*Pyrodinium bahamense, Gymnodinium catenatum,*	Saxitoxin & its derivatives
Diarrhetic shellfish Poisoning (DSP)	Scallops, mussels, short-necked clams	Vomiting, diarrhoea, nausea	*Dinophysis spp. Prorocentrum spp.*	Okadaic acid
Neurotoxin shellfish Poisoning (NSP)	clams, scallops, mussels, oysters	Neurological symptoms, vomiting, muscular aches, eye irritation dizziness & tingling of mouth	*Gymnodinium sp. Cochlodinium sp.*	Brevetoxins
Amnesic shellfish Poisoning (ASP)	clams, mussels, scallops	Nausea, vomiting, chronic memory loss & abdominal cramps	*Pseudo-nitzshia spp.*	Domoic acid & derivatives

Source: Bhat *et. al.*, (2006)