

## Red tides and other harmful algal blooms in Japan

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### History of HAB investigations in Japan

In Japan, one of the old historical books entitled “The History of Great Japan” (Dai Nippon Shi), edited more than 300 years ago, described 16 cases of red tide, seven in freshwater and the rest in the marine environment. The oldest was an occurrence in 731 AD. A case that occurred in 1234 AD was reported to cause fish mass mortality and human fatalities after eating fishes. The first scientific study on red tides was made by Nishikawa in 1900 on *Noctiluca scintillans*. From 1900 to 1950 the number of reports on red tides was about 10-25 per decade, and after 1950 it gradually increased. In 1970s, the number jumped drastically to more than 200 cases per year along with the development of heavy industry in coastal terrestrial zone and fish aquaculture industry in marine embayments. Fish mass mortality cases with serious economic loss also increased. After 1970, effort and large budgets were devoted to the elucidation of the biology and ecology of red tide organisms and their blooming mechanisms, modeling and mitigation methods.

Two types of harmful algal blooms (HABs) are known in Japan. The first one is a noxious algal bloom associated with the mass mortality of marine organisms, especially fish in aquaculture cages and shellfish hanging from rafts. Most of the noxious blooms cause water discoloration, *i.e.* red tides, but less than 20% of the red tides, as the term is used here, cause harmful effects. Most management and scientific research efforts have been devoted to the harmful ones, but successive occurrences of different harmful

species such as *Chattonella*, *Gymnodinium* and *Heterocapsa* along with changing environmental oceanographic conditions, make people concerned, feeling that the fight against red tides is endless.

The second type of HAB is a toxic algal bloom causing contamination of shellfish, either PSP (paralytic shellfish poisoning) or DSP (diarrhetic shellfish poisoning). The amount of toxin in shellfish and the cell number of toxic dinoflagellates are monitored regularly by local governments. Closures of aquaculture areas for the harvesting and marketing of shellfish occur depending on the amount of toxin, not the toxic plankton concentration. Therefore not all blooms of toxic plankton are recorded as HAB occurrences. Toxic plankton blooms with toxin contamination lower than the permitted level are not recorded as HAB occurrences.

In some fishing villages in northern Japan, warning about possible toxin contamination might be passed on from ancestors and known among elderly local fishermen. There are several traditional folk tales such as “Do not eat shellfish during snow water runoff into the sea”, *i.e.* it warns that shellfish may become inedible in early spring. After modern aquaculture developed, fishermen tried to sell their products year round but closure of marketing happens often in the spring. Thus we notice the wisdom from the experience that led to the folk tale. Perhaps this indicates that toxin contamination of shellfish has repeatedly occurred almost every year over a long time, leading to many tragedies among the local people.

The first recorded cases that were undoubtedly PSP and DSP occurred in 1948 and 1976, respectively. In the early 1970s, shellfish aquaculture techniques were more fully developed and the aquaculture industry spread into various areas, mainly along the coast of northern Japan. But at the same time several cases of human poisoning (PSP and DSP) were also reported.

The Fisheries Agency together with the Ministry of Health and Welfare set guidelines for monitoring and marketing regulation. After implementation of the official monitoring program there was no poisoning by marine products sold in market.

### Red tides that harm marine life

The number of algal bloom occurrences in the western part of Japan (Seto Inland Sea, Kyushu area, Tosa Bay and Kumano-nada: for locations see Figure 1) is shown in Figure 2. In the Seto Inland Sea red tide observations were very rare in the 1950s. Then, they dramatically increased and reached a peak of about 300 per year in the mid-1970s. After this period, they gradually decreased and recently remained stable. The number is now around 100 per year. This is thought to be mainly due to governmental regulations to control eutrophication based on laws such as the law concerning special measures for conservation of the environment of the Seto Inland Sea ("Seto Inland Sea Law") enforced in 1973. On the other hand, in the other three areas, the number of red tide events seems not to have decreased in last two decades. The sighting ranges per annum are 60 to 110 in Kyushu area, 3 to 16 in Tosa Bay, and 0 to 9 in Kumano-nada.

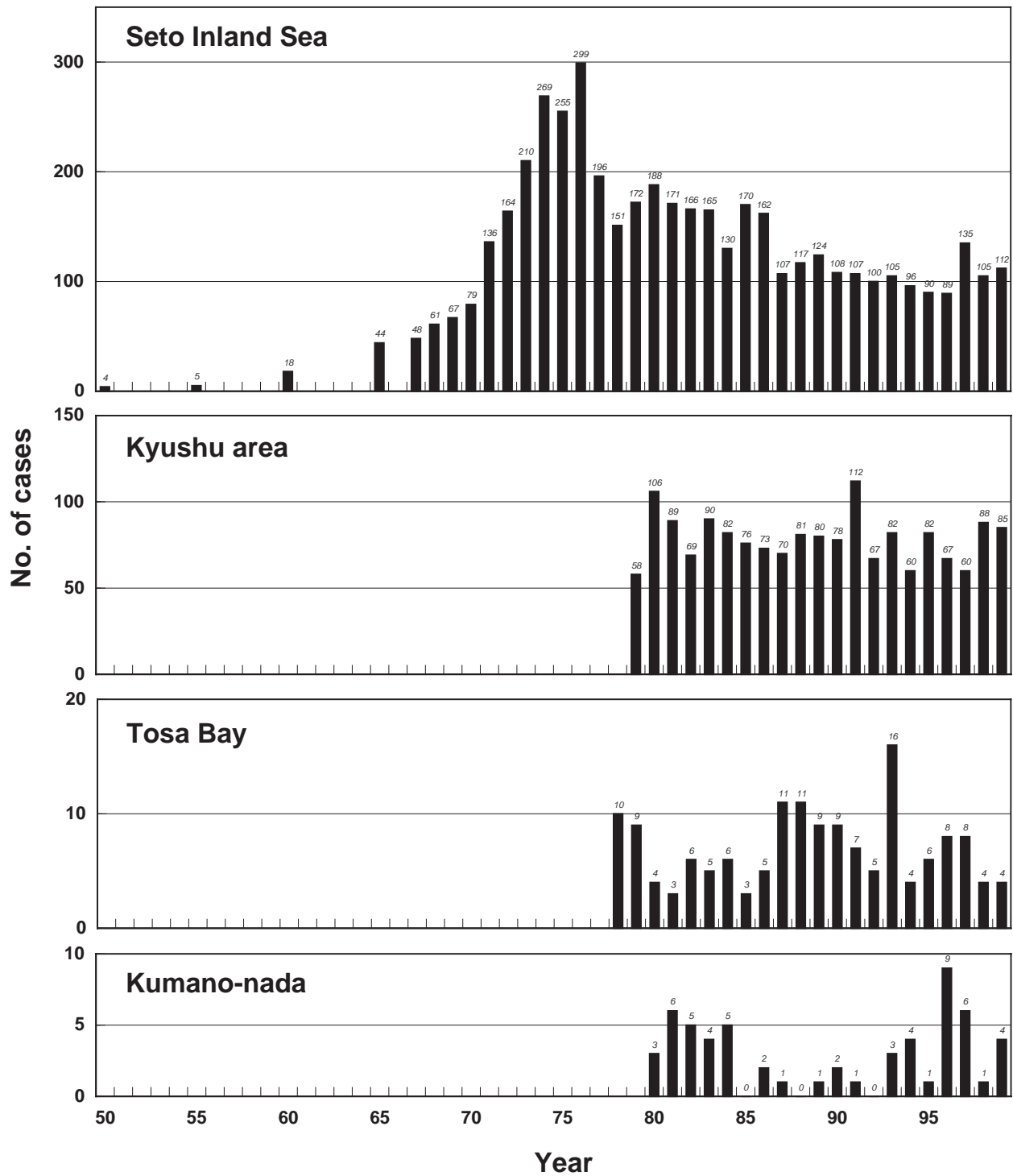
The number of red tides which have actually caused fisheries damage in Japan is shown in Figure 3. In last three decades, it seems to have been relatively stable. The numbers range from 20 to 50 per annum.



Fig. 1 Map of western Japan.

Figure 4 indicates the economic loss caused by red tides in Japan. Maximum economic loss was recorded in the summer of 1972. In that year the raphidoflagellate *Chattonella* red tide caused severe damage to cultured yellowtail in the eastern part of the Seto Inland Sea. The loss was about seven billion yen (about US\$70 million). Since then, the loss per year has decreased but, even in the 1990s, severe fisheries damage was sometimes recorded.

Table 1 demonstrates the major red tide species in Japan and the number of fisheries damage events due to these species. The most hazardous species are *Chattonella marina*, *C. antiqua* and dinoflagellate *Gymnodinium mikimotoi*. These three species mainly kill finfish and have frequently caused very severe fisheries damage of more than 100 million yen (about one million US\$). The dinoflagellate *Heterocapsa circularisquama* recorded for the first time from a small, semi-enclosed bay connected to Tosa Bay in 1988 was newly added to the list of hazardous species. This species only shows a harmful effect on shellfish, particularly on bivalves such as edible oysters, pearl oysters, short-necked clams and so on. Very severe fisheries damage caused by this species were occurred four times in last decade. In addition, the dinoflagellates *Cochlodinium polykrikoides* and *Gonyaulax polygramma* and raphidoflagellate *Heterosigma akashiwo* are also hazardous species in Japan.



**Fig. 2** Number of red tide occurrences per year. ND indicates no data. Refer to Figure 1 for names of area (Fisheries Agency 1973, 1999, 2000b).

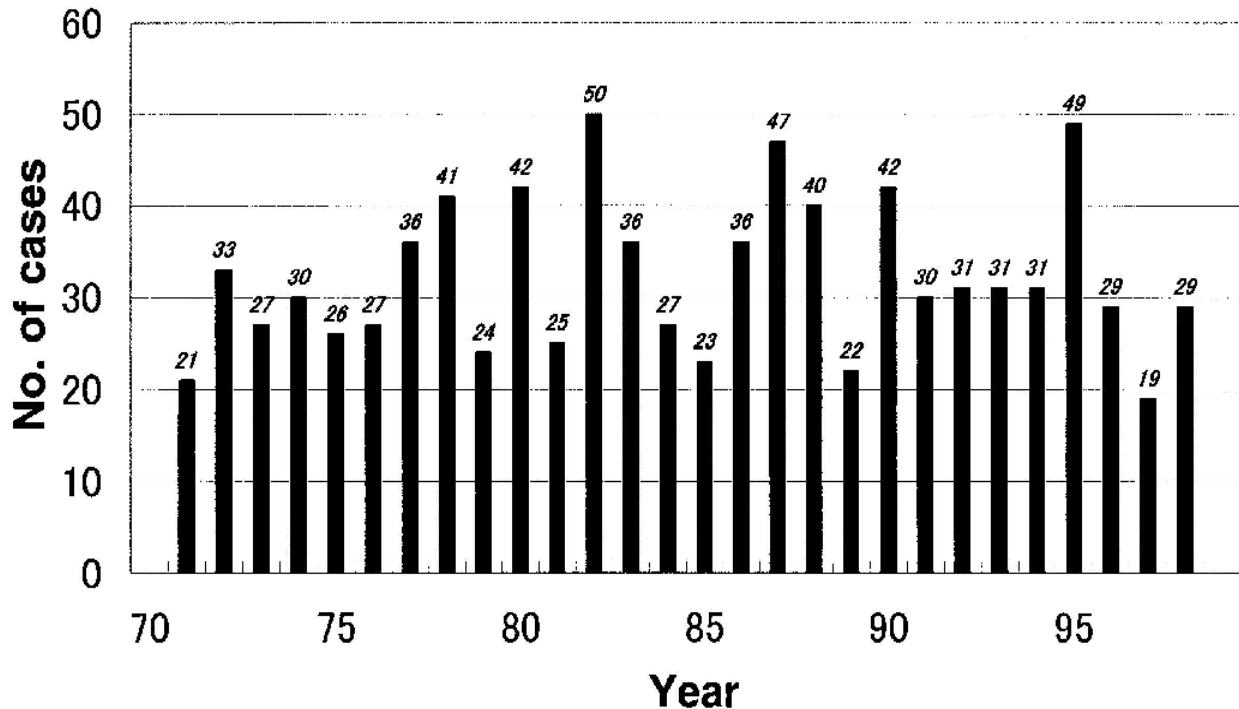
**Table 1** Major red tide species and the number of fisheries damaged by these species in Japan.

Causative organism (genus)	More than 100 million yen					10 ~ 100 million yen					Species name
	Seto Inland Sea (70~'98)	Kyushu area (79~'98)	Tosa Bay (78~'98)	Kumano-nada (80~'98)	Total	Seto Inland Sea (70~'98)	Kyushu area (79~'98)	Tosa Bay (78~'98)	Kumano-nada (80~'98)	Total	
<b>Dinophyceae</b>											
<i>Prorocentrum</i>	0	0	0	0	0	3(2)	1(0)	0	0	4(2)	<i>P. dentatum</i> etc.
<i>Cochlodinium</i>	0	0	0	0	0	0	12(1)	0	0	12(1)	<i>C. polykroides</i> etc.
<i>Gymnodinium</i>	16(2)	2(0)	1(0)	1(0)	20(2)	20(3)	9(1)	2(2)	0	31(6)	<i>G. mikimotoi</i> etc.
<i>Polykrikos</i>	0	0	0	0	0	1(1)	1(0)	0	0	2(1)	<i>Polykrikos</i> sp.
<i>Noctilca</i>	0	0	0	0	0	4(0)	0	0	0	4(0)	<i>N. scintillans</i> etc.
<i>Ceratium</i>	0	0	0	0	0	1(1)	1(0)	0	0	2(1)	<i>C. fusus</i> etc.
<i>Alexandrium</i>	0	0	0	0	0	0	1(0)	0	0	1(0)	<i>A. catenella</i>
<i>Gonyaulax</i>	2(0)	0	0	0	2(0)	0	0	0	0	0	<i>G. polygramma</i>
<i>Heterocapsa</i>	3(0)	1(0)	0	0	4(0)	1(0)	1(0)	0	0	2(0)	<i>H. circularisquama</i>
<b>Raphidophyceae</b>											
<i>Chatonella</i>	13(2)	5(0)	0	0	18(2)	11(1)	9(0)	2(1)	0	22(2)	<i>C. marina</i> , <i>C. antiqua</i> etc.
<i>Heterosigma</i>	0	1(0)	1(0)	0	2(0)	7(3)	0	2(1)	0	9(4)	<i>H. akashiwo</i>
<b>Chrysophyceae</b>											
<i>Distephanus</i>	0	1(0)	0	0	1(0)	0	0	0	0	0	<i>D. speculum</i>

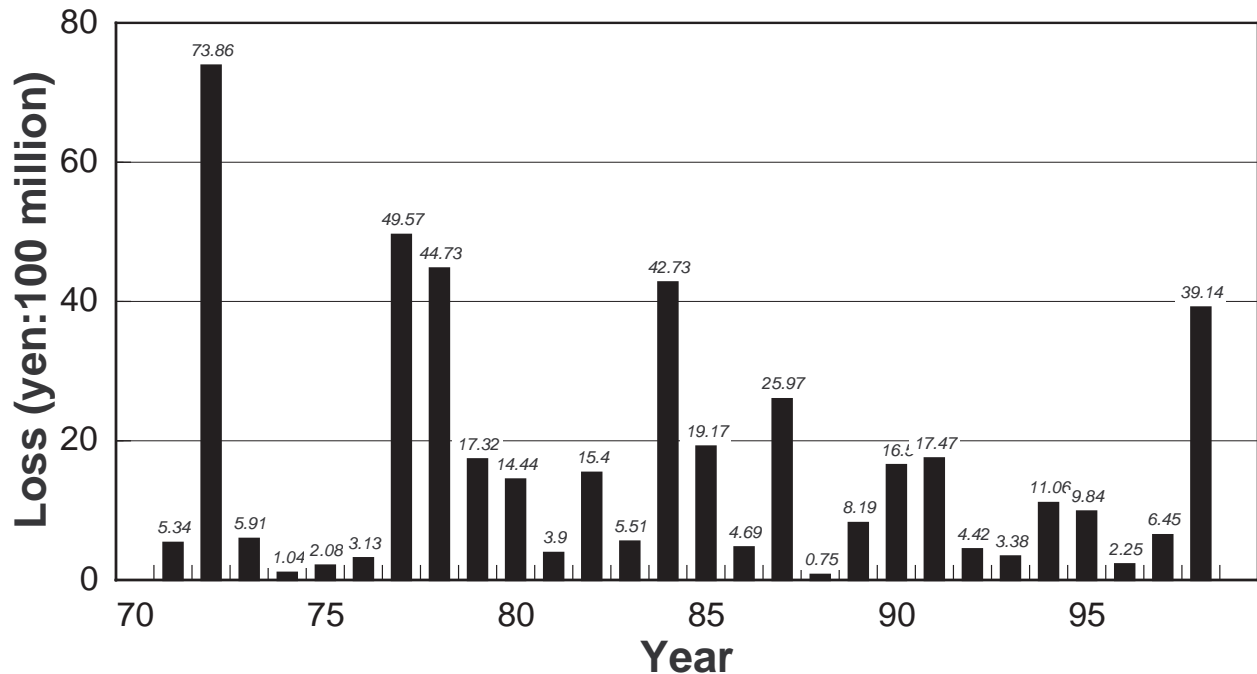
A dollar corresponds to 108 yen (Aug. 2000).

Figure in parentheses shows the number of the cases that more than 2 species caused a red tide (complex red tide).

(Fisheries agency 1999a, 2000a)



**Fig. 3** Number of red tides causing fisheries damage in Japan (Fisheries Agency 1996b).



**Fig. 4** Economic loss caused by red tide in Japan (Fisheries Agency 1999b).

#### Plankton blooms toxic to humans

Almost all microalgae responsible for various types of fish and shellfish poisoning of humans, such as PSP, DSP, ASP (amnesic shellfish poisoning), NSP (neurotoxic shellfish poisoning) and ciguatera (ciguatera fish poisoning), are present in Japanese coastal waters. Among them, PSP and DSP toxin contaminations in marine shellfish and filter feeders have been monitored officially since 1995 at 67 sea areas and 173 sampling stations

The official monitoring program started in 1978 in northern Japan and some other areas where shellfish aquaculture operated. Several trials to reduce toxins in shellfish, including the canning of scallops, were operated on a very limited scale. No fully effective countermeasure applicable to variety of affected marine products has been discovered and regular toxin monitoring is the only way to prevent the occurrence of poisoning. Modeling of bloom-forming mechanisms of the causative organisms and toxin accumulation in shellfish has also been tried in several ways in order to create a prediction system, but the current situation is still far from a good system, mostly because of

incomplete data acquisition and accumulation. This is the result of low cell number occurrences during blooming of causative plankton and the difficulty in analysis of shellfish physiology.

Among them *A. tamarense* and *A. catenella* appear in wider areas and often cause toxin contamination in scallops, mussels, clams and tunicates. In the first decade after the start of the monitoring program the area affected was mainly in northern Japan, but it gradually expanded to western Japan (Fig. 5). Closures of affected areas are decided by toxin quantification using the mouse bioassay, based on a modified AOAC method. Four MU (Mouse Units) of toxin in 1 gram of edible part of sample is the highest tolerable quarantine level. If the toxicity exceeds the level, closure of the area for harvesting the sample species is declared and three consecutive weeks of lower toxicity than that level is necessary to lift the closure. This means that it takes a minimum 21 days to re-open the market. The economic damage due to PSP toxin contamination is measured by the duration of closure. This monitoring and management system has been working successfully, and no PSP cases have

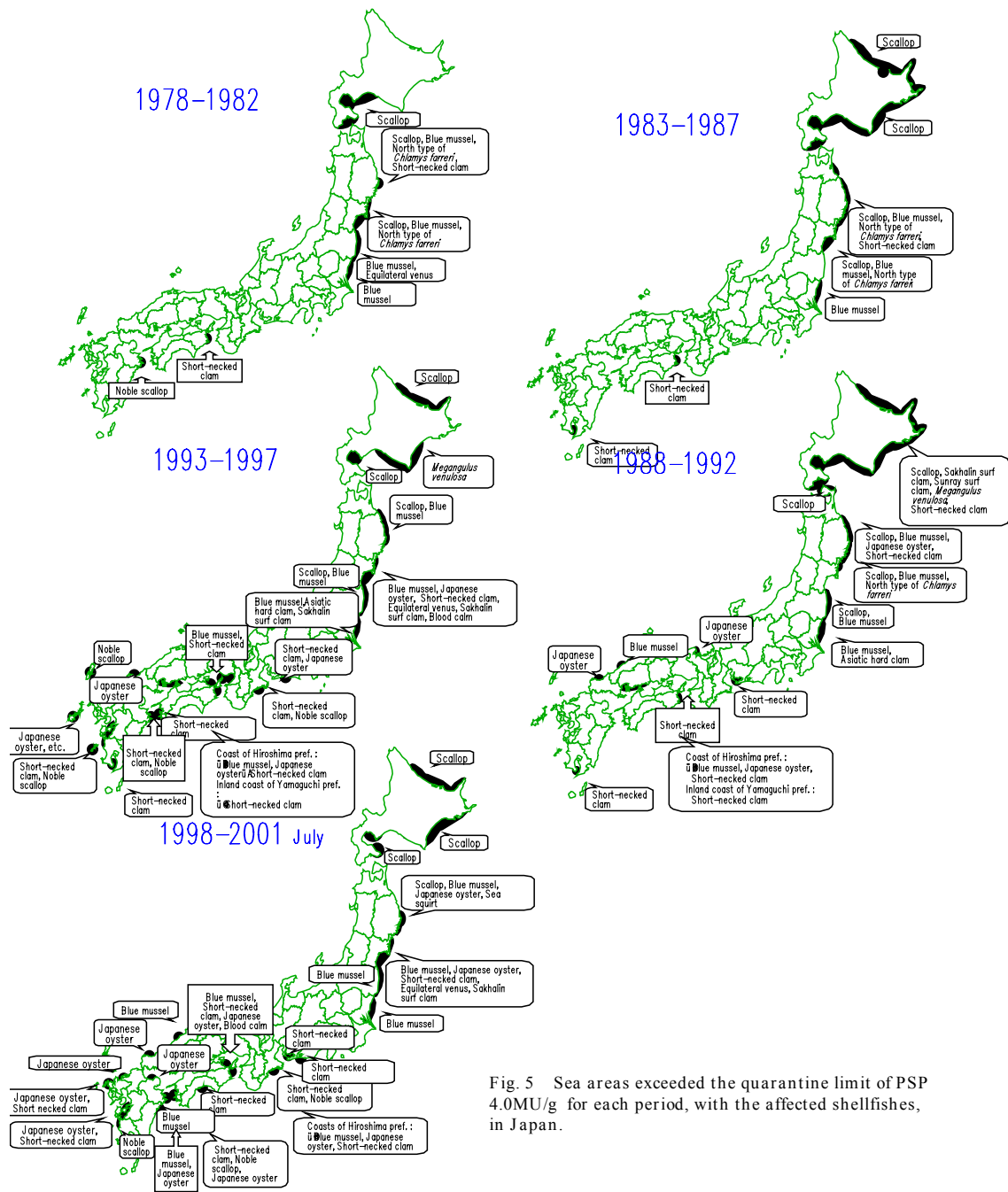


Fig. 5 Sea areas exceeded the quarantine limit of PSP 4.0MU/g for each period, with the affected shellfishes, in Japan.

**Fig. 5** Sea area exceeded the quarantine limit of PSP 4.0 MU/g for each period with the affected shellfishes in Japan.

occurred so far from eating shellfish sold in market, although some accidental cases have been reported from the consumption of shellfish collected by the victims.

### ***DSP causative plankton***

Dinoflagellates responsible for DSP contamination in Japanese waters are *Dinophysis fortii* and *D. acuminata*. Some other dinoflagellates such as *D. mitra* and *D. tripos* are also known to produce DSP toxins, but accumulation of toxins produced by these is less than the quarantine level. Closure of the affected area is decided by toxin quantification using mouse bioassay. A level of 0.05 MU in 1 gram of edible part of shellfish toxin is the highest permissive level. The management policy for the closure of aquaculture areas is same as that for PSP. After the operation of the monitoring program, the area affected was mainly in northern Japan (Fig. 6). In western Japan, *D. fortii* mysteriously never causes DSP toxin contamination in shellfish. After the establishment of the monitoring system, no poisoning case has been reported.

### ***Other toxic microalgae***

The diatom genus *Pseudo-nitzschia* is one of the commonest phytoplankton in Japanese waters, and about 10 different species bloom successively almost year round. Among them the production of domoic acid, which is responsible for ASP in North America, has been confirmed in *P. multiseriata*, *P. delicatissima* and *P. pseudodelicatissima*. However, domoic acid contamination in wild and aquacultured shellfish has not been detected so far, in spite of very intensive surveys conducted by the Japan Fisheries Agency and several universities. Therefore no regular monitoring program is set for ASP toxins.

For NSP and ciguatera, there is no monitoring program, although *Gymnodinium breve* and *Gambierdiscus toxicus*, causative organisms respectively for those types of poisoning, are sometimes found in western Japan, toxin contamination in shellfish and fish has not been detected.

## **Mitigation**

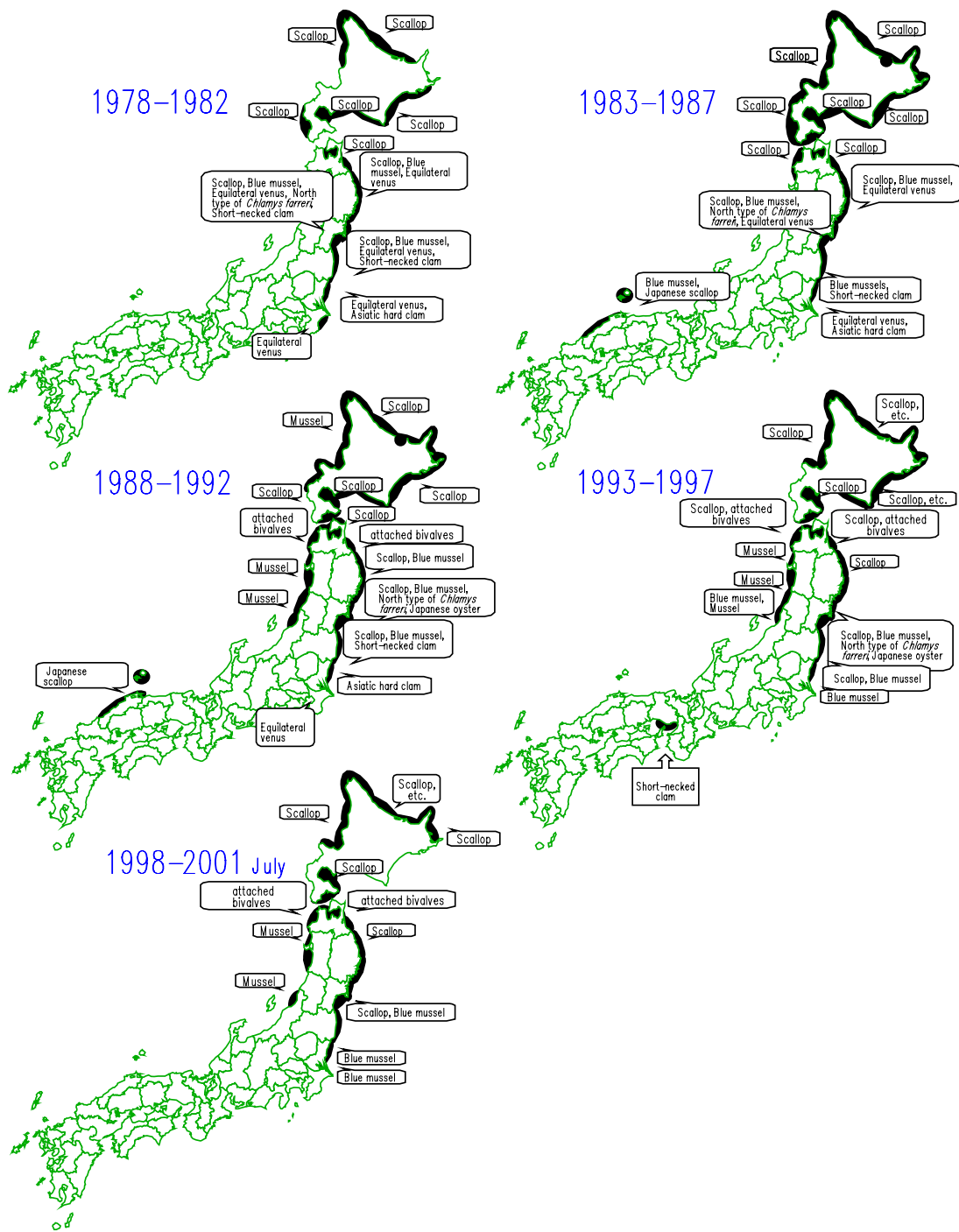
The incidence of fish killing red tides dramatically increased in frequency and scale in Japanese coastal waters, especially in the Seto Inland Sea, during 1960s and 1970s (see above), and huge fishery damages occurred repeatedly. The incidents have decreased thereafter as a long-term trend during the last two decades, but the number of known causative species has increased. The average economic loss associated with these red tides is about one billion Japanese yen per year.

Therefore, there is an urgent and compelling need for bloom mitigation strategies in aquaculture areas. In this section, plans for impact prevention of red tides attempted in the past are summarized, and the possibility of the utilization of diatoms as competitors for nutrients is described. Also the use of pathogenic microbes, such as bacteria and viruses, is introduced as these may be promising tools in reducing the impacts of red tides in the future.

Shellfish poisoning events (PSP and DSP) are also serious problems in Japan, but mitigation methods for these are not considered in this report because the strategies for these toxic blooms are expected to be different from those for red tides.

### **Plans for counteracting red tides attempted in the past in Japan**

Figure 7 illustrates the attempts at counteracting the impact of red tides in the past in Japan (Shirota 1989). The counteraction techniques are roughly divided into two categories, indirect and direct methods. Indirect methods are basically important as prevention of red tide occurrences on a long-term scale. Laws for regulations were established for the conservation of environments in the coastal sea such as the Seto Inland Sea. These laws have been effective in decreasing the direct discharge of polluted waste water into the coastal areas.

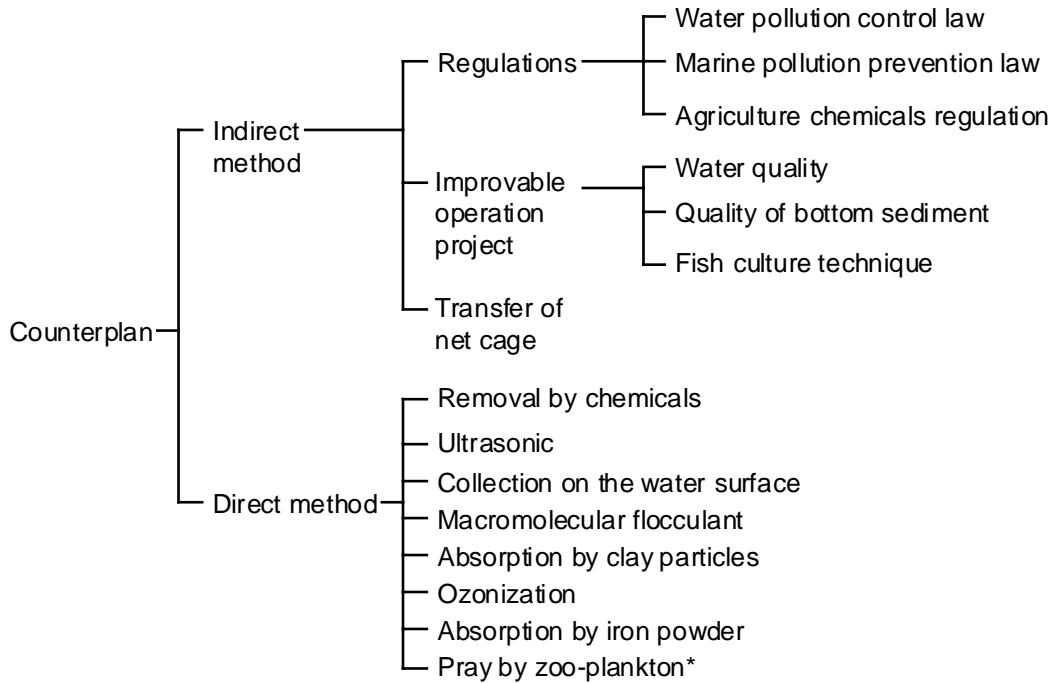


**Fig. 6** Sea area exceeded the quarantine limit of DSP 0.05 MU/g for each period with the affected shellfishes in Japan.



The resulting decline of nitrogen and phosphorus concentration in the water and sediments has led to a decrease of incidents of red tides. Developments of fish culture techniques, such as the use of "moist pellets" as a substitute for raw bait, keeping the proper scale and density of fish in aquaculture sites, and

transfer of net cages from red tide-prone areas, were effective in reducing the negative impacts of red tides. However, the most prevailing method is to stop feeding cultured fish just before and during red tides. This effectively reduces the mortality of fish in the cages, especially of yellowtail.



**Fig. 7** Counterplans at impact prevention of red tides attempted in the past in Japan (Shirota 1989).

As shown in Figure 7, rather many methods for the direct control of red tides had been attempted before 1985, but no physical and chemical control was successful as a whole. In other words, nature is beyond our direct control. Thereafter, these chemical and physical control options have received little attention. Shirota (1989) suggested that one promising strategy could be to treat red tides with flocculants such as clay, which scavenge particles, including algal cells, from seawater and carry them to bottom sediments. The feasibility of the treatment with clay has also been vigorously investigated in Korea in recent years. Field trials near fish farms have been successful to some extent. However, before this application can be implemented, considerable study is essential to determine the fate and effects of sedimented cells and toxins on benthic animals

and the collateral mortality of co-occurring planktonic organisms. Also, the decomposition of sedimented biomass and resulting oxygen depletion needs to be determined (SCOR-IOC 1998).

At present in Japan, treatment with clay is not practically applied because of high costs and the possible bad effects on marine organisms mentioned above.

**Biological control of red tides**

The biological control of red tides by using copepods and bivalves such as oysters has been examined, but the results were minimal because of the huge scale of red tides (Shirota 1989). Before 1989, microorganisms such as bacteria, viruses, parasites, and harmless phytoplankton

such as diatoms, had not been investigated for the application to biological control of red tides except for a few studies on bacteria. These microorganisms appear to be promising control agents against red tides, as they can be abundant in marine ecosystems, proliferate rapidly, and sometimes are host-specific (SCOR-IOC 1998). Basic studies have been conducted on biological control of red tides using microorganisms in Japan during the last decade. Feasibility is discussed in the following sections on diatoms, bacteria, and viruses, and some possible mitigation strategies are proposed.

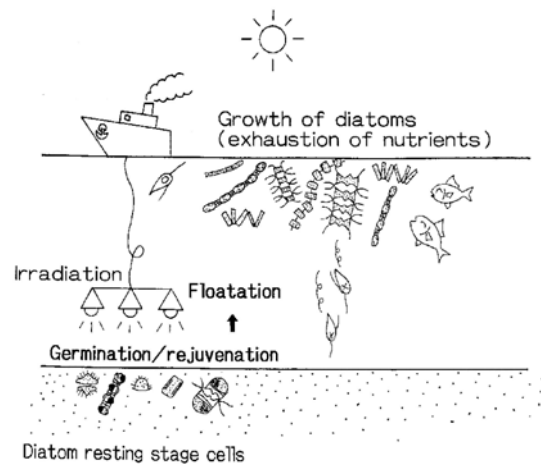
### ***Prevention of red tides by diatoms***

Aiming at controlling red tides due to *Chattonella* spp. (*C. antiqua* and *C. marina*) by using diatoms, a research project sponsored by the Environment Agency of Japan, "Technical development on the ecological control of noxious red tides", was conducted mainly by scientists of the Nansei National Fisheries Research Institute of Fisheries Agency from 1989 to 1993.

It is known empirically that *Chattonella* red tides occur when diatoms are scarce in surface water (Imai *et al.* 1998b). Diatoms (mainly Centrales) can dominate over *Chattonella* spp. due to their higher growth rates. However, diatoms form resting spores under conditions of nutrient limitation, especially of nitrogen, and they rapidly sink to the sea bottom. Accordingly, stratification and resulting exhaustion of nutrients will contribute to the formation of diatom resting spores. A timely mixing event, resulting in nutrient supply to the surface layer, after an appropriate stratification, which causes the nutrient depletion and sinking and/or inactivation of diatoms, is thought to be essential for the occurrence of *Chattonella* red tides.

*Chattonella* red tides are seeded by the germination of cysts (Imai and Itoh 1987). Diatom resting cells occur at densities of  $10^3$ - $10^6$  /g (wet sediment) in the coastal sea bottom (Itakura *et al.* 1997). *Chattonella* cysts can germinate in the dark at the sea bottom, but diatom resting cells need light for germination

and/or rejuvenation. If a sufficient intensity of light is provided to the diatom resting stage cells on the sea bottom, they are expected to germinate vigorously, and vegetative cells will be supplied to the surface layer (Fig. 8). Growth of diatoms will exhaust nutrients in the surface layer, which will reduce the growth of *Chattonella*. The crucial point of this mitigation strategy (prevention of *Chattonella* red tides) is the timing of irradiation to the sea bottom. The best timing is just after the supply of nutrients to the surface by a mixing event with a low cell density of *Chattonella* in a closed embayment.



**Fig. 8** A possible mitigation strategy of red tides using diatoms. Light is irradiated to the sea bottom in order to accelerate the germination and/or rejuvenation of diatom resting stage cells. Resulting diatom vegetative cells will grow and exhaust nutrients in the surface layer, and suppress the growth of *Chattonella*.

### ***Prevention of red tides by algicidal bacteria***

Aiming at controlling red tides by the use of bacteria, several research projects sponsored by the Fisheries Agency were conducted by scientists of several universities and the National Fisheries Research Institute from 1989 to 1999. Temporal fluctuations of algicidal microorganisms acting on the raphidophytes *C. antiqua* and *H. akashiwo*, were studied in northern Hiroshima Bay in the Seto Inland Sea (Imai *et al.* 1998a). The dynamics of *H.*

*akashiwo* killers revealed a close relationship with that of *H. akashiwo* populations (Fig. 9). *H. akashiwo* killers followed the increase of *H. akashiwo* cells, reaching a maximal level after the beginning of the decline of *H. akashiwo*. They maintained a high level for at least one week after the crash of bloom, and then decreased. *C. antiqua* killers consistently remained at low densities during the period of the *H. akashiwo* red tides. This result clearly indicates that algicidal microorganisms specifically associated with the occurrence and crash of *H. akashiwo* red tides, and contributed to the rapid termination of the red tides in the coastal seas.

Table 2 shows the list of algicidal bacteria isolated from the Japanese coastal seas (Yoshinaga 2000). These bacteria were classified phylogenetically using a database of SSU rDNA. Many algicidal bacteria are new species. The majority of algicidal bacteria are categorized into two groups,  $\gamma$ -proteobacteria (mainly the genera *Alteromonas* and *Pseudoalteromonas*) and a Cytophaga / Flexibacter / Bacteroides (CFB) group (mainly the genus *Cytophaga*). The algicidal bacteria, belonging to CFB group, are generally of the direct attack type, and the  $\gamma$ -proteobacteria are involved in the extracellular production of algicidal matter. SSU rDNA will be used for the development of PCR primers and DNA-probes for FISH (Fluorescence In Situ Hybridization) for specific detection of these algicidal bacteria.

A new aspect of ecology of algicidal bacteria has been the discovery that huge numbers of algicidal microorganisms (mainly bacteria) associate on the surface of macroalgae such as *Ulva* sp. (Chlorophyta) and *Gelidium* sp. (Rhodophyta) (Imai *et al.* in preparation). Maximum numbers of about  $10^5 - 10^6 \text{ g}^{-1}$  (wet weight) were detected for *Gymnodinium mikimotoi* (Dinophyceae), *Fibrocapsa japonica* (Raphidophyceae), and *H. akashiwo* (Raphidophyceae) (Fig. 10).

Algicidal microorganisms were also abundant in seawater collected at a seaweed bed in Obama Bay and Osaka Bay. Based on these findings,

we propose a new prevention strategy of red tides using macroalgae in aquaculture areas (Fig. 11). Co-culturing of *Gelidium* sp. and/or *Ulva* sp. and finfish such as red sea bream or yellowtail is proposed as an effective measure in cage culture. Many algicidal bacteria will be continually released from the surface of macroalgae to seawater, and contribute to reduce cell densities of phytoplankton, including harmful species.

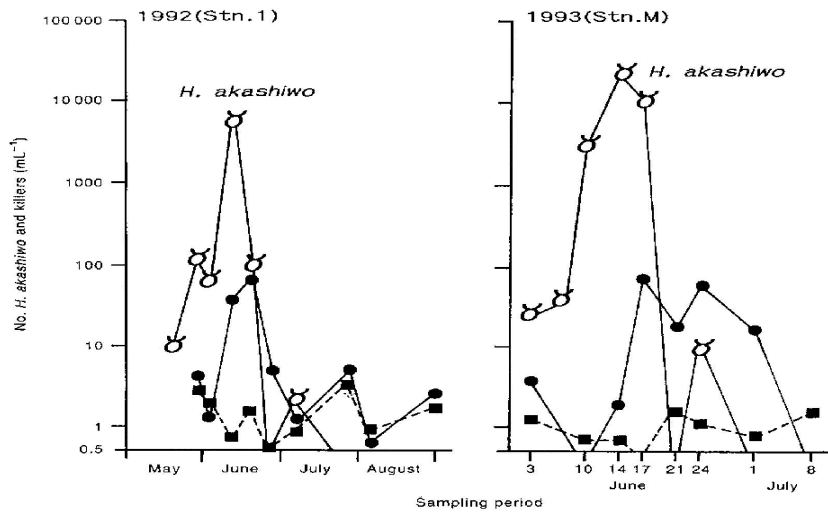
Consequently, these bacteria can probably play an important role in preventing occurrences of noxious red tides. This strategy may be effective in semi-enclosed and small inlets. Feasibility studies will be needed for the practical use of macroalgae in the future. The most excellent merit aspect of this strategy is that macroalgae have no negative image for aquaculture farmers and consumers. Moreover, *Ulva* sp. is actually being utilized as supplementary food for red seabream in some cage cultures in Mie and Ehime Prefectures in Japan

#### ***Extermination of red tides by algicidal viruses***

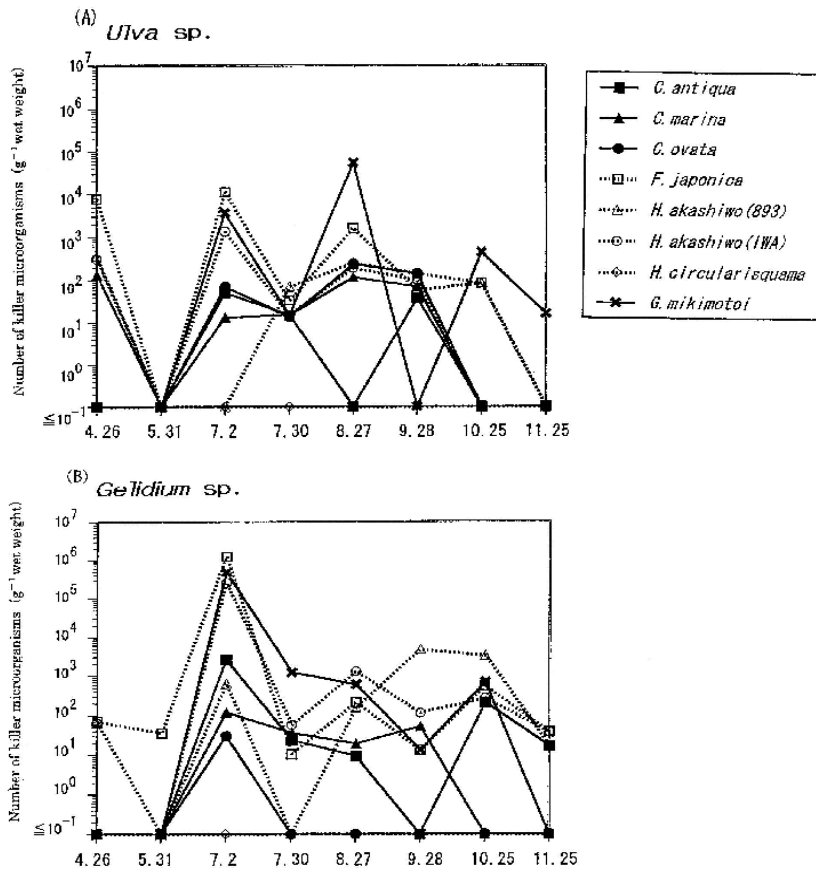
Algicidal viruses appear to be promising tools for the reduction or extermination of noxious red tides by virtue of their high host specificity and high replication rate.

In *H. akashiwo* red tides, HaV virus was found and isolated in the final stage of the red tide (Nagasaki *et al.* 1994, Nagasaki and Yamaguchi 1997). Viruses were also found and isolated from the notorious bivalve-killer dinoflagellate *Heterocapsa circularisquama* (Tarutani *et al.* submitted). For the encounter of viruses and host algal cells, relatively high cell densities of red tide organisms are thought to be essential.

Hence, viruses cannot be used as a tool for the prevention of red tides but as an effective tool for the extermination of red tides. As viruses have an extremely high replication rate, they can be utilized as a trump card after the failure of the prevention of red tides by diatoms and algicidal bacteria at earlier stages.



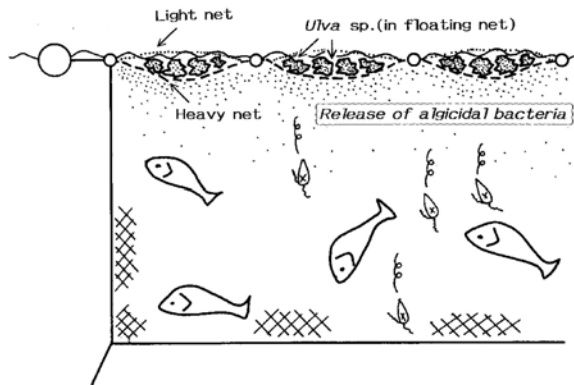
**Fig. 9** Fluctuations in densities of *Heterosigma akashiwo* (o), *H. akashiwo* killers (•), and *C. antiqua* killers (■) in the surface waters collected in northern Hiroshima Bay, the Seto Inland Sea (Imai *et al.* 1998a).



**Fig. 10** Seasonal fluctuations in densities of killer microorganisms associated with seaweeds against 8 red tide flagellates (Imai *et al.* in preparation). Algal samples were collected at the station located in macrophytic bed in Osaka Bay.

**Table 2** Algicidal microorganisms (mainly bacteria) isolated from the Japanese coastal seas.

Species and strain	Isolated year	Isolated area	Host microalga for isolation	Accession No. in DDBJ	Remarks
<i>Cytophaga</i> sp. /J18/M01	1990	Harima-Nada	<i>Chattonella antiqua</i>	AB017046	Direct attack
<i>Alteromonas</i> sp. /S	1991	Hiroshima Bay	<i>Chattonella antiqua</i>	AB040464	Extracellular production of algicidal matter
<i>Alteromonas</i> sp. /K	1991	Hiroshima Bay	<i>Chattonella antiqua</i>	AB040465	Extracellular production of algicidal matter
<i>Alteromonas</i> sp. /D	1991	Hiroshima Bay	<i>Chattonella antiqua</i>	AB040466	Extracellular production of algicidal matter
<i>Pseudoalteromonas</i> sp. /R	1991	Hiroshima Bay	<i>Chattonella antiqua</i>	AB040467	Extracellular production of algicidal matter
<i>Alteromonas</i> sp. /GY21	1994	Hiroshima bay	<i>Heterosigma akashiwo</i>	AB001335	Extracellular production of algicidal matter
<i>Alteromonas</i> sp. /GY27	1994	Hiroshima Bay	<i>Heterosigma akashiwo</i>	AB001334	Extracellular production of algicidal matter
<i>Cytophaga</i> sp. /GY9	1994	Hiroshima Bay	<i>Heterosigma akashiwo</i>	AB001332	Extracellular production of algicidal matter
<i>Flavobacterium</i> sp. /5N-3	1989	Uranouchi Inlet, Kochi Pref.	<i>Gymnodinium mikimotoi</i>	AB017597	Extracellular production of algicidal matter
<i>Alteromonas</i> sp. /E401	1991	Tanabe Bay	<i>Gymnodinium mikimotoi</i>	AB004313	Extracellular production of algicidal matter
$\gamma$ -proteobacterium /EHK-1	1999	Etauhi, Hiroshima bay	<i>Heterocapsa circularisquama</i>	AF228694	
<i>Cytophaga</i> sp. /AA8-2	1995	Ago Bay, Mie Pref	<i>Heterocapsa circularisquama</i>	AB017047	Direct attack type, the same as <i>Cytophaga</i> sp. J18/M01
<i>Cytophaga</i> sp. /AA8-3	1995	Ago Bay, Mie Pref	<i>Heterocapsa circularisquama</i>	AB017048	Direct attack type, the same as <i>Cytophaga</i> sp. J18/M01
<i>Pseudoalteromonas</i> sp. /A25	1994	Ariake Sea, Fukuoka Pref.	<i>Skeletonema costatum</i>	AF227237	Extracellular production of algicidal matter, specific to diatoms
<i>Pseudoalteromonas</i> sp. /A28	1994	Ariake Sea	<i>Skeletonema costatum</i>	AF227238	Extracellular production (protease) of algicidal matter, specific to diatoms, the same as A27, A29, A30, A42
<i>Cytophaga</i> sp. /A5	1990	Ariake Sea	<i>Skeletonema costatum</i>	AB008031	The same as A11, A14, A15, A20
<i>Cytophaga</i> sp. /A11	1990	Ariake Sea	<i>Skeletonema costatum</i>	AB008032	The same as A5, A14, A15, A20
<i>Cytophaga</i> sp. /A14	1990	Ariake Sea	<i>Skeletonema costatum</i>	AB008033	The same as A5, A11, A15, A20
<i>Cytophaga</i> sp. /A15	1990	Ariake Sea	<i>Skeletonema costatum</i>	AB008034	The same as A5, A11, A14, A20
<i>Cytophaga</i> sp. /A20	1990	Ariake Sea	<i>Skeletonema costatum</i>	AB008035	The same as A5, A11, A14, A15
<i>Cytophaga</i> sp. /A38	1994	Ariake Sea	<i>Skeletonema costatum</i>	AB008036	
<i>Cytophaga</i> sp. /A12	1992	Ariake Sea	<i>Skeletonema costatum</i>	AB008037	The same as A32, A35, A41
<i>Cytophaga</i> sp. /A32	1992	Ariake Sea	<i>Skeletonema costatum</i>	AB008038	The same as A12, A35, A41
<i>Cytophaga</i> sp. /A35	1992	Ariake Sea	<i>Skeletonema costatum</i>	AB008039	The same as A12, A32, A41
<i>Cytophaga</i> sp. /A41	1992	Ariake Sea	<i>Skeletonema costatum</i>	AB008040	The same as A12, A32, A35
<i>Flavobacterium</i> sp. /A16	1992	Ariake Sea	<i>Skeletonema costatum</i>	AB008041	
<i>Flavobacterium</i> sp. /A17	1992	Ariake Sea	<i>Skeletonema costatum</i>	AB008042	
<i>Flavobacterium</i> sp. /A43	1994	Ariake Sea	<i>Skeletonema costatum</i>	AB008043	
<i>Flexibacter</i> sp. /A37	1994	Ariake Sea	<i>Skeletonema costatum</i>	AB008044	
<i>Flexibacter</i> sp. /A45	1994	Ariake Sea	<i>Skeletonema costatum</i>	AB008045	
<i>Cytophaga</i> sp. /A23	1994	Ariake Sea	<i>Skeletonema costatum</i>	AB008046	
<i>Saprospira</i> /SS90-1	1990	Shrimp culture pond, Kagoshima Pref.	<i>Chaetoceros ceratosporum</i>	unavailable	Filamentous, multicellular, carotenoid, cell lysis by direct contact
<i>Saprospira</i> /SS91-40	1991	Shrimp culture pond, Kagoshima Pref.	<i>Chaetoceros ceratosporum</i>	unavailable	Filamentous, multicellular, carotenoid, cell lysis by direct contact
<i>Saprospira</i> /SS92-11	1992	Kagoshima Bay	<i>Chaetoceros ceratosporum</i>	unavailable	Filamentous, multicellular, carotenoid, cell lysis by direct contact
<i>Saprospira</i> /SS95-4	1995	Kagoshima Bay	<i>Chaetoceros ceratosporum</i>	unavailable	Filamentous, multicellular, carotenoid, cell lysis by direct contact
<i>Labyrinthula</i> /L93-3	1993	Seagrass, Kagoshima Bay	<i>Chaetoceros ceratosporum</i>	unavailable	Eucaryotes, cell lysis by direct contact
<i>Labyrinthula</i> /L95-1	1995	Seaweed, Kagoshima Bay	<i>Chaetoceros ceratosporum</i>	unavailable	Eucaryotes, cell lysis by direct contact



**Fig. 11** A possible prevention strategy of red tide by using macroalgae in aquaculture areas. Co-culture of macroalgae and finfish is proposed in the same cage.

Diatoms have not yet been tried for the prevention of harmful red tides. Uncertainties about host specificity, pathogen stability, and environmental impacts such as negative effects of these microorganisms on higher organisms, must be examined before the practical utilization of microorganisms as tools for the prevention and control of noxious red tide occurrences.

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