Life cycle strategies and occurrences of red tides of *Heterosigma akashiwo* and *Chattonella* spp. in temperate coastal sea

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Global distribution of raphidophytes and fish-kill (Edvardsen and Imai 2006)
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1. Life cycle strategy of *Chattonella antiqua* and *C. marina*

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Life cycle strategy of *Chattonella* spp.
1. *Chattonella antiqua* and *C. marina* are the summer red tide flagellates.

2. The optimum combination of temperature and salinity
   
   - *C. antiqua*: 25°C and 25psu
   - *C. marina*: 25°C and 20psu

3. Survival range of temperature
   
   - *C. antiqua*: 11 ~ 31°C
   - *C. marina*: 13 ~ 31°C

∴ *C. antiqua* and *C. marina* can not overwinter as vegetative cells in the Seto Inland Sea, and form cysts for overwintering.
Life cycle of *Chattonella antiqua* and *C. marina* based on DNA microfluorometry (Yamaguchi and Imai 1994)

*Chattonella antiqua* and *C. marina* are the diploid organisms.
Cysts of *Chattonella antiqua* and *C. marina* (Imai and Itoh 1988)

A & B: *C. antiqua*

C & D: *C. marina*
MPN method for enumeration of *Chattonella* cysts having germination ability in sediments (Imai et al. 1984).

Feasible for the cysts without information on morphology.
Comparison of temperature characteristics of germination and maturation in *Chattonella* cysts.
Seasonal changes in germinability of *Chattonella* cysts in sediments collected at 3 points in Suo-Nada, the Seto Inland Sea (Imai and Itoh 1987).
Annual life cycle of *Chattonella* in the Seto Inland Sea, including vegetative cells and cyst phase. (Imai and Itoh 1987)
Summary for *Chattonella*

1. *Chattonella* spp. are diploid organisms (2n).
2. *Chattonella* has cyst stage for overwintering.
3. Cyst formation was induced by nutrient depletion such as nitrogen under low light conditions.
4. Cysts needs winter for maturation.
5. Cysts effectively germinate at 20°C or higher.
6. *in situ* germination of cysts show great seasonality.
7. Summer red tides are seeded by the germination of cysts in sea bottom.
8. Life cycle strategy of *Chattonella* is well adapted to temperate shallow coastal areas; changes between cysts and vegetative cells are easy.
Life cycle strategy of *Heterosigma akashiwo*
Sampling stations for H. akashiwo study in Hiroshima Bay, the Seto Inland Sea.
Cysts of *Heterosigma akashiwo* discovered from the sediments of the Seto Inland Sea (Imai et al. 1993)

Feasible for the cysts without information on morphology.
Distribution of cysts of *Heterosigma akashiwo* in bottom sediments of Hiroshima Bay, the Seto Inland Sea (Imai and Itakura 1991).

Cysts were abundant in the coast.
Effects of temperature on the germination of Heterosigma akashiwo cysts in sediments determined by the MPN method (Imai and Itakura 1999).

Vigorous germination at 15°C or higher temperature
Seasonal fluctuations of the highest cell densities of *H. akashiwo* cells in water columns at 3 stations in Hiroshima Bay (Imai and Itakura 1999).

Blooms in May ~ June with great seasonality
Seasonal fluctuations of germinable cysts (MPN) in the surface sediments (0 - 1 cm) of Hiroshima Bay (Imai and Itakura 1999).

Many cysts are always physiologically germinable.

Fig. 5 Heterosigma akashiwo. Seasonal fluctuations in the number of germinable H. akashiwo cysts in surface sediments (top 1-cm layer) collected at three stations in northern Hiroshima Bay. Enumeration by extinction dilution method.
Seasonal fluctuations of surface (open circle) and bottom (closed circle) water temperatures at 3 stations in Hiroshima Bay.
Weekly changes in water temperature, *H. akashiwo* in surface water, and the total *H. akashiwo* live cysts in surface sediments (0-1cm) at Stn.1 in Hiroshima Bay (Imai and Itakura 1998). Total live cysts increased just after the peak of bloom and needed 1 week for maturation.

Table 1. *Heterosigma akashiwo*. Numbers of the total cysts enumerated by the direct count method and the germinable cysts enumerated by the extinction dilution method in surface (top 1-cm depth) sediments collected at St.1 on 24 June and 1 July, 1993.

<table>
<thead>
<tr>
<th>Date</th>
<th>June 24</th>
<th>July 1</th>
</tr>
</thead>
<tbody>
<tr>
<td>Total cysts</td>
<td>5676</td>
<td>5240</td>
</tr>
<tr>
<td>Germinable cysts</td>
<td>419</td>
<td>4223</td>
</tr>
<tr>
<td>(cm⁻³ wet sediment)</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>
Heterosigma red tides usually occur at 20°C or higher with wide range of salinity (Honjo 1993).


Honjo (1993)
Process and key factors for the occurrence of *Heterosigma akashiwo* red tide (Smayda 1998).

Figure 2. General model of key processes influencing bloom outbreaks of *Heterosigma akashiwo*; see text for details.
Summary for *Heterosigma akashiwo*

1. *Heterosigma akashiwo* has cyst stage for outliving such as overwintering.
2. Cyst formation was induced at the end of blooms and completed in the dark (Itakura et al. 1996).
3. New cysts need ca. 1 week for maturation.
4. Cysts effectively germinate at 15°C or higher.
5. *in situ* germination of cysts can always occur.
6. Red tides show great seasonality, and hence seeded by the germination of cysts in sea bottom.
7. Life cycle strategy of *H. akashiwo* is also well adapted to temperate shallow coastal areas; changes between cysts and vegetative cells are easy.
How do harmful raphidophyte blooms occur predominantly over diatoms in coastal sea?
Change of blooms from diatoms to *H. akashiwo* and to diatoms.
# Growth parameters of diatoms and raphidophytes

<table>
<thead>
<tr>
<th>Species</th>
<th>Nutrients</th>
<th>$\mu_{\text{max}}$ (division/day)</th>
<th>$K_s$ ($\mu$M)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Diatoms</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Chaetoceros didymum</em></td>
<td>NO3-N</td>
<td>1.82</td>
<td>1.29</td>
</tr>
<tr>
<td></td>
<td>PO4-P</td>
<td>2.16</td>
<td>0.48</td>
</tr>
<tr>
<td><em>Ditylum brightwellii</em></td>
<td>NO3-N</td>
<td>2.02</td>
<td>0.73</td>
</tr>
<tr>
<td></td>
<td>PO4-P</td>
<td>2.00</td>
<td>0.16</td>
</tr>
<tr>
<td><em>Thalassiosira rotula</em></td>
<td>NO3-N</td>
<td>1.69</td>
<td>1.28</td>
</tr>
<tr>
<td></td>
<td>PO4-P</td>
<td>1.70</td>
<td>0.23</td>
</tr>
<tr>
<td><strong>Raphidophytes</strong></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td><em>Chattonella antiqua</em></td>
<td>NO3-N</td>
<td>0.74</td>
<td>0.87</td>
</tr>
<tr>
<td></td>
<td>PO4-P</td>
<td>0.74</td>
<td>0.29</td>
</tr>
<tr>
<td><em>Heterosigma akashiwo</em></td>
<td>NO3-N</td>
<td>1.7 – 1.9</td>
<td>1.99 - 2.45</td>
</tr>
<tr>
<td></td>
<td>PO4-P</td>
<td>1.7 – 1.9</td>
<td>1.00 – 1.98</td>
</tr>
</tbody>
</table>
Raphidophytes have cysts.
Diatoms have resting stage cells.
Seeds of diatoms and raphidophytes are abundant in sediments of coastal sea.

<table>
<thead>
<tr>
<th>Species</th>
<th>Abundance (cm⁻³ · wet sediment)</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Raphidophyceae</strong></td>
<td></td>
</tr>
<tr>
<td><em>Heterosigma akashiwo</em></td>
<td>5.6 × 10¹ ~ 2.9 × 10⁴</td>
</tr>
<tr>
<td><em>Chattonella</em> spp.</td>
<td>0 ~ 7.7 × 10²</td>
</tr>
<tr>
<td><strong>Dinophyceae</strong></td>
<td></td>
</tr>
<tr>
<td><em>Alexandrium</em> spp.</td>
<td>5.0 × 10¹ ~ 1.3 × 10³</td>
</tr>
<tr>
<td><strong>Bacillariophyceae</strong></td>
<td></td>
</tr>
<tr>
<td><em>S. costatum</em></td>
<td>8.0 × 10³ ~ 2.1 × 10⁶</td>
</tr>
<tr>
<td><em>Chaetoceros</em> spp.</td>
<td>2.7 × 10³ ~ 6.6 × 10⁵</td>
</tr>
<tr>
<td><em>Thalassiosira</em> spp.</td>
<td>3.7 × 10³ ~ 1.5 × 10⁵</td>
</tr>
</tbody>
</table>

Note: Data of flagellates cysts were compiled from Imai and Itakura (1991),¹ Imai et al. (1993),² Yamaguchi et al. (1995)³
Abundance of diatom resting stage cells (cm⁻³ · wet sediment) were calculated from the present data.
Germination of raphidophyte cysts in the dark

Chattonella spp.  
Heterosigma kashiwo

[Graphs showing germination over time for Chattonella spp. and Heterosigma kashiwo]
Diatom resting stage cells need light for germination and rejuvenation.

**Fig. 4-1.** A schematic representation of the role of diatom resting stage cell in the coastal environment.
Germination experiment

H. akashiwo cysts
330 / g sed. medt

Diatom resting stage cells
1.6 x 10^5 / g sed.

Light: 110µE/ m^2/ sec
14hL : 10hD

Temperature: 20ºC
Effects of light intensity on germination and/or rejuvenation of diatom resting stage cells.

Diatom resting stage cells need light for germination.

Open circle: total diatoms
Closed circle: *S. costatum*
Black triangle: *Chaetoceros*
Star: *Thalassiosira*
Comparison of the effects of light intensity to cysts and diatom resting stage cells on dominancy of \textit{H. akashiwo}.

In surface water:

1) Diatoms dominated in higher light.
2) \textit{H. akashiwo} dominated in low light
Vertical profiles of light intensity in coastal sea

Harima-Nada, Seto Inland Sea
*Chaetoceros* spp. form sores in low nitrogen concentration

*Chaetoceros curvusetus, C. distans, C. lauderi*

Harima-Nada: 
July 14 - 18, 1991
Diatom resting hypothesis

Nutrient depletion after stratification induce resting stages of diatoms.
1. Diatoms are stronger than raphidophytes in vegetative growth.
2. Raphidophytes have cyst stage and diatoms have resting stage in their life cycles.
3. Raphidophyte cysts can germinate in the dark and diatom resting stage cells need light for germination.
4. Nutrient depletion induce resting stage in diatoms.
5. Low light irradiation to sediments induced predominancy of raphidophytes in water columns.
6. The diatom resting hypothesis is presented for the mechanism of occurrences of raphidophyte red tides.
A proposal of idea for prevention of harmful blooms by bottom irradiation

No toxic effects on environments

Problems for feasibility
1. Timing, period, and intensity of irradiation
2. Scale of irradiation