

Studies on growth interactions between harmful red tide dinoflagellate *Karenia selliformis* and diatoms

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Title : Studies on growth interactions between harmful red tide dinoflagellate *Karenia selliformis* and diatoms
(有害赤潮渦鞭毛藻 *Karenia selliformis* と珪藻類の増殖相互作用に関する研究)

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Thesis Summary

This study aimed to investigate the competitive interactions in growth between the harmful dinoflagellate *Karenia selliformis*, which caused a large-scale red tide and severe damage to fisheries in 2021, and other phytoplankton species, particularly diatoms, in relation to red tide formation. Co-culture experiments were first conducted using *K. selliformis* and either *Thalassiosira hendeyi* or *Chaetoceros diadema*, both of which are commonly present in the same coastal waters. The results indicated that the species with higher cell density significantly suppressed the growth of the other. Numerical analyses using the Lotka-Volterra model supported these results. In addition, a pronounced inhibitory effect on growth was observed in the cell-free filtrate of *K. selliformis*, suggesting that allelopathic compounds released by *K. selliformis* were involved in the suppression. Moreover, direct cell-to-cell contact was found to further enhance the inhibitory effect, indicating that both allelopathy and physical interactions contribute to its competitive advantage.

To further investigate the role of environmental factors in modulating competitive interactions, *K. selliformis* and *C. diadema* were co-cultured under a range of temperatures (5, 10, 15, and 20°C) and light intensities (0, 20, 50, and 150 $\mu\text{mol m}^{-2} \text{s}^{-1}$). Across all temperature treatments, *K. selliformis* demonstrated a stronger competitive advantage, which became more pronounced with increasing temperature. Similarly, under all tested light intensities (except complete darkness), *K. selliformis* eliminated *C. diadema* within nine days, highlighting its superior competitiveness. Neither species survived under dark conditions.

Results from competition and environmental stress experiments confirmed that *K. selliformis* exerted broad inhibitory effects on diatoms under multiple conditions. To further elucidate its allelopathic spectrum, the filtrate of *K. selliformis* was tested against ten microalgal species, among which *Prorocentrum minimum* and *Thalassiosira pseudonana* were most strongly affected. Subsequent assays with fractionated filtrates revealed that low molecular weight (<5000 Da) and hydrophobic fractions exhibited the greatest inhibitory effects on *T. pseudonana*, suggesting that allelopathic compounds are mainly concentrated in these fractions. Additionally, the inhibitory activity remained potent after heat treatment at 100°C, indicating the thermal stability of the allelopathic substances. Exposure experiments showed that the filtrate reduced *T. pseudonana* photosynthetic efficiency by approximately 50% within 60 minutes (as indicated by a sharp drop in Fv/Fm).

These findings strongly suggest that *K. selliformis* formed dominant red tides through the release of allelopathic compounds that inhibit the growth of competing algal species. The allelopathic compounds were determined to be relatively low-molecular-weight and heat-stable and the strength of this inhibitory effect was found to vary depending on the target species. Furthermore, the inhibition mechanism was shown to involve a marked suppression of photosynthetic activity in the affected species.