

## Changes in growth rate and photosynthetic properties of mixotrophic haptophyte and cryptophyte species with temperature

\*Shimada Ren<sup>1</sup>, Koji Suzuki<sup>2</sup>

1. Graduate School of Environmental Science Hokkaido University, 2. Faculty of Environmental Earth Science Hokkaido University

Mixotrophs are organisms that retain autotrophic and heterotrophic capabilities. Wilken et al. (2013) proposed the hypothesis that increasing temperature will make mixotrophs more heterotrophic, and several studies support this (e.g., Lin et al., 2018; Chen et al., 2024). However, contrary to the hypothesis, an article indicated that some species enhance autotrophy rather than heterotrophy at higher temperatures (Ferreira et al., 2022). Therefore, the trophic response to temperature change may differ among mixotroph species. On the other hand, temperature-related trophic changes of mixotrophs are little known in polar waters, where the effects of global warming are more sensitive. In this study, we report changes in growth and photosynthetic physiology in response to temperature change using the main mixotrophic haptophyte *Phaeocystis antarctica* and the cryptophyte *Geminigera cryophila*, both of which are dominated in the Southern Ocean.

In this study, *P. antarctica* and *G. cryophila* NBRC 102822 strains were grown in f/2 medium as batch cultures. Photosynthetically active radiation was adjusted to  $30 \mu\text{mol photons m}^{-2} \text{s}^{-1}$  using white LEDs with a 12 h:12 h light:dark cycle. Incubator temperatures were set at  $-1^\circ\text{C}$ ,  $4^\circ\text{C}$ , and  $9^\circ\text{C}$ . Cells were counted by light microscopy using a hemocytometer or Sedgwick Rafter chamber once every two days, and the growth rates were calculated based on the cell concentrations after 14 days of incubation. To estimate the photophysiological status of the mixotrophs, a Satlantic FRe fluorometer was used for evaluating the maximum quantum yield ( $F_v/F_m$ ) of photochemistry in photosystem II during the exponential growth phase. We also measured the light absorption coefficient values ( $a_{ph}^*$ ), and accessory pigment ratios normalized by chlorophyll *a* concentration. In addition, the maximum photosynthetic carbon fixation rate ( $P_{max}^*$ ) was calculated based on the photosynthesis-energy (P-E) curve using the  $^{13}\text{C}$  tracer method.

The haptophyte *P. antarctica* did not grow at  $9^\circ\text{C}$  but grew at  $-1^\circ\text{C}$  and  $4^\circ\text{C}$ , and the growth rates during the exponential growth phase were similar between the two temperatures. On the other hand, the  $F_v/F_m$  values at  $4^\circ\text{C}$  were significantly higher than those at  $-1^\circ\text{C}$ , suggesting that photosynthetic activity in photosystem II changed with temperature. Many carotenoid levels normalized by chlorophyll *a* concentration were significantly higher at  $-1^\circ\text{C}$  than at  $4^\circ\text{C}$ . Similar results were also obtained in the light absorption coefficient values ( $a_{ph}^*$ ) normalized by chlorophyll *a* concentration, suggesting increased accessory pigments at lower temperatures. The growth rates of the cryptophyte *G. cryophila* were highest at  $4^\circ\text{C}$ . The  $F_v/F_m$  values at  $4^\circ\text{C}$  were significantly higher than at  $-1^\circ\text{C}$ , similar to those of *P. antarctica*. To further evaluate changes in autotrophy to rise in temperature, we plan to compare the parameters from P-E curve experiments based on the electron transfer rate.

Keywords: mixotroph, *Phaeocystis antarctica*, *Geminigera cryophila*, the Southern Ocean