Estimation of Primary Production and Nitrogen Mass Balance During *Karenia mikimotoi* Bloom Based on Environmental Variables

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Introduction:
The harmful dinoflagellate Karenia mikimotoi frequently forms red tides in coastal waters worldwide and causes serious damage to fisheries. Because this species is motile and exhibits diel vertical migration, conventional bottle incubation methods are not suitable for accurately estimating its primary production. In addition, continued growth during advection and spatial expansion of blooms is highly probable. Therefore, a simple and spatially applicable method for estimating primary production is required. This study aimed to develop a predictive model of primary production based on environmental variables water temperature, salinity, photosynthetically active radiation PAR, and chlorophyll a concentration Chl a using field observations from the Yatsushiro Sea where red tides occurred. Furthermore, we applied the model to continuous monitoring data to conduct a nitrogen mass balance analysis and evaluate bloom persistence.

Materials and Methods:
Field surveys were conducted in Minamata Bay from March 2021 to July 2022 and throughout the Yatsushiro Sea in August 2024. A 48 hour continuous observation was carried out in Kusura Bay Yatsushiro Sea from 5 to 7 August 2024. Water temperature, salinity, Chl a, and PAR were measured at 1 to 3 hour intervals using an AAQ profiler. Primary production was determined by the 13C method in Minamata Bay n equals 569. In the Yatsushiro Sea survey, primary production estimated by pulse amplitude modulated fluorometry PAM was calibrated against the 13C method n equals 3605. During the continuous observation, nutrient concentrations NH4 N, NOx N, PO4 P, and SiO2 and Karenia mikimotoi cell densities were measured from CTD water samples. Regeneration and consumption fluxes of NH4 N and NOx N in both the water column and sediments were experimentally determined and integrated with standing stocks to evaluate daily nitrogen mass balance. A random forest regression model ranger package R version 4.2.3 was constructed using Chl a, PAR, water temperature, and salinity as explanatory variables and primary production microgram C per L per hour as the response variable.

Results and Discussion:
Primary production measured by PAM showed strong agreement with the 13C method n equals 12 r2 equals 0.853, allowing correction of PAM based estimates. The random forest model demonstrated high predictive accuracy under both bloom and non bloom conditions r2 greater than 0.9. Chl a and PAR were identified as the most important variables. During continuous monitoring, a significant positive correlation was observed between Chl a and Karenia mikimotoi cell density r equals 0.573 p less than 0.0001, indicating that Chl a served as a proxy for cell abundance during the study period.

Nitrogen mass balance analysis showed that the total standing stock of dissolved inorganic nitrogen DIN NOx N plus NH4 N was 8.25 mmol N per m2, while nitrogen based primary production was 3.83 mmol N

per m2 per day. Dividing DIN standing stock by primary production suggested that bloom growth could become nitrogen limited within approximately 2.7 days.

This study established a predictive model capable of quantitatively estimating primary production during red tide events and demonstrated that nitrogen mass balance analysis enables evaluation of bloom persistence. Future work will integrate this primary production model into particle tracking simulations that mimic advection processes to reproduce bloom growth dynamics.

Keywords: Red tide, Yatsushiro Sea, Empirical model, Primary production

