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# Harmful Algae



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Original Article

# Mapping harmful microalgal species by eDNA monitoring: A large-scale survey across the southwestern South China Sea



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### ABSTRACT

A large-scale sampling was undertaken during a research cruise across the South China Sea in August 2016, covering an area of about 100,000 km<sup>2</sup> to investigate the molecular diversity and distributions of microeukaryotic protists, with a focus on the potentially harmful microalgal (HAB) species along the east coast of Peninsular Malaysia. Environmental DNAs from 30 stations were extracted and DNA metabarcoding targeting the V4 and V9 markers in the 18S rDNA was performed. Many protistan molecular units, including previously unreported HAB taxa, were discovered for the first time in the water. Our findings also revealed interesting spatial distribution patterns, with a marked signal of compositional turnover between latitudinal regimes of water masses, where dinophytes and diatom compositions were among the most strongly enhanced at the fronts, leading to distinct niches. Our results further confirmed the widespread distribution of HAB species, such as the toxigenic *Alexandrium tamiyavaichii* and *Pseudo-nitzschia* species, and the fish-killing *Margalefidinium polykrikoides* and *Karlodinium veneficum*. The molecular information obtained from this study provides an updated HAB species inventory and a toolset that could facilitate existing HAB monitoring schemes in the region to better inform management decisions.

#### 1. Introduction

Micro-eukaryotic protists in the seawater ecosystem, including marine phytoplankton, play a critical role in primary production and nutrient cycling that sustains global biogeochemical, ecological, and climatic processes (Abreu et al., 2022). Their vulnerability to environmental change makes them a good indicator of water quality and climate change (Payne, 2013). Likewise, the excessive proliferation of selected harmful protistan species can lead to an ecological phenomenon known as 'harmful algal bloom' (HAB), which affects not only humans but also the ecosystem structure and function (Anderson et al., 2012). Events of HABs have become more prominent in many coastal countries, particularly in Southeast Asia (Yñiquez et al., 2021). This can be attributed to intensified monitoring efforts associated with the expansion of aquaculture production (Hallegraeff et al., 2021). More importantly, the geographical extent has expanded under the influence of climate change and excessive anthropogenic activities (Griffith and Gobler, 2020).

Marine micro-eukaryotic protists are a diverse group of organisms, with over 300 species associated with HAB (Hallegraeff, 2003); more than half of the HAB-associated algal species produce biotoxins (Lundholm et al., 2023) and are responsible for poisoning syndromes in human/marine organisms such as paralytic shellfish poisoning (PSP), diarrhetic shellfish poisoning (DSP), amnesic shellfish poisoning (ASP), azasprasid poisoning (AZP), neurotoxic shellfish poisoning (NSP), and ciguatera poisoning (CP) (e.g., Anderson et al., 2012). Some nontoxic microalgae formed high biomass blooms and caused mortality of fish and marine organisms at multiple trophic levels, by clogging gills or triggering hypoxia/anoxia conditions in the ambient water (e.g., Mohd-Din et al., 2020). This global ecological phenomenon has resulted in remarkable socioeconomic losses due to massive fish kills and fishery closure (e.g., Ritzman et al., 2018; Brown et al., 2020). This included a decrease in revenue for the aquaculture industries and tourism and caused additional costs for monitoring programs and public health services (Griffith and Gobler, 2020).

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