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The prevalence of benthic dinoflagellates associated with ciguatera fish poisoning in the central Red Sea

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ABSTRACT

This study confirms the presence of the toxigenic benthic dinoflagellates *Gambierdiscus belizeanus* and *Ostreopsis* spp. in the central Red Sea. To our knowledge, this is also the first report of these taxa in coastal waters of Saudi Arabia, indicating the potential occurrence of ciguatera fish poisoning (CFP) in that region. During field investigations carried out in 2012 and 2013, a total of 100 *Turbinaria* and *Halimeda* macroalgae samples were collected from coral reefs off the Saudi Arabian coast and examined for the presence of *Gambierdiscus* and *Ostreopsis*, two toxigenic dinoflagellate genera commonly observed in coral reef communities around the world. Both *Gambierdiscus* and *Ostreopsis* spp. were observed at low densities (<200 cells g^{-1} wet weight algae). Cell densities of *Ostreopsis* spp. were significantly higher than *Gambierdiscus* spp. at most of the sampling sites, and abundances of both genera were negatively correlated with seawater salinity. To assess the potential for ciguatoxicity in this region, several *Gambierdiscus* isolates were established in culture and examined for species identity and toxicity. All isolates were morphologically and molecularly identified as *Gambierdiscus belizeanus*. Toxicity analysis of two isolates using the mouse neuroblastoma cell-based assay for ciguatoxins (CTX) confirmed *G. belizeanus* as a CTX producer, with a maximum toxin content of $6.50 \pm 1.14 \times 10^{-5}$ pg P-CTX-1 eq. $cell^{-1}$. Compared to *Gambierdiscus* isolates from other locations, these were low toxicity strains. The low *Gambierdiscus* densities observed along with their comparatively low toxin contents may explain why CFP is unidentified and unreported in this region. Nevertheless, the presence of these potentially toxigenic dinoflagellate species at multiple sites in the central Red Sea warrants future study on their possible effects on marine food webs and human health in this region.

1. Introduction

Globally, marine algal toxins account for greater than 60,000 human intoxications annually, with an overall mortality rate of 1.5% (Van Dolah, 2000). Ninety percent of harmful phytoplankton species are flagellates, notably dinoflagellates (Smayda, 1997), and their biotoxins are responsible for an array of human illnesses, often associated with seafood consumption. The leading non-bacterial illness associated with seafood consumption is ciguatera fish poisoning (CFP) (WHO, 2009).

Certain species and strains of *Gambierdiscus* dinoflagellates produce ciguatoxins (CTX) and gambiertoxins that are precursors to CTXs. CTX is a liphophilic neurotoxin, and the consumption of fish that have accumulated these toxins leads to CFP (see reviews by Anderson and Lobel, 1987; Lehane and Lewis, 2000; Dickey and Plakas, 2010). These ciguatoxin-producing dinoflagellates are macroalgal epiphytes and are thus consumed by herbivorous

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and omnivorous fishes during grazing. Due to their bioaccumulation in the food web, levels of ciguatoxin are highest in carnivorous fish, particularly piscivores. Estimates of CFP incidence are uncertain as the disease is under-reported and often misdiagnosed (Friedman et al., 2008); however, as many as 200,000–1,000,000 people may be affected annually (Fleming et al., 1998; HARRNESS, 2005).

Thus far *Gambierdiscus* is the only dinoflagellate genus that has been definitively linked to ciguatera; however, several other co-occurring tropical and sub-tropical epiphytic species belonging to the genera *Ostreopsis*, *Prorocentrum*, and *Amphidinium* also produce toxins. These genera are well represented in most established benthic dinoflagellate communities in regions such as the Caribbean Sea, Pacific Ocean, and Indian Ocean (ciguatera is present in all these regions (Lehane and Lewis, 2000; Dickey and Plakas, 2010)), but the prevalence or impact of their toxins in the coral reef food chain is not well-documented or understood.

From a biogeographical perspective, *Gambierdiscus* species tend to be found in relatively low densities throughout most tropical and subtropical regions where they occur. Average *Gambierdiscus* spp. cell densities are similar in both the Atlantic and Pacific regions, with a slightly higher frequency of high density samples (i.e., >1000 cells g^{-1} wet weight algae) in the Pacific compared with the Atlantic (Litaker et al., 2010). *Ostreopsis* spp. cell densities vary enormously among geographic regions, although abundances observed in tropical regions are generally lower than in temperate regions. For example, cell densities reported from tropical regions range from <100 to 57,000 cells g^{-1} wet weight algae (Grzebyk et al., 1994; Kohler and Kohler, 1992), whereas in temperate regions cell densities as high as 8.54×10^6 cells g^{-1} wet weight algae have been reported (Cohu et al., 2013). A variety of macroalgae have been reported to host significant numbers of *Gambierdiscus* and other epiphytic dinoflagellates (Cruz-Rivera and Villareal, 2006), although these dinoflagellates are also found to be free living in sediments and coral rubble (Hallegraeff, 1993; Suburova et al., 2013), and in the water column (e.g., Mangialajo et al., 2008).

Despite recent advances in characterizing the global distribution of *Gambierdiscus*, there is limited information regarding the presence of *Gambierdiscus* and the incidence of CFP in the Red Sea. A recent study was the first to record the presence of the genus in the Arabian Gulf and in the northern Red Sea (Suburova et al., 2013), but little else is known regarding the diversity, distribution, and toxicity of *Gambierdiscus* species in the main body of the Red Sea. Similarly, there is scant data regarding toxin levels in fish or the incidence of CFP from the Red Sea region, and none of the few potential cases have been confirmed to arise from Red Sea fish (Ruprecht et al., 2001; de Haro et al., 2003). As ciguatera is frequently unreported or misdiagnosed even in areas where it is well-known, it is possible that the incidence of CFP in this region has been overlooked. Notably, the major species of fish caught by traditional fisheries in the Saudi Arabian Red Sea include grouper, snapper, emperors, barracuda, jacks, trevallies, kingfish, and tuna (Jin et al., 2012), which are known to be ciguatoxic elsewhere in the world. The Red Sea generally suffers from a lack of sustained and intensive ecological research compared with areas such as the Caribbean (Berumen et al., 2013), so CFP or CTX may be present but remains undocumented. The objectives of the present study were to: 1) determine whether ciguatera-associated dinoflagellates (e.g., *Gambierdiscus* spp. and *Ostreopsis* spp.) are present in coral reefs in the central Red Sea, 2) carry out a preliminary assessment of their abundances and distribution at selected sampling sites, and 3) characterize the taxonomy and toxicity of *Gambierdiscus* species present at these sites.

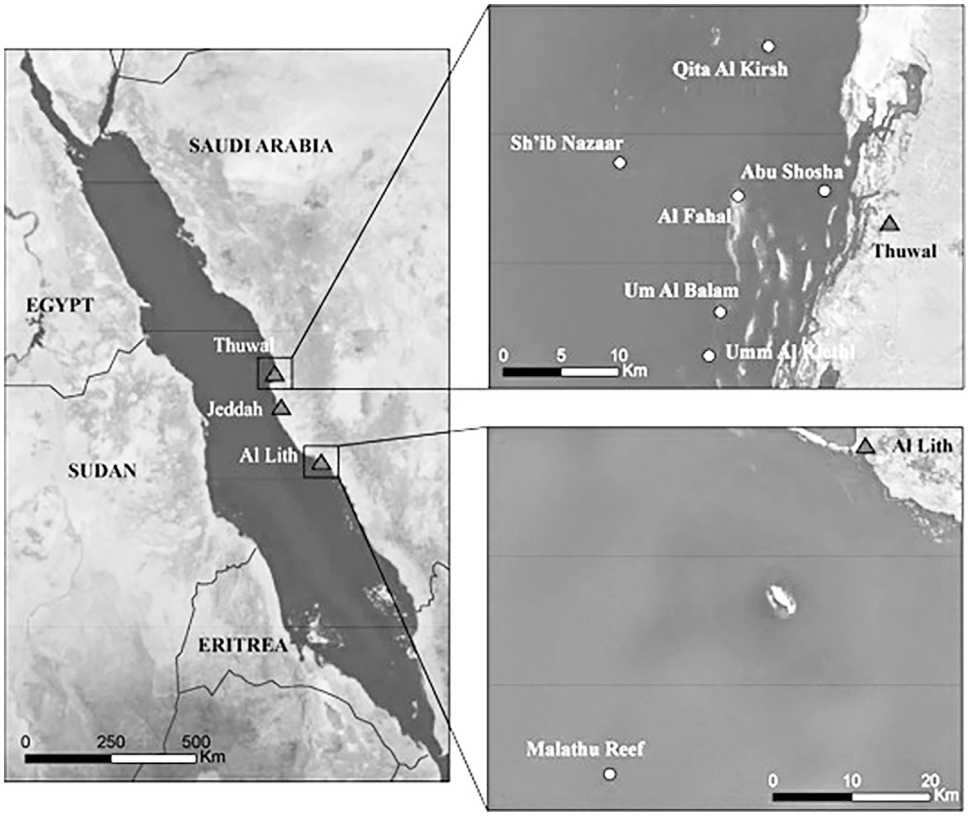

Fig. 1. Sampling sites used in this study included six coral reefs off the coast of Thuwal and four coral reefs off the coast of Al-Lith, Saudi Arabia. Macroalgae collected at these reefs were examined for the presence of ciguatera-associated benthic dinoflagellates.