

## Short Communication

# Autecology of the Toxic *Dinoflagellate Gambierdiscus toxicus* Adachi et Fukuyo (Dinophyceae) in Central Coastal Areas of Tanzania

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**Key words:** *Gambierdiscus toxicus*, Ciguatera, Western Indian Ocean

**Abstract**—The spatial and temporal variability of the toxic dinoflagellate *Gambierdiscus toxicus* in central coastal areas of Tanzania was studied over a period of 14 months between February 2003 and March 2004. *G. toxicus* density on coral rubble algal tufts and seagrasses ranged from 0 to 879.5 cells/gFW and from 0 to 92.6 cells/gFW, respectively. Significantly higher density ( $U = 208.4$ ,  $p << 0.05$ ) was found on coral rubble algal tufts collected near Bawe Island, Zanzibar, compared to coral rubble algal tufts collected near Mbudya Island, Dar es Salaam. However, there was no significant difference in the density of *G. toxicus* collected on coral rubble algal tufts ( $U = 67.6$ ,  $p >> 0.2$ ) between the northern and southern monsoon periods. Also, there were no significant correlations between the biomass of *G. toxicus* with any of the environmental parameters measured in this particular study i.e., water temperature, salinity, nitrate and phosphate concentrations. It is concluded that *G. toxicus* exists in the Tanzanian coastal waters in background concentrations similar to those reported from areas where ciguatera outbreaks has been reported. That algal tufts covering dead corals seems to provide good substrate for developments of *G. toxicus* in the area. A monitoring programme is therefore called for in order to safeguard the health of seafood consumers in the country.

## INTRODUCTION

*Gambierdiscus toxicus* Adachi et. Fukuyo (Dinophyceae), an armored disc or lens shaped dinoflagellate of approximately 80  $\mu\text{m}$  in diameter, was first discovered from the Gambier Islands in French Polynesia (Yasumoto *et al.*, 1977a, b). This was a result of a survey to search for the origin of ciguatoxins that accumulate in fish and cause the seafood poisoning called ciguatera, a form of the most common fish poisoning in tropical marine waters such as the tropical Pacific Ocean, Indian Ocean and the Caribbean Sea (Holmes 1998). Holmes *et al.* (1991) and Satake *et al.* (1993) extracted ciguatoxins from cultured *G. toxicus* confirming the circumstantial evidence of the link

between the dinoflagellate and ciguatera poisoning. Since then, *G. toxicus* has been implicated as one of the principal causative organism of ciguatera fish poisoning. In the Indian Ocean, ciguatera was first described in Mauritius (Dr. JP Quod, personal comm.) and since then it has been repeatedly reported in the Island States of the Western Indian Ocean (Quod and Turquet, 1996).

A qualitative survey of potentially harmful microalgae in coastal waters off Zanzibar was conducted by Kyewalyanga and Lugomela (2001) and identified *G. toxicus* to be among the potentially toxic dinoflagellates that are found along the Tanzanian coastal waters. This implied a potential risk facing seafood consumers along the coast. Indeed seafood intoxication is not

uncommon in Tanzania and human deaths as a result of eating intoxicated seafood have been repeatedly reported through the mass media. However, there has been no any quantitative study that can explain the spatial and temporal distribution of toxic microalgae along the Tanzanian coast. Also, factors regulating their distribution are not known. The current study was therefore aimed to provide baseline information of the spatial and temporal distribution of *G. toxicus* in the central coastal areas of Tanzania.

## MATERIAL AND METHODS

Samples for the *G. toxicus* identification and quantification were collected monthly by SCUBA

diving and/or snorkeling depending on the water depth at the sampling site and by walking in the intertidal areas. Samples were collected from two locations near Dar es Salaam and two locations in the Zanzibar area. The first sampling station was in coral reef area near Mbudya Island, Dar es Salaam, with a maximum depth of about 5 m during high tide. The second station was at Oysterbay, Dar es Salaam, which is an intertidal rocky shore area (Fig. 1). For comparison purposes of coral reef samples, station three was also on a coral reef near Bawe Island, Zanzibar, with a maximum depth of about 10 m during high tide. Station four was in a fishpond at Makoba, Zanzibar (Fig. 2). The coral rubble algal tufts were comprised of essentially brown algae filaments of

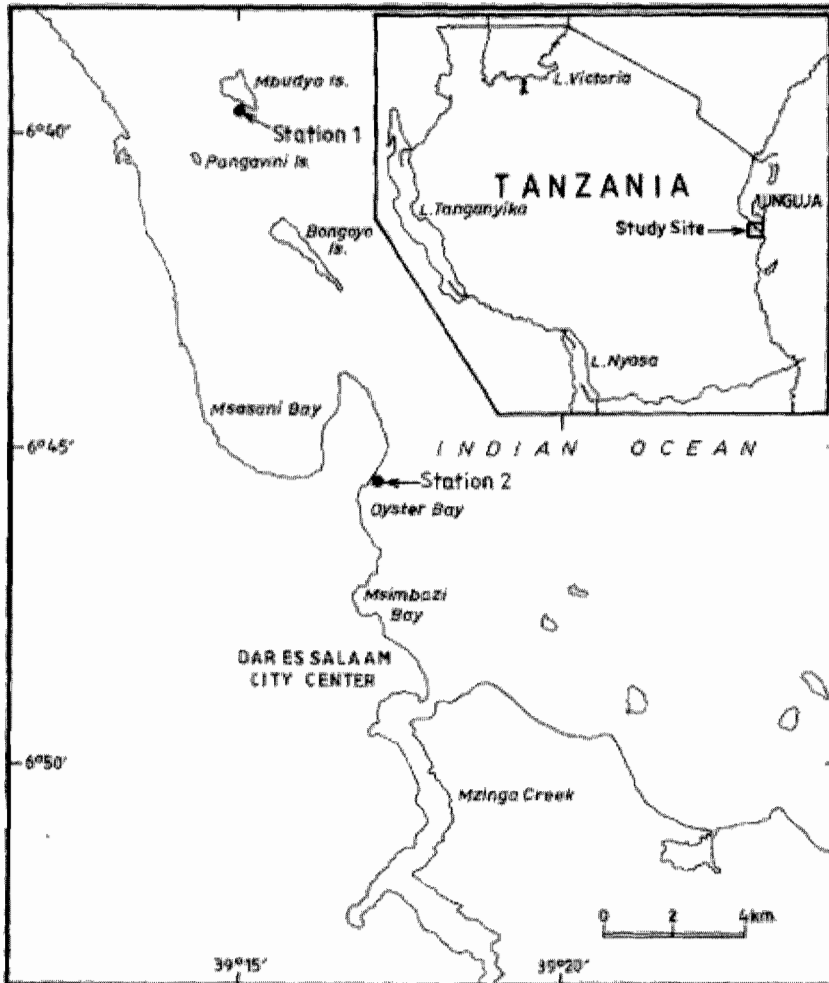


Fig. 1. Map of Dar es Salaam showing the sampling sites (arrows) at Mbudya (Station 1) and Oysterbay (station 2)

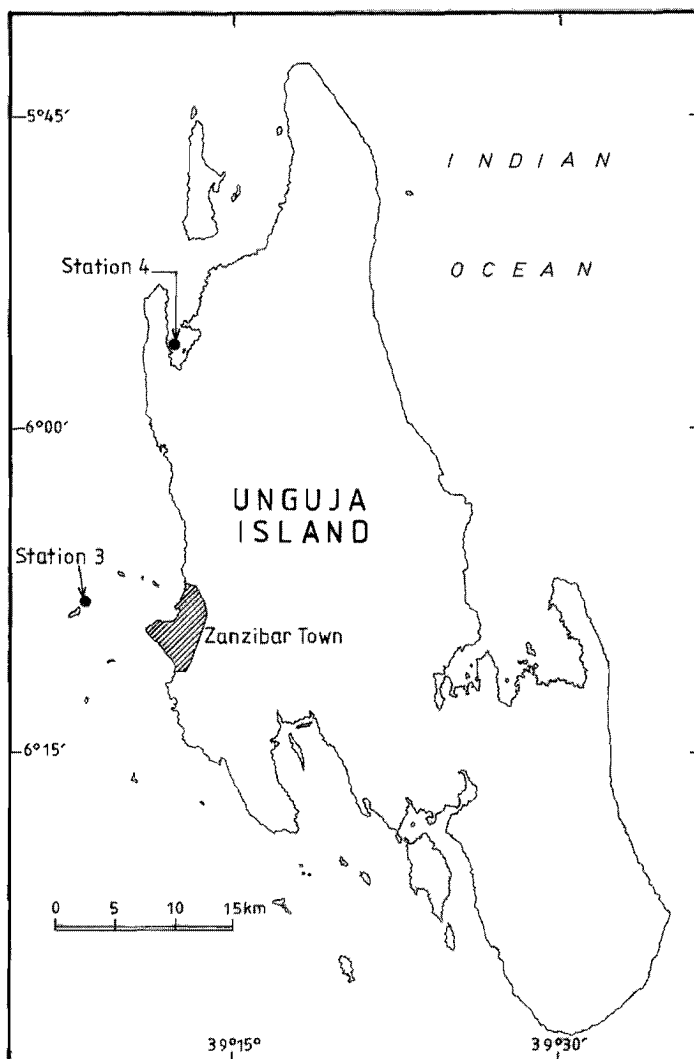


Fig. 2. Map of Zanzibar showing the sampling sites (arrows) at Bawe (Station 3) and Makoba (station 4)

unknown identity mixed with some filamentous cyanobacteria. The seagrass species sampled at Mbudya was *Thalassodendron ciliatum* while the seagrass species sampled at Oysterbay was *Thalassia hemprichii*. In the fish ponds at Makoba samples of *Gracilaria* sp. were collected for isolation of the benthic dinoflagellates.

To avoid unnecessary shaking algal tufts and dead coral pieces were carefully picked by hand and kept in plastic bags while underwater. On the deck the bags were kept in cool boxes containing local seawater to maintain ambient temperature during transport to the laboratory. To detach the

dinoflagellate from the algal tufts the bags were manually shaken vigorously for five minutes while coral rubble was scrubbed using a stiff brush in a bucket half filled with seawater. The suspension resulting from these treatments were filtered through four sets of sieves; 850  $\mu\text{m}$ , 125  $\mu\text{m}$ , 38  $\mu\text{m}$  and 20  $\mu\text{m}$  (Turquet *et al.*, 1998). Dinoflagellates retained in fractions 38  $\mu\text{m}$  and 20  $\mu\text{m}$  were collected in 100 ml filtered seawater and fixed using acid lugol's solution. The samples were then stored in the dark at room temperature before microscopical analyses. Identification and quantification of *G. toxicus* were done in a

Sedgewick Rafter Cell (Woelkerling *et al.*, 1976) under a light microscope. The abundance of *G. toxicus* was expressed per gram of fresh weight algal/seagrass biomass retained in 850  $\mu\text{m}$  and 125  $\mu\text{m}$  sieves.

Water samples were collected from each sampling site for nutrients ( $\text{NO}_3^-$  and  $\text{PO}_4^{+}$ ) determination. The samples were filtered through glass fibre filters into acid cleaned 100 ml plastic vials and kept cool on ice during transport to the laboratory. In the laboratory the samples were kept frozen at  $-20^\circ\text{C}$  before analysis. Analysis of the inorganic nutrients was done using standard methods as described by Parsons *et al.* (1984). Temperature and salinity were measured in the field using a thermometer and refractometer, respectively.

## RESULTS AND DISCUSSION

Water temperature during the study period ranged from  $25.5^\circ\text{C}$  as recorded at Mbudya station in March 2004 to  $32.5^\circ\text{C}$  at Oysterbay in April 2003 (Fig. 3). In general low temperature values were recorded during the southern monsoon period

(May, June, July, August and September) as compared to the northern monsoon period (November, December, January, February, and March). This observation is similar to previous records in the area (e.g. Bryceson 1977). There was no significant correlation between the density of *G. toxicus* collected on coral rubble algal tufts and the water column temperature at both Mbudya and Bawe stations ( $r = -0.118182$ ,  $p = 0.54905$ ). Salinity in the study area ranged from a minimum value of 34 ‰ recorded at Oysterbay during May 2003 to a maximum value of 43 ‰ at Makoba in March 2004 (Fig. 4). In general Makoba station had higher salinity values compared to the other station possibly due to higher evaporation rates in the enclosed fishponds. There was no significant correlation between the density of *G. toxicus* collected on coral rubble algal tufts and salinity of the water at both Mbudya and Bawe stations ( $r = -0.133727$ ,  $p = 0.497512$ ).

Nitrate concentrations in the water column ranged from  $0.49 \mu\text{g at-NO}_3^-/\text{l}$  at Mbudya station in February 2003 to  $7.8 \mu\text{g at-NO}_3^-/\text{l}$  at Oysterbay station during September 2003 (Fig. 5). Higher nitrate concentrations were generally recorded at

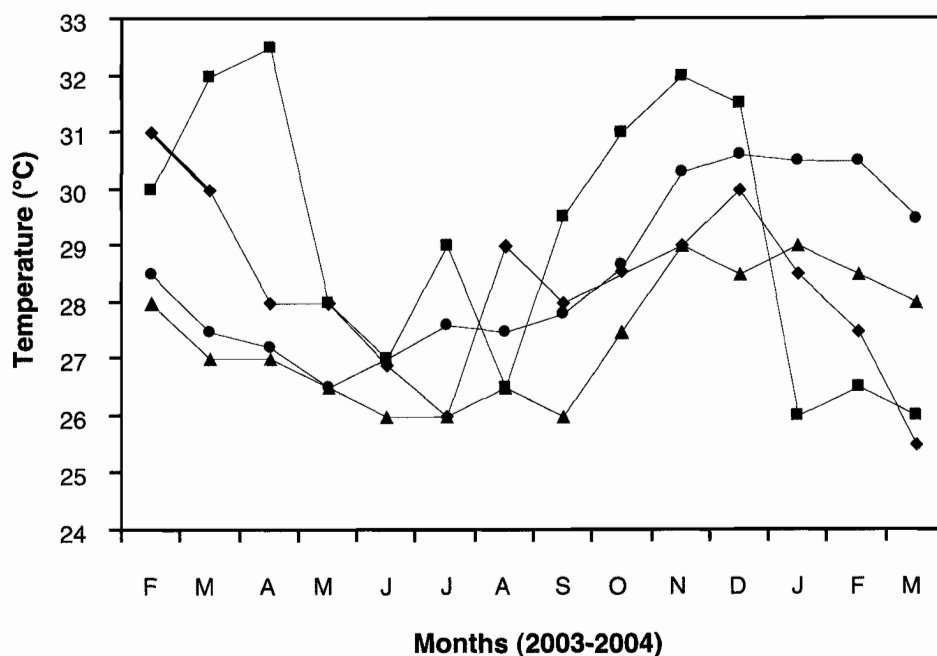


Fig. 3. Water Temperature during the study period at station 1 (diamonds), station 2 (squares), station 3 (triangles) and station 4 (circles)

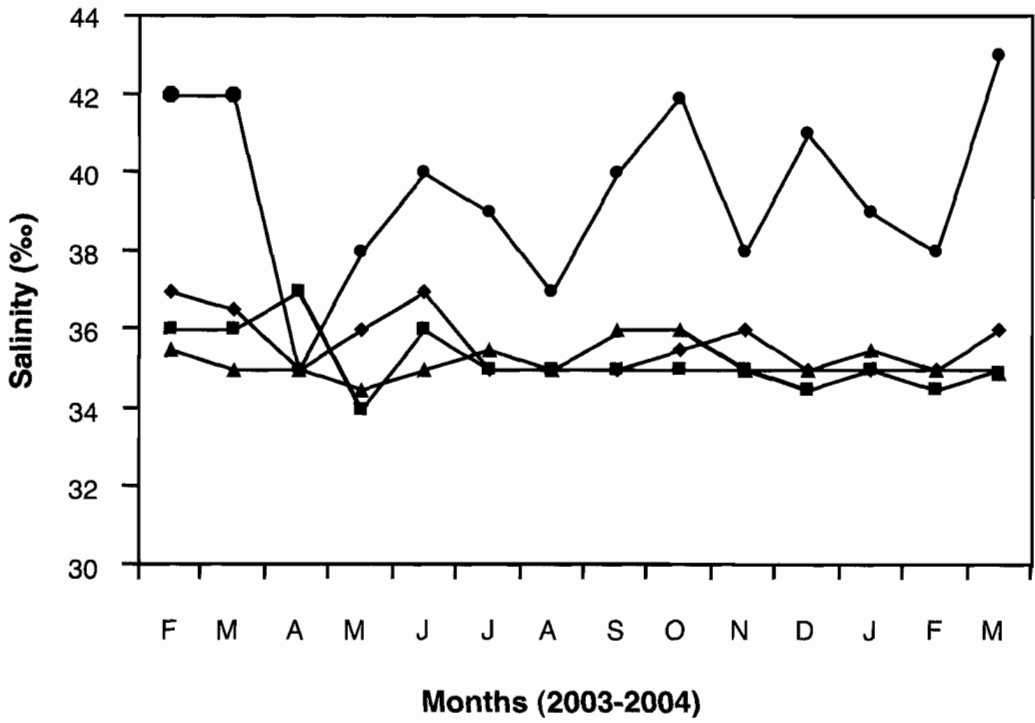


Fig. 4. Salinity in the water column during the period at station 1 (diamonds), station 2 (squares), station 3 (triangles) and station 4 (circles)

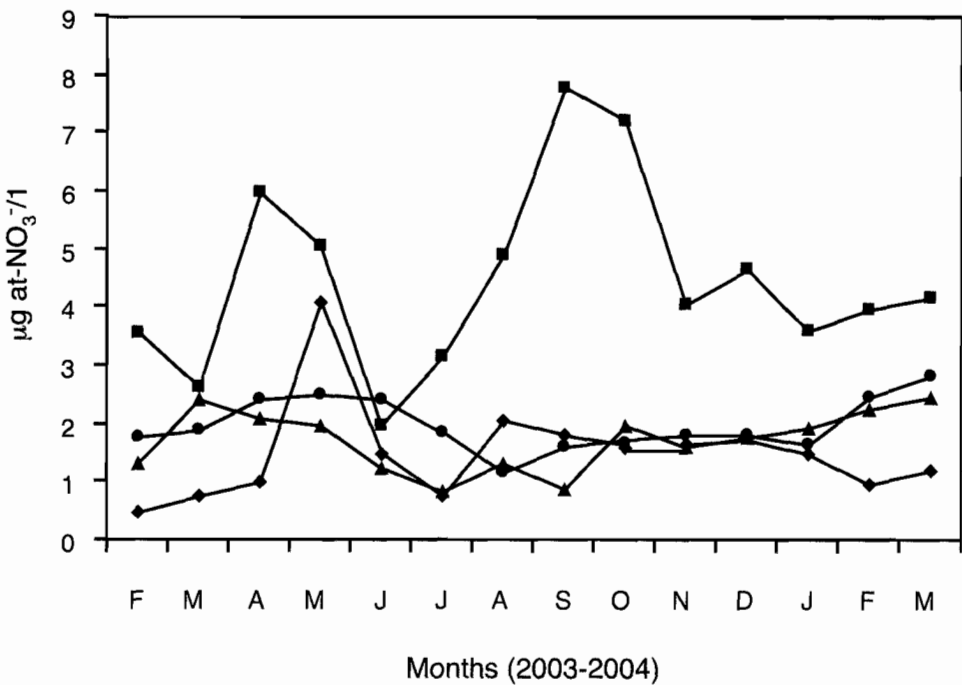


Fig. 5. Concentration of inorganic nitrate ( $\text{NO}_3^-$ ) during the study period at station 1 (diamonds), station 2 (squares), station 3 (triangles) and station 4 (circles)

Oysterbay station possibly due to greater influence of land/terrestrial based water inflow at this intertidal location. However, there was no significant correlations between the density of *G. toxicus* collected on coral rubble algal tufts and the concentration of  $\text{NO}_3^-$  in the water column at both Mbudya and Bawe stations ( $r = 0.031027$ ,  $p = 0.875455$ ). Phosphate concentration ranged from  $0.1 \mu\text{g at-PO}_4^-/\text{l}$  as recorded at Oysterbay during October 2003 to a maximum value of  $1.62 \mu\text{g at-PO}_4^-/\text{l}$  recorded at Bawe station during July 2003 (Fig. 6). Low phosphate concentrations were generally observed during the months of August, September, October and November. Also, there was no significant correlations between the density of *G. toxicus* collected on coral rubble algal tufts and the concentration of  $\text{PO}_4^-$  in the water column at both Mbudya and Bawe stations ( $r = 0.0023164$ ,  $p = 0.439430$ ).

*Gambierdiscus toxicus* was identified by following the general morphological description provided by Fukuyo (1981), Faust (1995), Holmes (1998) and Hansen *et al.* (2001). The collected

specimens were anterior-posterior compressed with cells more or less rounded or slightly ellipsoid. Cell surfaces were smooth with barely visible numerous and more or less evenly distributed pores. The cells had ascending narrow cingulum with a short sulcus. Cell dimensions ranged from  $60 - 150 \mu\text{m}$  in length and from  $45 - 140 \mu\text{m}$  in width.

The density of *G. toxicus* collected on coral rubble algal tufts ranged from zero (in many cases) to  $879.5 \text{ cells/gFW}$  algae that was recorded in August 2003 at Bawe station (Fig. 7). On the seagrasses, the density of *G. toxicus* ranged from zero (in most cases) to  $92.6 \text{ cells/gFW}$  seagrass as recorded in January 2004 at Mbudya. Note that *G. toxicus* was never found on the seagrass collected from intertidal pool at Oysterbay (Fig. 8). The values obtained in this study are generally high compared to those reported by Holmes *et al.* (1998) on Singapore reefs but comparable to those reported by Turquet *et al.* (1998) off Reunion Island, SW Indian Ocean, and Heil *et al.* (1998) off Heron Island, Australia. Turquet *et al.* (2001)

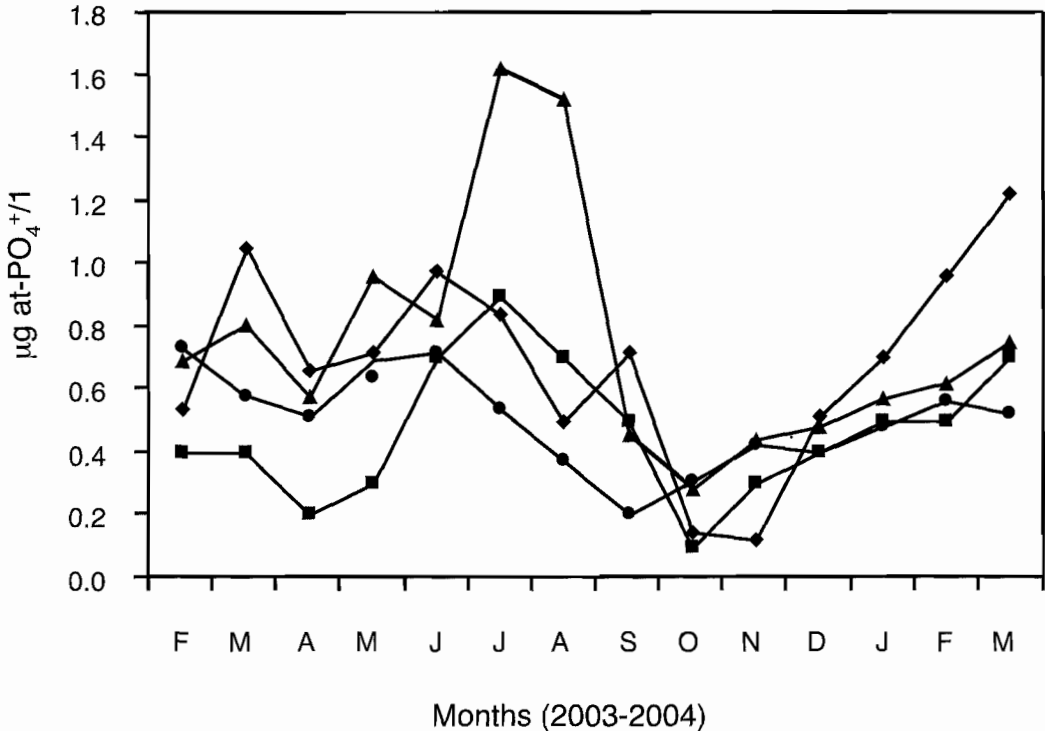


Fig. 6. Concentration of inorganic phosphate ( $\text{PO}_4^-$ ) during the study period at station 1 (diamonds), station 2 (squares), station 3 (triangles) and station 4 (circles)

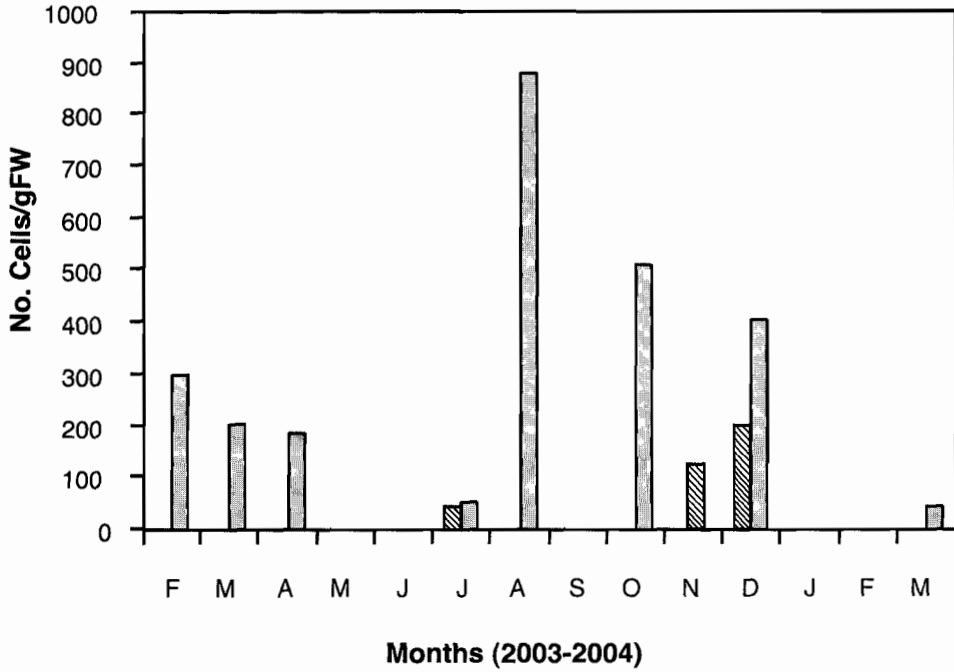


Fig. 7. Biomass of *G. toxicus* on coral rubble algal tufts during the study period at Station 1 (striped bars) and station 3 (dotted bars)

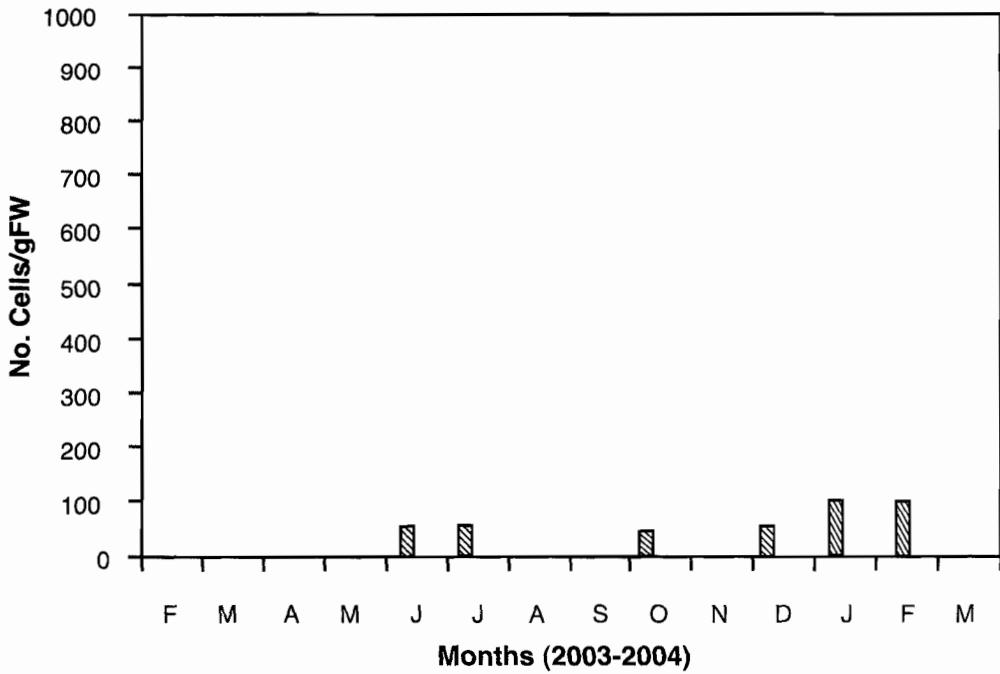


Fig. 8. Biomass of *G. toxicus* on seagrasses during the study period at Station 1 (striped bars) and station 2 (dotted bars)

recorded an unusually high density of *G. toxicus* reaching cell concentration of 60,463 cells per gram of algae during October 1998 in Mayotte Island, Comoros. The authors associated this to coral bleaching during that year, which resulted in proliferation of multispecific algal turfs on bleached corals, a good condition for epiphytic microalgae such as *Gambierdiscus*, *Ostreopsis* or *Prorocentrum* species.

Significantly higher density ( $U = 208.4$ ,  $p < 0.05$ ) of *G. toxicus* was found on coral rubble algal turfs collected near Bawe Island as compared to those found on coral rubble algal turfs collected near Mbudya Island. The reason to this variation is not clear and calls for further study. The lack of *G. toxicus* on seagrass collected from the intertidal areas at Oysterbay suggests that the microalgae in question does not grow in the intertidal flats possibly due to the high fluctuation of environmental parameters such light intensity, temperature and desiccation, conditions that usually prevail in such an ecosystem. *Gambierdiscus* was also not observed on the macrophytes collected from Makoba bay fishponds. It is assumed that no seed populations were brought into the fishponds as a result of tidal inundation since this alga primarily lives a benthic life. It is also possible that environmental parameter, not quite apparent from this study did not allow for the growth of the algae in the ponds. This implies that the fishpond may be free from *Gambierdiscus* induced ciguatera.

There was no clear trend in the temporal variability of the *G. toxicus* density in the study area (Fig. 7 and 8). Indeed, there were no significant difference ( $U = 67.6$ ,  $p >> 0.2$ ) between the northern monsoon period (November, December, January, February and March) and the southern monsoon period (May, June, July, August and September). However, a high fluctuation in *G. toxicus* density between different sampling dates was apparent (Fig. 7 and 8). This implies a high spatial-temporal fluctuation (patchiness) in the distribution of this microalga. This observation is similar to that reported elsewhere for benthic microalgal species (e.g. Pinckney *et al.*, 1994).

## CONCLUSIONS

From this study it is possible to say that *G. toxicus* exists in the Tanzanian coastal waters in background concentrations similar to those reported from areas where ciguatera outbreaks has been reported. That algal tufts covering dead corals seems to provide good substrate for developments of *G. toxicus* in the area. Monitoring programme for *G. toxicus* biomass and ciguatoxins in seafood from coral reef areas need to be implemented so as to safeguard the health of seafood consumers in the country. The lack of significant correlation between the biomass of *G. toxicus* with any of the environmental parameters that were measured in this study suggests that other variables not tested in this particular study may be more responsible for regulating the observed spatial and temporal variability. Thus long-term studies in order to establish good trends and separate from short term variations of some parameters are required so as to establish the cause of population dynamics of this important dinoflagellate.

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## REFERENCES

- Bryceson, I. (1977) An ecological study of phytoplankton of the coastal waters of Dar es Salaam. PhD thesis, University of Dar es Salaam, 165 pp.
- Faust, M.A. (1995) Observation of Sand-dwelling toxic dinoflagellates (Dinophyceae) from widely differing sites, including two new species. *J. Phycol.* **31**: 996-1003.



- Fukuyo, Y. (1981) Taxonomical study on benthic dinoflagellates collected in coral reefs. *Bull. Jpn. Soc. Sci. Fish.* **47(8)**: 967-978.
- Hansen, G., Turquet, J., Quod, J.P., Ten-Hage, L., Lugomela, C., Kyewalyanga, M., Hurbungs, M., Wawiye, P., Ogongo, B., Tunje, S. & Rakotoarinjanahary, H. (2001) Potentially harmful microalgae of the Western Indian Ocean - a guide based on a preliminary survey. IOC Manuals and Guides No. 41, Intergovernmental Oceanographic Commission of UNESCO, 105 pp.
- Heil, C.A., Bird, P. & Dennison, W.C. (1998) Macroalgal habitat preference of ciguatera dinoflagellates at Heron Island, a coral cay in the southeastern great barrier reef, Australia. In: Reguera, B., Blanco, J., Fernández, M.L. & Wyatt, T. (eds.) Harmful Algae. Xunta de Galicia and Intergovernmental Oceanographic Commission of UNESCO, pp. 52 - 53.
- Holmes, M.J. (1998) *Gambierdiscus yasumotoi* sp. nov. (Dinophyceae), a toxic benthic dinoflagellate from southeastern Asia. *J. Phycol.*, **34**: 661- 668.
- Holmes, M.J., Lee, F.C., Teo, S.L.M. & Khoo, H.W. (1998) A survey of benthic dinoflagellates on Singapore reefs. In: Reguera, B., Blanco, J., Fernández, M.L. & Wyatt, T. (eds.) Harmful Algae. Xunta de Galicia and Intergovernmental Oceanographic Commission of UNESCO, pp. 50 - 51.
- Holmes, M.J., Lewis, R.J., Poli, M.A. & Gillespie, N.C. (1991) Strain dependent production of ciguatoxin precursors (gambiertoins) by *Gambierdiscus toxicus* (Dinophyceae) in culture. *Toxicon*, **29**: 761-775.
- Kyewalyanga, M. & Lugomela, C. (2001) Existence of potentially harmful microalgae in coastal waters around Zanzibar: a need for monitoring programme? In: Richmond MD and Francis J (eds.) Marine science development in Tanzania and Eastern Africa. Proceedings of the anniversary conference on advances in marine science in Tanzania. 28<sup>th</sup> June - 1<sup>st</sup> July 1999, Zanzibar, Tanzania. IMS/WIOMSA, pp. 319 - 328.
- Parsons, T.R., Maita, Y. & Lalli, M. (1984) A manual of chemical and biological methods for seawater analysis. Pergamon press, 173 pp.
- Pinckney, J., Piceno, Y. & Lovell, C.R. (1994) Short term changes in the vertical distribution of benthic microalgal biomass in intertidal muddy sediments. *Diat. Res.* **9**: 143 - 153.
- Quod, J.P. and Turquet, J. (1996) Ciguatera in Reunion Island (SW Indian Ocean): epidemiology and clinical patterns. *Toxicon*, **34**: 779-785.
- Satake, M., Ishimaru, T., Legrand, A.M. & Yasumoto, T. (1993) Isolation of a ciguatoxin analog from cultures of *Gambierdiscus toxicus*. In: Smayda, T.J. & Shimizu, Y. (eds.) Toxic phytoplankton blooms in the sea. Elsevier, Amsterdam, pp. 575 - 579.
- Turquet, J., Quod, J.P., Couté, A. & Faust, M.A. (1998) Assemblage of benthic dinoflagellates and monitoring of harmful species in Reunion Island, SW Indian Ocean, 1993-1996. In: Reguera, B., Blanco, J., Fernández, M.L. & Wyatt, T. (eds.) Harmful Algae. Xunta de Galicia and Intergovernmental Oceanographic Commission of UNESCO, pp. 44 - 47.
- Turquet, J., Quod, J.P., Ten-Hage, L., Dahalan, Y. & Wendling, B. (2001) Example of a *Gambierdiscus toxicus* flare-up following the 1998 coral bleaching event in Mayote Island (Comoros, South-West Indian Ocean). In: Hallegraeff, G.M., Blackburn, S.I., Bolch, C.J. & Lewis, R.J. (eds.) Algal Blooms 2000: Proceedings of the Ninth International Conference on Harmful Algal Blooms, pp. 50 - 53.
- Yasumoto, T., Bagnis, R., Thevenini, S. & Garçon, M. (1977a) A survey of comparative toxicity in the food chain of ciguatera. *Bull. Jpn. Soc. Sci. Fish.* **43**: 1015-9.
- Yasumoto, T., Nakajima, I., Bagnis, R. & Adachi, R. (1977b) Finding of a dinoflagellate as a likely culprit of ciguatera. *Bull. Jpn. Soc. Sci. Fish.* **43**: 1021-6.
- Woelkerling, W.J., Kowal, R.R. & Gough, S.B. (1976) Sedgewick-Rafter cell counts: a procedural analysis. *Hydrobiologia*, **48**: 95-107.