toxin) so that the resulting hydrophilic metabolites can be eliminated into urine. The oxidation of CTX-4B to CTX could thus be regarded as a kind of detoxification process.

What actually happens to CTX-4B is the opposite of detoxification; the toxicity of the oxidised product (CTX) is in fact enhanced ninefold.

Therefore, we can say that moray eels are more toxic than parrotfish, both because the former are at a higher trophic level and because they accumulate toxins in the most toxic forms. If someone finds the enzymes which catalyse the oxidation, such enzymes will not only help us understand the metabolic fate of toxins, but will also help chemists run reactions which they cannot run with reagents.

Recently we have determined the structure of maitotoxin (MTX), the biggest natural product ever to be elucidated. MTX is nearly three times bigger than CTX, having a molecular formula $C_{164}H_{256}O_{68}S_2Na_2$ and a molecular weight of 3422 Da.

It consists of a C142 carbon chain, 32 ether rings, 28 hydroxyls, and 21 methyls. Analogous with CTX, most of the ether rings in MTX are fused in a ladder shape. Nevertheless, the two toxins are entirely different molecular entities. MTX does not contain CTX as a part of its structure.

Therefore, it is quite clear that there will be no possibility of converting MTX to CTX by feeding MTX to fish or bacteria, as speculated earlier by some people.

Also in our recent work, one *G. toxicus* strain was confirmed as producing CTX analogues in cultures. Structures of CTX-3C and CTX-4A in the cultures were unambiguously confirmed by spectroscopic measurements. This result puts to an end the long-running argument about whether *G. toxicus* is the true source of ciguatera toxins or not. The strain (RAI1 strain) was one of six tested.

If renewed effort were made to collect and screen more strains, there would be chances of finding other strains producing CTX analogues, hopefully in even higher yields than our RAI1 strain. As future progress on ciguatera studies depends on an adequate supply of toxins, and as the current supply from fish is very limited, the prospect of obtaining toxins by algal cultures is very encouraging.

We can even dream that some day we will be getting CTX by oxidising CTX precursors produced by the alga with liver enzymes. We would like to urge biologists to collect and test as many *G. toxicus* strains as possible for production of valuable toxins.

Evaluation of Hawaiian reef fishes with the solid-phase immunobead assay (SPIA)

by Y. Hokama,
Department of Pathology,
University of Hawaii, Honolulu

This study was published in the *Journal of Clinical Laboratory Analysis* in 1993. It presents data on the evaluation of a laboratory-made ciguatera testing system based on the solid-phase immunobead assay (SPIA) for the detection of ciguatoxin and related polyethers in Hawaiian reef fishes. The SPIA was performed on fish caught by volunteer fishermen throughout the State of Hawaii.

A total of 1,067 fish representing 61 different species was tested by the SPIA system, as reported in the *Journal of Clinical Laboratory Analysis* in 1990. Of the 1,067 fishes tested, 510 were from the island of Oahu, 402 from Hawaii (Big Island), and 75 from Maui. Other fish included 23 from Molokai, 20 from Kauai and 7 from Lanai. Twenty per cent of the total fish tested were positives, 41 per cent borderlines and 39 per cent negatives in the SPIA assay. The highest percentages of SPIA-positive fish were from the island of Hawaii (27%), followed by Oahu (19%) and Kauai (15%).

These results correlate with the incidents reported from the State Department of Health of actual ciguatera fish poisoning in the State of Hawaii.

Unfortunately fish in all categories were eaten, though warnings strongly emphasised that all borderline and positive SPIA-tested fish were *not* to be eaten. All 332 negative fish eaten (80% of 416 fish) caused no poisoning, therefore *no false negatives*.

However, of the 201 borderline SPIA value fish eaten (46% of 433 fish), 4 caused ciguatera poisoning symptoms. These fish included 2 **papio** (*Caranx* sp.), 1 mullet (*Mugilcephalus*), and 1 **po'ou** (*Cheilinus rhodochrous*). Finally of the 17 SPIA-positive-tested fish eaten (8% of 218 fish), 5 caused ciguatera poisoning. This involved 2 **papio** (*Caranx* sp.), a **kole** (*Ctenochaetus strigosus*), an **uhu** (*Scarus*) and a **weke** (*Mulloidichythys auriflamma*).

The SPIA test used by the fishermen was successful in protecting the public when SPIA-negative fish eaten caused no illness, that is, there were *no false negatives*.

We have contended that a person who is genetically more susceptible, or has had long-term exposure to reef fish consumption in endemic regions, will be most likely to become ill from eating SPIA-borderline or positive fishes. Indeed, this appeared to be the case.

The data suggested that the probability of getting ill with SPIA-positive fish is 1 out of 3; with the borderline fish, 1 out of 50. As indicated, if the fish is negative by SPIA the possibility of ciguatera is nil. The *Caranx* spp. (**papio** or **ulua**) appeared to be the major culprits causing ciguatera. This is compatible with the Department of Health reports for ciguatera in Hawaii.

References

Hokama, Y. 1990. Simplified solid-phase immunobead assay for detection of ciguatoxin and related polyethers. J. Clin. Lab. Anal. 4:213–217.

Hokama, Y., A.Y. Asahina, E.S. Shang, T.W.P. Hong & J.L.R. Shirai. Evaluation of the Hawaiian reef fishes with the solid-phase immunobead assay. J. Clin. Lab. Anal. 7:26_30.

Hokama, Y., A.Y. Asahina, E. Titus, J.L.R. Shirai, T.W.P. Hong, S. Chun, J.T. Miyahara, D. Takata, A. Muranaka, E. Pang, I.A. Abbott & D. Ichinotsubo. In press. A survey of ciguatera: assessment of Puako, Hawaii, associated with ciguatera toxin epidemics in humans. J. Clin. Lab. Anal.

Ciguatera fish poisoning: the situation in New Caledonia

by Philippe Amade, CR1, INSERM, La Darse, Villefranche/mer, France

Situation

The coral reef surrounding the islands of New Caledonia has a particularly rich biological diversity with numerous fish species.

However, visiting the fish market in Noumea, one is surprised that relatively few reef species of fishes are sold, compared with sales of deep-slope and pelagic fishes. Ciguatera fish poisoning is believed to be responsible for this situation.

An investigation performed in March 1992 in Noumea on a representative sample of 500 people, indicated that 124 of them (nearly 25%) had been intoxicated at least once (Laurent et al.1993). This percentage varied according to the ethnic groups: Polynesians 44 per cent, Asians 34 per cent, Europeans 24 per cent, Melanesians 23 per cent and Wallisians 18 per cent.

According to the April 1989 census, the population of the city, excluding children under 10 years old, was 79,167 . It is therefore possible to estimate that 20,000 persons were affected by this intoxication. This result suggests that the current estimates from the South Pacific Commission (based on figures reported by health authorities in New Caledonia) may be significantly below the real incidence of ciguatera intoxications in New Caledonia, mainly because a large number of cases only concern weak intoxications, not declared to doctors or hospitals, and frequently cured by traditional medicine(s).

A widespread phenomenon

Ciguatera poisoning is widespread in the outer islands of New Caledonia. According to local reputation rather then scientific analysis, the north of the mainland has non-toxic fishes as compared with the south. A similar comparison can be made between the island of Ouvea and the other Loyalty Islands. Some places are reputed always to harbour toxic fishes.