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Ciguatera fish toxicity in French Polynesia: Size does not always matter

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ABSTRACT

Accumulation of ciguatoxins (CTXs) in tropical reef fish tissues during their life is responsible of the most prevalent human seafood intoxication in the South Pacific called Ciguatera Fish Poisoning (CFP). It has been assumed for a long time that CTXs are transferred and accumulated along the trophic food chain, and consequently that smaller individuals within a given fish species are safer to eat than larger ones. However, the relationship between toxicity and fish size has been studied for a limited number of species only and the conclusions are often contradictory. The toxicity of 856 fishes from 59 different species sampled in six islands in French Polynesia between 2003 and 2011 was assessed by Receptor Binding Assay. Among them, 45 species × island and 32 families × island for which the number of individuals was ≥6 allowed testing the relationship between toxicity and size. Except for six specimens of *Lutjanus bohar* caught in Fakarava ($P < 0.01$; $R^2 = 0.854$), the 44 remaining species × island showed no significant increase of CTXs concentration with fish total length (TL). Moreover, the proportion of toxic individuals decreased significantly for *Epinephelus polyphekadion* from Fakarava ($n = 24$; $P < 0.05$) and *Kyphosus cinerascens* from Raivavae ($n = 29$; $P < 0.05$), while no significant variation was detected for the other 43 species × island. At the family level, only three positive and three negative relationships between size and CTXs concentration were observed among the 32 family × island analyzed. No relationship between the proportion of toxic fish within a family and the relative total length of individuals were observed. The lack of relationship between toxicity and size observed for most of the species and families from the six islands suggests that fish size cannot be used as an efficient predictor of fish toxicity in French Polynesia. These results highlight the need for improving our knowledge about metabolic processes which may play a role in CTXs bio-accumulation and depuration among the different trophic levels of fishes.

1. Introduction

Ciguatera is a food poisoning caused by consumption of tropical reef fishes which affects between 10,000 and 50,000 people worldwide each year (Friedman et al., 2008;

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Lewis, 2001). People affected by Ciguatera Fish Poisoning (CFP) generally suffer from gastrointestinal, neurological and cardiovascular symptoms but also from hallucinations and paralysis (Oh et al., 2012). Paresthesia of the extremities, tingling around the mouth, metallic taste and cold allodynia (dysesthesia when touching cold water or objects) are also characteristic to CFP (Vetter et al., 2012). Currently, only aqueous extract of *Heliotropium foertherianum*, and especially its major active compound rosmarinic acid, has been identified as an inhibitor of cytotoxicity induced by the Pacific ciguatoxin P-CTX-1B (Rossi et al., 2012). The whole CFP symptoms result from persistent activation of the voltage-gated sodium channel (Nicholson and Lewis, 2006) caused by absorption of even small concentrations of liposoluble toxins called ciguatoxins (CTX). The odorless, colorless and heat stable chemical properties of CTX have always hampered the resolution of CFP issue in the concerned areas worldwide.

Unicellular din flagellate algae from the genus *Gambierdiscus* reported as the main responsible of CFP (Bagnis et al., 1980) have been identified as the primary source of CTX (Adachi and Fukuyo, 1979). There are also evidences of the presence of CTX-like compounds in marine cyanobacteria *Hydrocoleum* spp., causing food poisonings by consumption of giant clams (Laurent et al., 2008), or in sea urchins (Pawlowiez et al., 2013), but these toxins have not yet been involved in fish toxicity. Herbivorous fishes grazing, or browsing on, macro-algae hosting the toxic dinoflagellates remain the major route of CTX entrance into the marine food web. Several studies on *Gambierdiscus* ecology have provided models which can be used to predict their population dynamics and estimate the lag time between peak cell densities and the first occurrences of CFP cases (Chateau-Degat et al., 2005; Parsons et al., 2010). The growth of dinoflagellates, which influences toxin production, depends on environmental factors such as temperature, salinity and irradiance (Chinain et al., 2010a; Kibler et al., 2012). Nonetheless, it has been proved that toxin production is also linked to *Gambierdiscus* assemblages species composition (Chinain et al., 2010a). As a consequence, the amount of CTX bioavailable and potentially transmitted to herbivorous fishes is not directly related to single-cell algae abundances and biomasses.

As CTX presence do not change the aspect or smell of the fishes, different strategies have been developed by consumers in order to distinguish safe and toxic individuals. One of these strategies is based on the assumption that smaller individuals within a given fish species are safer to eat that larger ones. This assumption comes from the food chain hypothesis first stated by Randall (1958) and reinforced by Lewis and Holmes (1993). In short, CTX concentration in fish tissues depends primarily on dietary intake, efficiency of assimilation, depuration rate and fish growth. However, even though changes in liver protein metabolism of toxic groupers and moray eels have been documented (Jiang et al., 2012), CTX depuration process has never been clearly demonstrated in fish. Thus if depuration does not occur, or occurs at very a low rate, larger individuals within a given species should be the ones with the highest CTX concentrations (Lehane and Lewis, 2000). Surprisingly, studies about the relationship between toxicity and fish size, or weight, are available only for a limited number of species and the results they present are often contradictory. For instance, O'Toole et al. (2012) demonstrated that CTX concentrations in the liver, blood and muscle of *Sphyraena barracuda* in the Bahamas are not correlated with fish length. Similar observations have also been reported for the peacock grouper (*Cephalopholis argus*) in Hawaii (Dierking and Campora, 2009; Bienfang et al., 2012) and for various species of *Seriola* spp. and *Acanthocybium solandri* in the Canary Islands (Caillaud et al., 2012). On the contrary, Chan et al. (2011) observed a positive relationship between CTX concentration in muscles and liver of moray eels (*Gymnothorax* spp.) from Kiribati Islands and the weight of the individuals. Such observations suggest that CTX concentration in fish tissues may be the result of biological and physiological processes more complex than the merely positive relationship with the size or weight of individuals currently postulated by several authors (Oshiro et al., 2010; Clua et al., 2011).

The main goal of this study was to test if CFP toxicity increased with fish size for several fish species from different families obtained from different islands of French Polynesia. Firstly, we tested whether CTX concentration significantly increased with fish total length (TL), i.e. if larger individuals were more toxic than smaller ones. Secondly, we tested whether the proportion of toxic individuals increased with TL. In other words whether, for a given taxa, it was possible to detect a size above which the probability for a fish to be toxic was significantly higher. These two questions were addressed both at the species and the family levels in order to circumvent the problem of uncertain identifications that is often observed for tropical fish species, and to provide practical suggestions useful for local populations.

2. Materials & methods

2.1. Sampling and study locations

From 2003 to 2011, several eco-toxicological field monitoring surveys were conducted by the Louis Malardé Institute (ILM) of Tahiti in six distinct CFP-prone islands of French Polynesia, namely Nuku Hiva (Marquesas Archipelago), Fakarava (Tuamotu Archipelago) and Rurutu, Tubuai, Raivavae and Rapa (Australes Archipelago) (Fig. 1). Fishes were sampled by spear-fishing and species known as presenting a high potential risk of CFP were preferentially targeted. Each individual was identified at the species level using Bacchet et al. (2006), with their total length (TL) and weight measured to the nearest centimeter and gram, respectively. Fillets of flesh were stored at −30 °C until toxicological analysis which were carried out a few weeks after sampling.

2.2. CTXs extraction and quantification

CTXs were extracted from 5 g flesh portions purified on Sep-Pak C18 + cartridges (Watters®) using different proportions of aqueous methanol (Darius et al., 2007). Then, fish toxicity was measured using the Receptor Binding