Ciguatera Poisoning in Antigua and Barbuda: Working Towards a Risk Management Strategy

Envenenamiento por peces Ciguatera en Antigua y Barbuda: trabajando hacia una estrategia de gestión de riesgos

Intoxication par les poissons de la ciguatera à Antigua-et-Barbuda: Vers une stratégie de gestion des risques

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EXTENDED ABSTRACT

Introduction

Ciguatera Poisoning (CP) is a foodborne illness caused by consumption of fish contaminated with ciguatoxins (CTX). Whilst rarely fatal CP causes gastrointestinal distress, and severe neurological symptoms (among many others) in those acutely exposed, with up to 500,000 people estimated to be affected worldwide annually. Ciguatoxins are produced by tropical marine microalgae of the genus *Gambierdiscus* spp. which once ingested by marine herbivores can become bioaccumulated through the food web, and impact upon important fisheries species such as hogfish, snapper, grouper, and mackerel. Globally, CP is distributed broadly across all subtropical and tropical oceans with some variability observed in toxin profiles between regions. CTXs are large polycyclic, polyether compounds, and within the Caribbean, two main chemical isomers have been most commonly reported known as C-CTX-1 and -2, and many other related toxins identified but yet to be elucidated (Robertson et al, 2014).

Within the Caribbean, CP is well documented with regions around many islands considered hyperendemic, and as such can have profound effects on fisheries with associated socio-economic burdens in remote coastal communities. It is known that CP occurrence can be highly localised in its prevalence, both geographically and within fish species. For instance, fish with high site fidelity (e.g., groupers; parrotfish) may be consistently toxic in a specific region, however many migratory fish (e.g., amberjack, mackerel) also forage in reef areas, but may then move to new areas and cause illness following harvest. Much of the species and regional variability has been accumulated through local knowledge from recreational, artisanal, and commercial fishers. Investigations that have worked with local fisheries managers and fishers have greatly improved available risk assessments but more data across and within regions is needed to fully establish safe harvest zones, seasonality, and marine species of highest risk. A recent study examining CTX prevalence in the US Virgin Islands that was conducted in collaboration with local fishermen, found that while only 4% of the pre-market fish (i.e., fish known to be caught and sold for consumption) had levels above the USFDA guidance limit of 0.1 μ g/Kg, there was significant variation in toxicity between species and region (E, W, SE and SW of St. Thomas), which may be linked to oceanic wave energy (Loeffler et al, 2018). Similarly, a study examining CTX levels in lionfish (*Pterois volitans*) collected from the US Virgin Islands found that 12% of fish examined exceeded FDA limits with clear regional differences (Robertson et al 2014).

We are currently working with the Antigua Fisheries Board to assess occurrence and levels of CTXs within edible fish tissues from Antigua and Barbuda. A number of fish species from sites believed to be high, medium and low risk will be assessed for the presence of CTX and toxicity levels determined to see if there is any correlation with current local knowledge. This partnership demonstrates how the scientific community can collaborate with local fishers and resource managers to improve evidence-based CP management strategies.

Methodologies

A range of fish species covering multiple trophic levels, including commercially important species such as red hind (*Epinephelus guttatus*); and a variety of snapper species (*Lutjanus* spp.), hogfish, and parrotfish will be compared to commonly avoided apex predators such as barracuda (*Sphyraena barracuda*), with samples collected by commercial fishers from a range of sites within Antigua and Barbuda. These sites will comprise of locally-suspected high, medium and low CTX risk areas. Once collected, morphometric data will be collected (fork and total length, weight, sex) otoliths retrieved for age assessment, and tissues harvested and sent to our respective laboratories for further assessment. Samples will be

chemically extracted for CTX and toxicity assessed via two methods as described in Turner et al; (manuscript in submission). Total toxicity will be assessed using a neuroblastoma N2a cytotoxicity assay, with molecular confirmation of CTX presence performed using liquid chromatography coupled with tandem mass spectrometry (LC-MS/MS). Relationships between CTX prevalence in fish will be assessed based on location, species and morphometric characteristics.

Results and Discussion

Initial pilot data from a proof-of-concept study conducted in Spring 2019 is promising with mass spectrometric data confirming the molecular identification of CTX in a number of predatory fish species from Antigua. As shown in Figure 1 below, LC-MS/MS chromatograms monitoring the triply dehydrated pseudo-molecular ion of m/z 1087.6 > 1087.6 reveals the presence of two distinct peaks with retention times of 2.64 and 2.73 mins, consistent with Caribbean CTX-1 and -2 in bar jack, snapper and barracuda extracts. This supports the hypothesis that the two CTX analogues are both prevalent in Antiguan waters and also in a range of fish species. At this time, it cannot be discounted that other congeners that have not yet been elucidated or detected in this study may be present even in the fish with lower levels of CTX-1 and 2, so composite toxicity testing will be essential to evaluate risk. In support of this work additional bulk ciguatoxic barracuda material was collected and is currently being extracted and purified in an attempt to generate a Caribbean CTX reference materials as part of a larger collective effort.

The data presented here show for the first time the molecular confirmation of the presence of Caribbean CTXs in a range of fish species from Antigua. Further investigations into toxicity levels between species, trophic levels and geographical sites will provide invaluable knowledge into the extent of CTX prevalence within the region. Due to the lack of commercially available analytical standards for C-CTXs, the generation of Caribbean CTX reference material will be an invaluable asset to ongoing CTX studies



Figure 1. UHPLC-MS/MS chromatograms of tissue extracts from a number of fish species harvested from Antiguan waters.

in the Caribbean region. In addition to providing a reference to enable a quantitative approach, it will also allow a thorough assessment of potential new C-CTX isomers through use of other mass spectrometric and cell toxicity approaches.

This study highlights the applicability and validity of a science-based approach to CTX risk mapping within the Antiguan region which has the potential to be used throughout the Caribbean. Such an approach has enormous potential for augmenting local fisheries management and in facilitating a reduction in the incidence of CP in these regions.

LITERATURE CITED

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