

# Mass mortality of marine invertebrates associated with the presence of yessotoxins in northern Chile

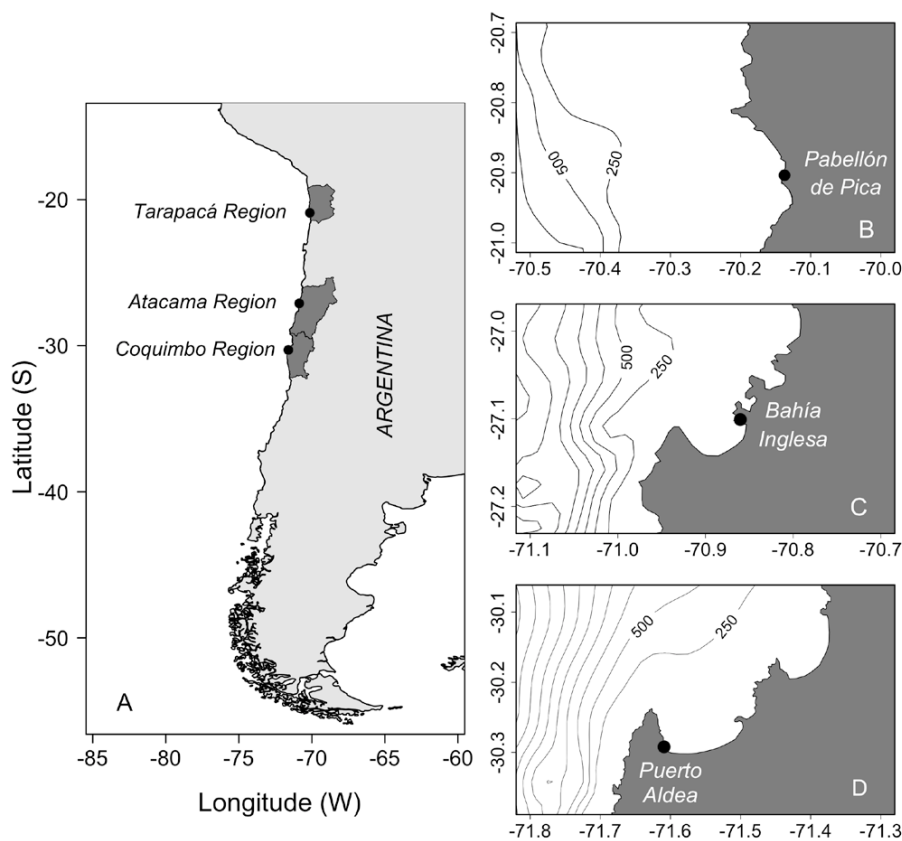


Fig. 1. Map of the study area showing A) Chilean coast; B) Pabellón de Pica, Tarapacá Region; C) Bahía Inglesa, Atacama Region; D) Puerto Aldea, Coquimbo Region

During the austral summer of 2019, mass mortalities of marine invertebrates were detected at different locations throughout an extensive geographic area (up to 1000 km coastline) along the northern Chilean coast (Fig. 1). On January 24<sup>th</sup>, massive strandings of starfish (*Stichaster striatus*), red sea urchins (*Loxechinus albus*), and clams (*Ameghinomya antiqua*) were detected in Pabellón de Pica, Tarapacá Region (Fig. 2A, B). Unfortunately, accurate in-

formation about the total stranded biomass of each species was not obtained. The estimated density of sea urchins was between 15 and 20 individuals m<sup>2</sup>. Toxin analyses of digestive tissue samples of the stranded specimens carried out by Liquid Chromatography- High Resolution Mass Spectrometry (LC-HRMS) revealed the presence of yessotoxins (YTX) with concentrations ranging between 0.1 and 0.4 mg YTX kg<sup>-1</sup> (Table 1).

Two weeks later, on February 10-11<sup>th</sup>, a mass mortality of Humboldt squid (*Dosidicus gigas*) was detected in Bahía Inglesa, Atacama Region (Fig. 2C, D). During this episode, thousands of dying or dead squid were washed ashore or appeared floating near the shore of one of the most visited beaches in northern Chile. At the end of this event, the density of beached squid on the sand was estimated to be between 45 and 50 individual m<sup>-2</sup> (approximately 15 tons). The health risk posed by the contaminated dead animals led the Ministry of Health to carry out a social media campaign to prevent their consumption. In addition, the City Council organized the beaches to be cleaned, to mitigate the negative impact on tourism caused by the decaying animals. LC-HRMS analyses of pooled samples of the viscera from individuals collected on February 11<sup>th</sup> revealed the presence of YTX with a concentration of 0.42 mg YTX kg<sup>-1</sup>.

By the end of summer, on March 31<sup>th</sup>, a new mass mortality event of squid *D. gigas* was detected at the artisanal fishing harbor of Puerto Aldea, Coquimbo (Fig. 2E, F). During this event which took place during the night and early morning, thousands of individuals were found dead or dying, mainly lying on the surface of harbor facilities and, to a lesser extent, on nearby beaches. A total of 130 tons of stranded biomass was quickly collected and sold by fishermen to fish processing plants and the local market. However, in order to guarantee seafood safety the health authorities demanded an analysis of the squid in all of the fishing plants involved and withdrawal of the squid being sold in the local market. LC-HRMS analyses of the viscera from pooled samples revealed the presence of YTX with a concentration of 0.12 mg YTX kg<sup>-1</sup>.

Table 1. Concentration of yessotoxin detected in different marine invertebrate species during the mass mortality events detected along the northern Chilean coast.

Date	Species	Location	Type of sample	mg YTX kg <sup>-1</sup>
01/24/19	<i>Stichaster striatus</i>	Pabellón de Pica	Digestive tissues	0.10
01/24/19	<i>Loxechinus albus</i>	Pabellón de Pica	Digestive tissues	0.11
01/24/19	<i>Ameghinomya antiqua</i>	Pabellón de Pica	Digestive gland	0.45
02/11/19	<i>Dosidicus gigas</i>	Bahía Inglesa	Visceral mass	0.42
31/03/19	<i>Dosidicus gigas</i>	Puerto Aldea	Visceral mass	0.12



Fig. 2. A-B) Stranded specimens of the red sea urchin *Loxechinus albus* on the beach at Pabellón de Pica, Tarapacá Region; C) Specimens of the Humboldt squid *Dosidicus gigas* on the beach at Bahía Inglesa, Atacama Region; D) Control of the Chilean army to avoid commercialization of stranded organisms; E-F) Squid *Dosidicus gigas* on the beach at Puerto Aldea, Coquimbo Region.

The relationship between YTX and mass mortality events of invertebrates has been reported in other geographical areas around the world. In 2011, a mortality event associated with the presence of the dinoflagellate *Gonyaulax spinifera* and low levels of YTX (<0.1 mg kg<sup>-1</sup>) was reported in Sonoma County, California affecting different marine invertebrates such as the red sea urchin (*Strongylocentrotus franciscanus*), the purple sea urchin (*S. purpuratus*), the starfish (*Pisaster ochraceus*) and the abalone (*Haliotis rufescens*) [1-2]. Recently, in 2017 Pitcher et al [3] reported a mass mortality of 250 tons of the abalone (*H. midae*) in different aquaculture farms along the South African

coast associated with a bloom of *Gonyaulax spinifera*. During this episode, the toxin profile in the digestive gland was dominated by homo-YTX, 45-hydroxy-YTX, and a minor contribution of YTX, with average concentrations of 0.73; 0.21 and 0.09 mg kg<sup>-1</sup>, respectively. The gill tissues, with an average concentration of 1.1 mg kg<sup>-1</sup> (homo-YTX), 0.33 mg kg<sup>-1</sup> (45-hydroxy-YTX) and 0.11 mg kg<sup>-1</sup> (YTX), was the most contaminated organ. It is worth noting that the YTX concentrations found during these episodes were very similar to those reported from Northern Chile. These coincidences suggest that yessotoxins may have been the main cause of marine invertebrate mortalities. More

research is needed to determine the mechanism of action and the toxin effects on tissues and cells of the main affected species. Finally, there is a need to establish an educational plan to protect the public and avoid the consumption and commercialization of potentially toxic marine invertebrates.

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