# Monitoring and management of paralytic shellfish toxins in Southern Rock Lobster, Tasmania, Australia

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## Abstract

The Tasmanian Southern Rock Lobster (*Jasus edwardsii*) fishery has been challenged by recurrent dinoflagellate blooms of *Alexandrium catenella* since 2012. The initial bloom resulted in the first ever closure of an Australian lobster fishery due to marine biotoxins and exposed several key knowledge gaps for managing food safety and market access risk. To fill these gaps, experimental studies were conducted to determine paralytic shellfish toxin (PST) toxicokinetics. Adult male lobsters fed highly toxic mussels (6 mg STX.2HCl eq $kg^{-1}$) accumulated PST in the hepatopancreas at an exponential rate of 6% per day, reaching a maximum level of 9 mg STX.2HCl eq $kg^{-1}$ in three weeks. However, lobsters exposed to toxic algae culture suspensions at 2 x $10^5$ cells $L^{-1}$ did not accumulate any toxin. Neither accumulation of PST nor exposure to toxic cells resulted in any gross impact on the health of lobsters, as assessed by a comprehensive range of behavioural, immune, nutritional and biochemical indicators. Field studies over a period of eight years confirmed the ability of lobster hepatopancreas to rapidly accumulate and depurate toxins in the wild, with a high degree of variability. Analysis of 496 hepatopancreas samples collected during *A. catenella* blooms identified high risk sites and seasons; demonstrated the usefulness of mussels as sentinel species for indicating PST risk; and enabled quantification of the confidence level associated with current risk management sampling practices. The combined experimental and field results have led to improved risk management for this AUD 97M wild fishery.

*Keywords:* PST, toxin, lobster, hepatopancreas, non-traditional vector, toxicokinetics, lobster health, *Alexandrium*, risk management

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## Introduction

An extensive dinoflagellate bloom of *Alexandrium catenella* occurred on the east coast of Tasmania in 2012, causing the first ever Australian lobster fishery harvest closure due to marine biotoxins (Campbell *et al*., 2013). During the bloom, Southern Rock Lobster, *Jasus edwarsdii*, accumulated PST to 3.9 mg STX.2HCl equiv $kg^{-1}$. Since 2012, recurrent blooms of *A. catenella* have occurred during winter and spring months when water temperatures are between 10 and 15°C and coastal waters may stratify (Condie *et al*., 2019). These blooms have had an on-going impact on both the commercial lobster fishery in Tasmania, valued at AUD 97 M, and the significant recreational fishing sector. In order to better manage the associated public health and market access risks, a series of experimental and field studies was undertaken. Initial work focused on the risks to human health from PST accumulation in Southern Rock Lobster, looking at the concentration of PST in the hepatopancreas, assessing the fate of PST during cooking, and consumer exposure levels (Madigan *et al*., 2018a,b; McLeod *et al*., 2018; Turnbull *et al*., 2018).

Risk management of PST in lobster in Tasmania has adopted the bivalve PST maximum level (ML) as the regulatory level (DPIPWE, 2020), however, lobster sampling strategies are necessarily different from those for molluscs due to the geographical spread of the wild fishery, the different way the animals are consumed, and the high level of variability among individual animals. Lobsters are keystone marine species, so concern was also raised over potential impacts on lobster health.

Further experimental and field studies were undertaken, seeking knowledge of the toxicokinetics of PST accumulation and depuration; of supply chain risks of exposure to toxic algal cells; impacts on lobster health; and effective methods to monitor PST levels in the field.

## Material and Methods

*Experimental studies*

Two controlled experiments took place in a biosecure facility using adult male lobsters housed in individual tanks in a flow-through aquaculture system (Fig. 1). In the first experiment, lobsters were fed mussels containing 6 mg STX.2HCl equiv $kg^{-1}$ for 27 days then moved to a non-toxic diet for a further 36 days (Turnbull *et al*., 2020a). PST in the hepatopancreas was examined at regular intervals during uptake and depuration using LC-MS/MS (Boundy *et al*., 2015; Turner *et al*., 2015). Exponential uptake and depuration rates were calculated and changes in the toxin profile noted. At the peak of uptake, PST was also analysed in the hindgut, antennal glands, gills and haemolymph.

In the second experiment, lobsters were exposed to toxic cultures of *A. catenella* at field relevant concentrations of 2 x $10^5$ cells $L^{-1}$ for three weeks (Turnbull *et al*., 2021b), replicating potential exposure in the supply chain. PST was measured in the hepatopancreas at three time points.

Fig. 1. Biosecure experimental facility where lobster housed in individual tanks were exposed to PST through either food or toxic algal cultures.

During both experiments, lobster health was assessed at each harvest point via a comprehensive set of behavioural, immune, nutritional and biochemical responses, measured by the same operator in the same order on each harvest day (Turnbull *et al*., 2020a). Histological analysis of the toxic algae exposed lobsters was conducted using formalin fixed paraffin embedded gill tissues samples cut into three micron thickness and stained with haemotoxylin and eosin.

*Field studies*

Lobster sampling (n = 496) occurred on a regular basis from 2012 to 2020 in eight lobster biotoxin management zones on the east coast of Tasmania during *A. catenella* blooms (Turnbull *et al*., 2021a; Fig. 2).

On each sample occasion, lobster hepatopancreas from each site were analysed individually for PST (n = 5 animals). Blue mussels, *Mytilus galloprovincialis,* from adjacent aquaculture farms or specifically installed mussel lines were sampled over the same time period as potential sentinel species. Samples were analysed for PST using either HPLC-FLD (Lawrence *et al*., 2005) or LC-MS/MS (Boundy *et al*., 2015; Turner *et al*., 2015).

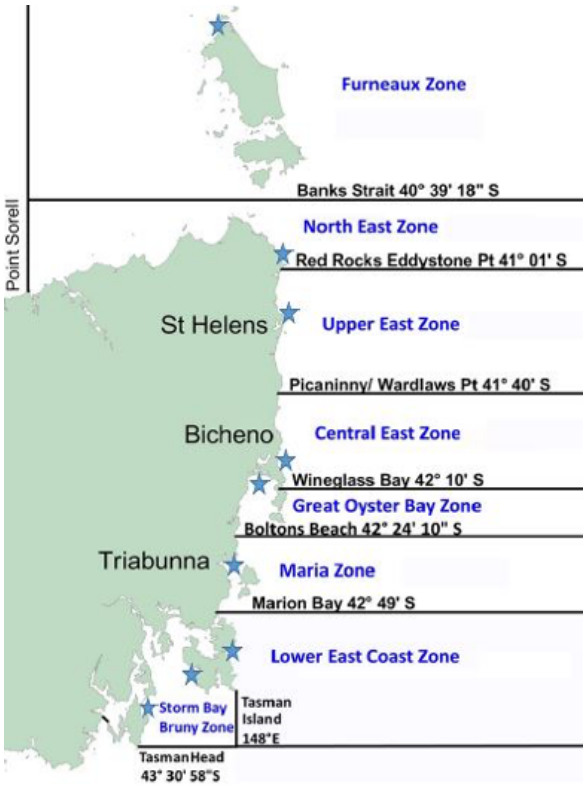


Fig. 2. PST lobster management zones on the east coast of Tasmania, Australia. Sentinel mussel sample locations are indicated with an asterisk. Source: DPIPWE, 2020.

Uptake and depuration rates for lobster were calculated for events with four or more consecutive sampling occasions at the same site and compared to concurrent mussel uptake and depuration rates. Data from the start and end of blooms were examined to determine the number of lobsters required to give a 95% confidence that the population is below the bivalve maximum level.

## Results and Discussion

When lobsters were fed toxic mussels, PST accumulated rapidly in the hepatopancreas at an exponential rate of 6% per day, reaching

a mean of 6.7 mg STX.2HCl eq $kg^{-1}$ after 27 days (Turnbull *et al*., 2020a). The majority of toxins in the hepatopancreas during uptake were GTX2,3 C1,2, and GTX1,4, with the proportion of the latter decreasing as uptake continued (Fig. 3). In comparison the mussel feed contained mostly GTX1,4 and GTX 2,3. The lobsters depurated at a rate of 7% per day once toxic feed was removed. PST was detected in lobster antennal glands and gills (possible excretion routes for PST), however, it was not detected at significant levels in lobster haemolymph. The majority of toxin in the antennal glands and gills were GTX2,3 and dcGTX2,3.

Exposure to PST did not result in mortality nor significant changes in any of the behavioural, health, nutritional and haemolymph biochemical parameters measures suggesting limited gross impact on lobster performance and indicating adult lobster are relatively tolerant to PST (Turnbull *et* al., 2020b).

Lobsters exposed to highly toxic algal cultures of *A. catenella* did not accumulate PST and no negative impact on lobster health or gill tissue was observed (Turnbull *et al*., 2021b). The results indicate that PST uptake can only occur through the consumption of toxic prey and hence that there are no food safety or quality risks from exposure to toxic cells during wet storage in boat wells, sea cages or specialized wet storage facilities in the supply chain.

Field studies and regulatory monitoring over the eight year period showed high variability in toxin levels between individuals, sites, months and years. The central Tasmanian coast was identified as the greatest risk site, but confined to the months of July to January

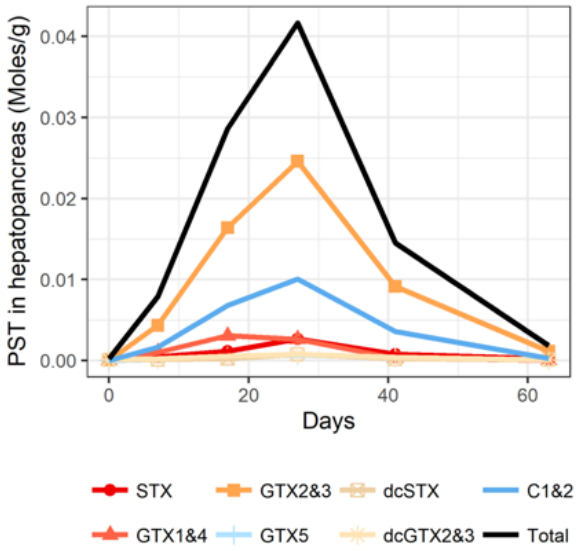


Fig. 3. PST analogue mean molar content in hepatopancreas of exposed *J. edwardsii* during 27 days of uptake and 35 days of depuration.

(Turnbull *et al*., 2021b; Fig. 4). Relatively high PST uptake rates were observed in lobster hepatopancreas (exponential rate of 2% per day), similar to but consistently less than rates seen in filter-feeding mussels. Lobsters were relatively fast detoxifiers following bloom demise, losing up to 3% PST per day. Mussel sentinel lines were a cost-effective means of indicating PST risk in lobsters, with an annual baseline monitoring cost of <0.1% of the industry value. The current practice of analysing multiple lobster from a site and closing on a conservative trigger level provides a 97.5% confidence level that any lobster from that site would be below the bivalve maximum level of 0.8 mg STX eq $kg^{-1}$..

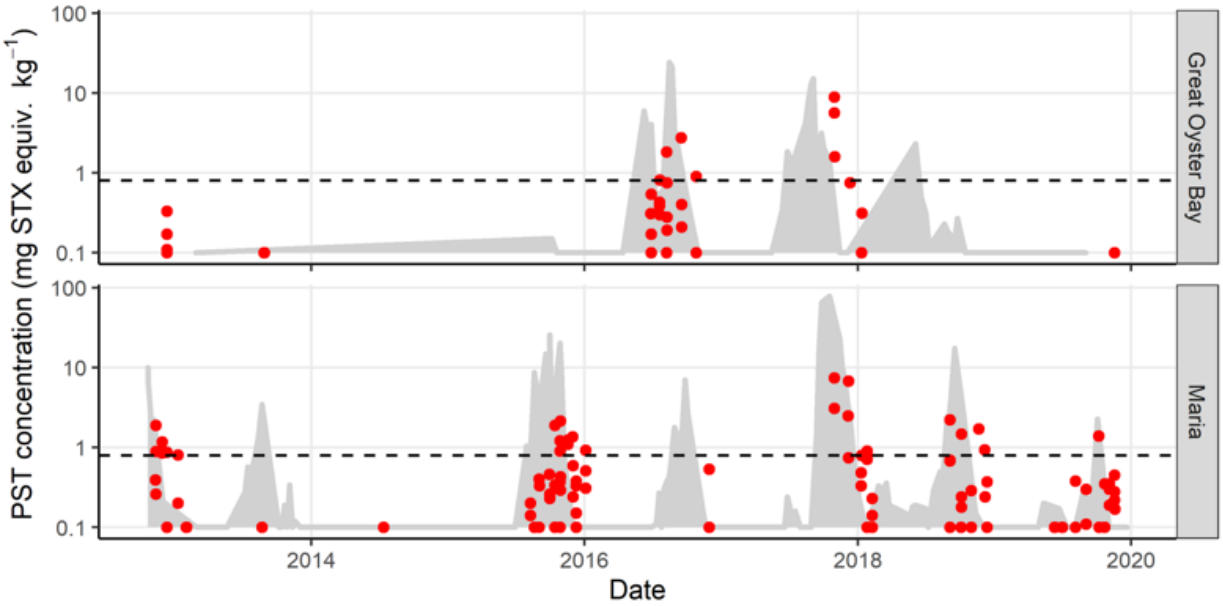


Fig. 4. PST levels in lobster hepatopancreas (red dots) and mussels (grey areas) from the central east coast of Tasmania from 2012 to 2020 inclusive. Modified after Turnbull *et al*. (2021).

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