Table 1Highest toxin levels reported for echinoderms and tunicates.

Species	Toxins	Toxin concentration	Analysed tissue	Local	Reference
Echinoderms					
Suspension-feeder					
Echinoidea					
Dendraster excentricus	ASP	15 mg DA kg ⁻¹	Whole body	California (CA), USA	(Kvitek et al., 2008)
Carnivorous					
Asteroidea					
Asterins amurensis	PSP	$^{\mathrm{a}}$ 1440 μg STX eq. kg $^{\mathrm{-1}}$	Whole body	Hiroshima, Japan	(Asakawa et al., 1997)
Asterina pectinifera	PSP	$^{\mathrm{a}}$ 2250 μg STX eq. kg $^{\mathrm{-1}}$	Whole body	Hiroshima, Japan	(Ito et al., 2003)
Astropecten scoparius	PSP	nr	Nr	Pingtung, Taiwan	(Lin et al., 1998)
Tunicates					
Suspension-feeder					
Ascideacea					
Pyura chilensis	ASP	16 mg DA kg ⁻¹	Edible part	South of Chile	(López-Rivera et al., 2009)
Microcosmus vulgaris	PSP	807 µg STX eq. kg^{-1}	Whole body	Adriatic sea, Croatia	(Roje-Busatto & Ujević, 2014)
Halocynthia roretzi	PSP	$^{\mathrm{a}}$ 3420 µg STX eq. kg $^{-1}$	Mantle	Iwate, Japan	(Nagashima et al., 1984)
	PSP	$^{\rm a}$ 252000 µg STX eq. kg $^{-1}$	Digestive Gland	Iwate, Japan	(Nagashima et al., 1984)

nr: not reported.

in late 2007, the rapid alert system on foodstuffs and feed (RASFF) of the EU Commission emitted a sanitary alert claiming that *Murex* spp. samples from Portugal were exceeding the legal limits for PSP toxins and were the cause of a human poisoning incident in Spain that lead a person into coma. Toxicity as high as 1510 μ g STX kg⁻¹ were determined by Spanish competent authorities using the EU reference method for PSP toxins determination, which at the time was a biological testing method that exposes mice through an intraperitoneal injection of an acidic shellfish extract. However, the gastropod species was misidentified, it was not *Murex* spp. but the trumpet shell *Charonia lampas lampas* and subsequent chemical analysis via liquid chromatography with tandem mass spectrometry revealed that the trumpet shell was highly contaminated not with PSP toxins but with tetrodotoxin (TTX) in the visceral mass (Rodriguez et al., 2008).

The possibility for the presence of TTX has long been recognized in the Japanese *Charonia sauliae* (Noguchi, Narita, Maruyama, & Hashimoto, 1982). The source of toxins in carnivorous gastropods has been difficult to trace. For example, in the previous case, the authors have confirmed its presence in the starfish *Astropecten polycanthus*, a common prey of this trumpet shell. Many of the older scientific reports resorted to the mouse bioassay as the only testing method. It is possible that in several instances STX analogues were mistaken with TTX analogues. Or even a mixture of both could have occurred in the samples analysed. TTX and its analogues are not regulated in the EU legislation. However, fish from family Tetraodontidae, Molidae, Diodontidae and Canthigasteridae must not be placed on markets since they harbor or are suspected to harbor TTX (EC, 2004c).

4. Relevance of crustaceans as toxins vectors

Crustaceans are not included in EU directives for marine toxins monitoring (EC, 2004c), but the number of documented studies and reported cases of toxins accumulated in crustaceans (Table 3) is higher than the number of studies reporting the presence of toxins in echinoderms and tunicates.

Several human poisoning incidents due to the consumption of contaminated crustaceans have been reported worldwide, including European waters from Portugal to Norway (Torgersen, Aasen, & Aune, 2005; Vale & Sampayo, 2002). While PSP toxins have been the toxins most commonly associated with crustaceans throughout the world, when it regards to Europe, the cases of toxin accumulation were related with DSP toxins. In 2001, a diarrhetic

poisoning episode in Portugal, was associated with consumption of contaminated crabs (*Carcinus maenas*) containing 322 μ g OA eq. kg⁻¹ of edible tissue (Vale & Sampayo, 2002). In Norway, several hundred people presented DSP symptoms after eating brown crabs *Cancer pagurus* (Torgersen et al., 2005). Similarly to the Portuguese episode, the toxin profile of the contaminated crabs was substantially (>90%) composed by esterified derivatives of the OA that are collectively designated as DTX3.

Although not associated with any human poisoning event in Europe, DA has been detected in crabs (Carcinus maenas, Polybius henslowii) reaching concentrations up to 323 mg DA kg⁻¹ whole body (Costa, Rodrigues, Botelho, & Sampayo, 2003; Vale & Sampayo, 2002). DA is a water-soluble molecule that is generally rapidly eliminated from shellfish, however, DA was found in most of the swimming crab samples obtained along the Portuguese coast during 2002, even when DA was not detected in bivalve mollusks neither toxigenic diatoms were observed in seawater (Costa et al., 2003). While there is no regulation for DA in crustaceans caught and commercialized in Europe, a specific limit for Dungeness crabs, higher than that for bivalve mollusks, has been established in the USA. Dungeness crab is an important fisheries resource in California and Washington State of the USA that commonly accumulate high levels of DA. In order to prevent acute intoxications US authorities concluded that Dungeness crabs must not be consumed when 30 mg DA kg⁻¹ is detected in crabs viscera (Mariën, 1996).

Preventive actions against consumption of contaminated crustaceans in the USA are extensive to PSP toxins contamination. Although there is not a regulatory limit for PSP toxins in crustaceans, the Food and Drugs Administration (FDA) advise against consumption of American lobster guts content, used as a sauce and locally known as tomalley, when high levels of PSP toxins are registered in the Atlantic coast (FDA, 2008).

5. EU production of echinoderms, tunicates, marine gastropods and crustaceans

The increase of seafood consumption and the search for new fisheries or aquaculture resources to satisfy the growing demand for these products may be exposing EU populations to new and not regularly monitored toxin vectors. Novel delicacy products and innovative solutions for fisheries and aquaculture have stimulated the production and capture of species for which current monitoring programs have not been appropriately designed for.

The EU production or capture of echinoderms and tunicates is

 $^{^{}a}$ Mouse Units were converted into STX equivalents, considering 1MU $= 0.18~\mu g$ STX.