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Harmful Algae 4 (2005) 717–723
HARMFUL ALGAE
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Investigation on the presence of domoic acid in Greek shellfish

I. Kaniou-Grigoriadou *, T. Mouratidou, P. Katikou

National Reference Laboratory on Marine Biotoxins, Centre of Veterinary Institutions of Thessaloniki, Ministry of Rural Development and Food, 3A Limnou Street, 546 27 Thessaloniki, Greece

Received 29 May 2003; received in revised form 2 August 2004; accepted 17 October 2004

Abstract

In the framework of the national monitoring program for marine biotoxins in bivalve molluscs for 2002 and 2003, a total of 862 samples were examined to investigate the presence of domoic acid (DA). Samples mainly consisted of mussels (*Mytilus galloprovincialis*) and venus clams (*Venus verucosa*) from various Greek harvesting areas. In 2002, 83% of all samples examined contained less that 1 μg DA/g, while in 2003 this percentage was 95%. In both years, DA was only detected in springtime without any sample exceeding the regulatory level. In 2002, the highest value was 14.0 μg/g in mussels, while in 2003 these were 4.2 μg/g in mussels and 5.6 μg/g in venus clams. This is the first report regarding the presence of ASP toxins in Greek shellfish.

Keywords: Domoic acid; Shellfish; Bivalve molluscs; Mussels; Venus clams; ASP

1. Introduction

Severe cases of amnesic shellfish poisoning (ASP) were first reported in late 1987 in the estuary of the Prince Edward Island River, Canada (Quilliam and Wright, 1989). Symptoms of the disease in humans included abdominal cramps, vomiting, headache, amnesia, coma and death (Todd, 1993).

The causative agent of this poisoning is domoic acid (DA), a widespread toxin, produced by several microalgal species belonging to the genus *Pseudo-nitzschia* (Bates, 1989). Domoic acid is a glutamate agonist that acts by binding to various glutamate receptors (Hampson and Manolo, 1998). Besides DA, several natural geometrical isomers have been identified which are not de novo products of the plankton, but are considered to be photo-isomers, less toxic than domoic acid itself. The C5′ diastereomer (epidomoic acid) is an isomer that shows very similar binding efficacy to the kainite receptor as DA itself and is included in the whole ASP toxicity.

In Europe, DA has been detected in wild and cultivated shellfish from Scotland (Gallacher et al., 2000) and Mediterranean coasts of France (Amzil

* Corresponding author. Tel.: +30 2310 552928; fax: +30 2310 566581.
E-mail address: nrlbiotoxins@in.gr (I. Kaniou-Grigoriadou).

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doi:10.1016/j.hal.2004.10.002

et al., 2001) and Italy (Orsini et al., 2002). In Greece, during the last few years, microalgae of the toxin-producing genus *Pseudo-nitzschia* (*P. pungens f. pungens*, *P. pseudodelicatissima*) have been found in Thermaikos Gulf (Northwest Aegean Sea) with the characteristic of high levels of abundance and short duration of their exponential phase of growth (Nikolaidis, 2002). ASP toxicity has been systematically monitored since late 2001 as part of the national monitoring program.

Internationally, the regulatory level of DA that has been adopted for public health is 20 μg DA/g; this limit has also been established by the European Union (Directive 97/61 amending Directive 91/492). High levels of DA can be detected by using the mouse bioassay for PSP toxicity based on acidic extraction (AOAC, 1990). However, for routine testing, the reversed-phase HPLC-UV method of Quilliam et al. (1995) is more sensitive, reliable and provides quantitative data.

In the present study, the 2002 and 2003 data on the ASP toxicity monitored in Greece are reported.

2. Materials and method

2.1. Sampling

A total of 13 provinces participated in the Greek monitoring program implemented in 2002 and 2003 for marine biotoxins in shellfish (Fig. 1). Samples of the species *Mytilus galloprovincialis*, *Venus verucosa*, *Modiolus barbatus*, *Cardium* spp., and *Arca* spp. were examined on weekly basis for the presence of DA.

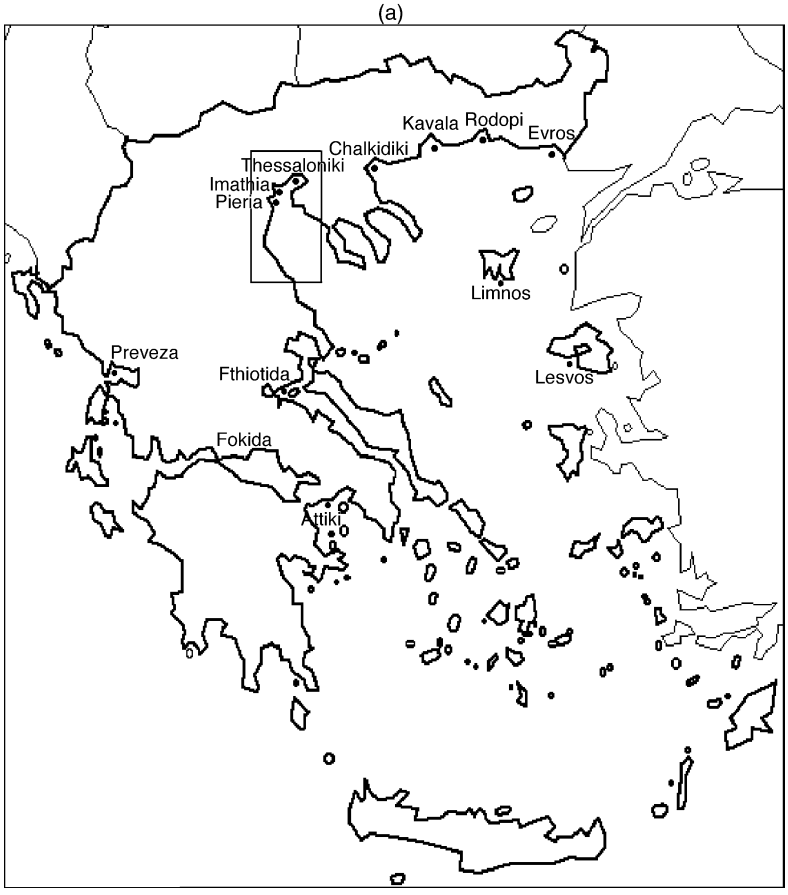

Fig. 1. (a) Locations of cultivated and wild shellfish on Greek coasts (b) sampling points along Thermaikos Gulf.