

Agri-Food & Biosciences Institute

VETERINARY SCIENCES DIVISION

Chemical Surveillance Branch

Annual Report National Reference Laboratory Marine Biotoxin

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Introduction

The Agri-Food and Biosciences Institute assumed the role of National Reference Laboratory on the 1st April 2008 and under the terms of the contract is required to provide an annual report on its activities. The UK-NRL for Marine Biotoxins is appointed by the FSA to carry out the following requirements and duties, as set out in Article 33 of Regulation (EC) 882/2004:

- 1. Collaborate with the Community Reference Laboratory (CRL) in their area of competence.
- 2. Co-ordinate, for their area of competence, the activities of official laboratories responsible for the analysis of samples.
- 3. Where appropriate, organise comparative tests between the official national laboratories and ensure an appropriate follow-up of such comparative testing.
- 4. Ensure the dissemination to the competent authority and official national laboratories of information that the Community Reference Laboratory supplies.
- 5. Provide scientific and technical assistance to the competent authority for the implementation of co-ordinated control plans adopted in accordance with Article 53.
- 6. Be responsible for carrying out other specific duties provided for in accordance with the procedure referred to in Article 62(3), without prejudice to existing additional national duties.

Collaboration with the Community Reference Laboratory:

The NRL collaborated with the CRL on arrange of issues including,

- The application of EC 226/2002 in respect of the testing of scallops for the presence of marine biotoxins and in particular, domoic acid (ASP).
- The updating of information on the testing of gastropods, tunicates and echinoderms for the presence of marine biotoxins. The NRL, in collaboration with the CRL and other EU NRLs, compiled information on the status of testing throughout the EC. The NRL presented this data at the 11th CRL-EU NRL meeting in Ljubljana in October 2008.
- The extension of CRL proficiency tests to allow the participation of both UK monitoring laboratories in 2009.
- Participation of both laboratories in proficiency tests for lipophilic toxins (DSP), PSP and ASP
- Responses to the reports on the CRL proficiency tests in 2008. The NRL raised concerns on the decisions taken to exclude data in both the lipophilic toxin test and the PSP test. The issues arising were addressed in the UK NRL response to the DG

SANCO request for details on the test protocols used within Member States. This report is currently with Food Standards Agency for consideration.

- Participation in the working group on development of an LC-MS/MS method for the detection of lipophilic toxins and subsequent participation in a performance trial of the method.
- Participation in the 11th CRL-EU NRL meeting in Ljubljana and production of the minutes of the meeting. The UK NRL was asked and has agreed to act as rapporteur at future meetings.
- The NRL has sought CRL advice on a number of technical issues relating to the application of methods. These include, for example, the use of species-specific laboratory reference material in the analysis of PSP by HPLC and the application of current methods to species other than bivalve molluscs.

Co-ordination of the Activities of the Monitoring Laboratories

Under its remit to co-ordinate the activities of the monitoring laboratories, the NRL has;

- Initiated a review of all UK NRL Standard Operating Procedures through correspondence and video conference with the UK monitoring laboratories. This process is addressing ambiguities in the current protocols and updating these, where necessary, as a consequence of recent validation data. Where possible, a more performance-based approach to the NRL SOPs is being undertaken. The protocol for the determination of domoic acid by HPLC with UV detection has been selected for redrafting in this format.
- Reissued draft guidelines for the collection of bulk contaminated material which is currently undergoing redrafting in response to the comments made.
- Collated the responses from the United Kingdom to the European Food Safety Authority's (EFSA) request for data on marine biotoxin testing in Member States. To date, data has been supplied on domoic acid, lipophilic toxins (okadaic acid, dinophysis toxins, azaspiracid, pectenotoxins and yessotoxin) and the PSP group.

Meetings of the UK NRL network group were held in June and November 2008 and minutes have been published and circulated. Action points arising from the June meeting have been addressed and action points from November are being progressed.

During 2008-2009, Food Standards Agency Scotland undertook a review of the Scottish biotoxin monitoring programme. The UK NRL was invited to participate in the working groups in an advisory capacity. The Working group has issued its report, which is currently under consideration by Food Standards Agency Scotland.

Dissemination of information Received from the Community Reference Laboratory

The NRL has circulated all documents, received from the CRL, to FSA and CEFAS. These have included papers associated with, the CRL network meeting, working groups on the development of an LC-MS/MS protocol for lipophilic toxins and a HPLC protocol for PSP toxins. Information on homogeneity and stability testing of proficiency test samples has also been circulated. All reports relating to pre-validation studies and proficiency tests have been issued to the UK NRL network.

To assist in the dissemination of information, the Biometrics Branch of Agri-Food and Biosciences Institute is currently developing web pages within the AFBI web site, specific for NRL Marine Biotoxin services. The NRL is currently liaising with the CRL to permit information from the CRL to be placed on the site. It is anticipated that the pages will be accessible during 2009-2010

Comparative Tests between the Official National Monitoring Laboratories

Official Monitoring for the United Kingdom is sited at two official control laboratories. With only two participants, the statistical evaluation of the results of comparative tests is impossible. In an attempt to rectify this problem, preliminary approaches were made by the NRL to run a joint programme with the Republic of Ireland. The NRL sought the agreement of the CRL to extend the current CRL proficiency tests to include both UK monitoring laboratories; agreement was reached for the inclusion of both UK laboratories in the proficiency test programmes for 2009.

Following analysis of the CRL proficiency test samples, the NRL provided CEFAS with the remaining tissue. The data produced by CEFAS could not be included in the statistical analysis but provided the UK competent authorities with information on the performance of both UK laboratories. Insufficient sample remained to allow analysis for Domoic Acid (ASP) by both laboratories, however both labs participate in the QUASIMEME programme and the laboratory results for Round 55 are included in the table below.

	Analyte	Sample Number	Result	Assigned	Z-Score	Method
AFBI	Lipophilic Toxins	CRLMB/08/L/04 (Mussel WF)	Positive	Positive	N/A	Mouse Bioassay
AFBI	Lipophilic Toxins	CRLMB/08/L/03	304.5	291	0.616	LCMS-MS
	(µg/kg)	(R.Clam WF)				
AFBI	Lipophilic Toxins	CRLMB/08/L/04	585.7	432	1.638	LCMS-MS
	(µg/kg)	(Mussel WF)				
AFBI	Lipophilic Toxins	CRLMB/08/L05	34.9	No Value	N/A	LCMS-MS
	$(\mu g/kg)$	(Mussel WF)				
CEFAS	Lipophilic Toxins	CRLMB/08/L03			N/A	LCMS-MS
	(µg/kg)	(R. clam WF)	258	291		
CEFAS	Lipophilic Toxins	CRLMB/08/L04			N/A	LCMS-MS
	(µg/kg)	(Mussel WF)	479	432		
CEFAS	Lipophilic Toxins	CRLMB/08/L05			N/A	LCMS-MS
	$(\mu g/kg)$	(Mussel WF)	59.4	No Value		
AFBI	PSP	CRLMB/08/P/01	1302	2554	-1.634	Mouse Bioassay
	(µg/kg)					
AFBI	PSP	CRLMB/08/P/01	1604	2554	Rejected	HPLC
	(µg/kg)					
AFBI	PSP	CRLMB/08/P/02	Negative	N/A	N/A	MBA and HPLC
CEFAS	PSP	CRLMB/08/P/01	1546	2554	N/A	HPLC
	(µg/kg)					
CEFAS	PSP	CRLMB/08/P/02	Negative	N/A	N/A	HPLC
	(µg/kg)					
AFBI	ASP	CRL/08/A/01	49.85	49.4	0.089	HPLC
	(µg/g)					
AFBI	ASP	CRL/08/A/02	<loq< td=""><td>Neg</td><td>N/A</td><td>HPLC</td></loq<>	Neg	N/A	HPLC
	(µg/g)					
AFBI	ASP	QST059SS	0.93	0.87	0.3	HPLC
	$(\mu g/g)$					
AFBI	ASP	QST060BT	6.52	6.30	-0.1	HPLC
	(µg/kg)					
AFBI	ASP	QST061BT	27.76	25.57	0.7	HPLC
	(µg/g)					
CEFAS	ASP	QST059SS	0.88	0.87	0	HPLC
	$(\mu g/g)$				\$	
CEFAS	ASP	QST060BT	6.08	6.30	-0.3	HPLC
	(µg/g)	-			0.0	
CEFAS	ASP	QST061BT	25.81	25.57	0.1	HPLC
	(µg/g)	-			0.1	

UK results in CRL Proficiency Tests 2008

Provide Scientific and Technical Assistance to the Competent Authority

In response to concerns raised on the testing of species other than bivalve molluscs, the NRL reissued a questionnaire to EU NRLs on the testing of gastropods, echinoderms and tunicates. In response to a request from the monitoring laboratories and the Competent Authority, the NRL undertook a review of literature on the potential of using indicator species for assessing the risk of biotoxin contamination in gastropods, tunicates and echinoderms and presented the report to Food Standards Agency in August 2008.

On behalf Food Standards Agency, the NRL is currently undertaking studies to determine the effectiveness of the current methodologies for testing gastropods for the presence of marine biotoxins. The study is under way on periwinkles and will include whelks and abalone.

The NRL has agreed to undertake a study on the extraction and clean-up protocols for those toxins within the lipophilic toxin group. This work will be undertaken in 2009-2010.

In response to requests from DG SANCO through Food Standards Agency and through their representative at the CRL-NRL Network meeting, the UK NRL has supplied detailed information on the application of the test protocols for marine biotoxins within the United Kingdom. The NRL response is currently with FSA for consideration before any response is made to DG SANCO.

Date	Venue	Subject		
16 th April 2008	FSA, Aviation House, London	NRL Set-up Meeting		
30 th April 2008	DGSANCO, Brussels	CRL Working Group on		
		LCMS		
24 th June 2008	FSA, Aviation House, London	14 th NRL Network Meeting		
16 th October 2008	FSANI, Clarendon Dock, Belfast	Progress meeting with FSA		
22 nd October 2008	Ljubljana, Slovenia	CRL Working Group on EFSA		
		reports on Biotoxins		
23 rd -24 th October 2008	Ljubljana, Slovenia	11 th CRL-EU NRL meeting		
$3^{\rm rd} - 7^{\rm th}$ November 2008	Hong Kong	The 13 th International		
		Conference on Harmful Algae		
25 th November 2008	FSA, Aviation House London	15 th NRL Network Meeting		
8 th December 2008	FSAS, Aberdeen	FSAS Biotoxin Monitoring		
		Redesign Working Group		
		meeting		
12 th February 2009	Millennium Hotel, Glasgow	Biotoxin Monitoring Redesign		
		Working Group meeting		
4 th March 2009	AFBI - Stormont	Video Conference with		
		CEFAS on UK NRL SOPs		

Meetings Attended