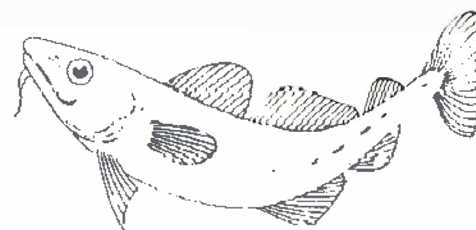
ISSN 0142-2499

AQUATIC ENVIRONMENT MONITORING REPORT
Number 35



Monitoring and Surveillance of Biological Contaminants and Disease in the Aquatic Environment 1990

Directorate of Fisheries Research
Lowestoft, 1993

MINISTRY OF AGRICULTURE, FISHERIES AND FOOD
DIRECTORATE OF FISHERIES RESEARCH

AQUATIC ENVIRONMENT MONITORING REPORT
Number 35

Monitoring and Surveillance of Biological Contaminants and Disease in the Aquatic Environment 1990

LOWESTOFT
1993

This report has been compiled by D. J. Alderman, BSc, PhD, MI Biol. of the MAFF, Fish Diseases Laboratory, 14 Albany Road, Granby Industrial Estate, Weymouth, Dorset DT4 9TH, from whom further copies may be obtained.

Aquat. Environ. Monit. Rep., MAFF Direct. Fish. Res., Lowestoft, (35), 35pp.

© Crown copyright, 1993

Requests for reproduction of material contained within this report should be addressed to MAFF

CONTENTS

Page

FOREWORD

BIOTOXINS

1. The monitoring programme for PSP and other shellfish toxins ... 7
- 1.1 Introduction ... 7
- 1.2 The 1990 sampling programme ... 7
- 1.3 Materials and methods ... 8
  - *1.3.1 Sampling and analysis* ... 8
  - *1.3.2 'Action' levels and procedures* ... 8
- 1.4 Results ... 9
  - *1.4.1 Molluscs* ... 9
  - *1.4.2 Crustaceans* ... 13
- 1.5 Discussion ... 14
- 1.6 Chemical analysis of PSP toxins ... 14
  - *1.6.1 Introduction* ... 14
  - *1.6.2 Materials and methods* ... 15
  - *1.6.3 Results* ... 15
  - *1.6.4 Discussion* ... 16
- 1.7 Chemical analysis of DSP toxins ... 16
  - *1.7.1 Introduction* ... 16
  - *1.7.2 Materials and methods* ... 17
  - *1.7.3 Results* ... 17
- 1.8 Investigation of other algal blooms ... 17

MARINE FISH DISEASES MONITORING PROGRAMME

2. The monitoring programme for determination and surveillance of disease in marine fish ... 17
- 2.1 Historical background ... 17
- 2.2 Materials and methods ... 18
  - *2.2.1 Areas monitored* ... 18
  - *2.2.2 Sampling methods* ... 18
  - *2.2.3 Recording of gross pathological changes in dab* ... 18
  - *2.2.4 Organs sampled for histological examination and assessment of stress in dab* ... 19
  - *2.2.5 Recording of gross pathological changes in other fish species* ... 19
  - *2.2.6 Fish disease returns from MAFF's Fisheries Inspectorate* ... 19
- 2.3 Results ... 19
  - *2.3.1 RV CORYSTES Cruise 6a, 5-11 May 1990* ... 19
  - *2.3.2 RV CIROLANA Cruise 8, 15 August-17 September 1990* ... 20
  - *2.3.3 RV CORYSTES Cruise 11, 18-22 October 1990* ... 26
  - *2.3.4 Monitoring catch landed at ports on the north-east coast of England* ... 27

STATUTORY MONITORING FOR FISH AND SHELLFISH DISEASE

3. The monitoring programme for control of disease in farmed stocks and its transmission to wild stocks ... 28
- 3.1 Introduction ... 28
- 3.2 Materials and methods ... 29
  - *3.2.1. Sampling methods* ... 29
  - *3.2.2 Laboratory tests* ... 29
- 3.3 Field investigations under the Diseases of Fish Acts (1937 and 1983) ... 29
  - *3.3.1 Investigations into the cause of disease outbreaks in fish* ... 29
  - *3.3.2 Investigations to monitor for specific diseases of fish* ... 30
  - *3.3.3 Re-tests for specific diseases of fish on designated sites* ... 30
- 3.4 Field investigations under the Sea Fisheries (Shellfish) Act (1967) ... 30
  - *3.4.1 Investigations to monitor for specific diseases of shellfish* ... 31
  - *3.4.2 Tests for gaffkaemia* ... 32
  - *3.4.3 Tests for crayfish plague* ... 32

4. References ... 33

ANNEX 1. **Areas of monitoring mentioned in the text and staff responsible for the projects ... 35**

FOREWORD

Environmental monitoring is the basis of any scientific regulation for the protection of the environment. One of the major responsibilities of MAFF's Directorate of Fisheries Research (DFR) is to conduct such monitoring for the aquatic environment and the results of these studies are published annually in two separate reports on radioactive and non-radioactive contaminants respectively.

Although these reports do reflect work on the biological effects of contaminants, a major new development in recent years has been the idea that some diseases of marine organisms may be related to contamination. This has led to an intensification of work on this subject and the time therefore seems right to publish the data on a regular basis alongside that in other areas of MAFF's environmental scrutinies.

This then is the first report on environmental contamination and disease and covers the work in 1990. It includes the last major study by DFR of the seasonal distribution of paralytic shellfish poisoning toxins. This area of work has now been transferred to the Torry Research Laboratory, but future developments in DFR for the monitoring of the algae which produce these and other toxins is likely to develop further under recent EC Directives.

The association between fish disease and contaminants continues to be an emotive subject. This difficult area of study can only be dealt with properly by the careful conduct of work and the regular publication of all data.

Statutory fish disease work continues to be the principal government responsibility in relation to fish farming and towards ensuring that fish farms and natural fish populations co-exist without harm to each other. New developments within the EC single market measures require additional monitoring in this context and future reports will reflect this increased commitment.

It is expected that this component of MAFF's Aquatic Environment Report Series will in future be published annually.

C. E. Purdom
Deputy Director
(Aquatic Environment Protection)
Ministry of Agriculture, Fisheries and Food
Directorate of Fisheries Research

1. THE MONITORING PROGRAMME FOR PSP AND OTHER SHELLFISH TOXINS

1.1 Introduction

Since 1968 a survey for the presence of paralytic shellfish poisoning (PSP) toxins in bivalve shellfish has been undertaken annually by the Directorate of Fisheries Research (DFR) of the Ministry of Agriculture, Fisheries and Food (MAFF). These surveys commenced as a result of the high levels of PSP contamination of mussels (*Mytilus edulis*) which occurred in 1968 and which resulted in 78 persons being affected by paralysis after the consumption of mussels from the Holy Island and Budle Bay areas of Northumberland (McCollum *et al.*, 1968; Wood and Mason, 1969).

The 1968 outbreak clearly demonstrated that there was a significant risk from PSP toxins to consumers of bivalve shellfish from the north-east coast of Britain in spring and early summer. At that time, no other Department or Authority was able to take on a monitoring programme to protect consumers and, in addition, the reputation of the British shellfish industry was also at risk (P. C. Wood, pers. comm.). It therefore fell to DFR to institute a monitoring programme, which operated from April to August from 1969 to 1990.

From 1969 to 1987, the programme was based at DFR's Burnham-on-Crouch Laboratory, but was transferred to the Fish Diseases Laboratory (FDL) at Weymouth from the 1988 season, with the handing over of responsibility for research into shellfish hygiene to that laboratory.

In the years after 1968, peaks of PSP toxicity were detected during each season, although never at the levels reached in 1968 itself. Warnings against the consumption of mussels were posted in affected areas when levels of PSP approached safety limits and no new cases of PSP in humans were reported after the commencement of DFR's monitoring and advisory programme. The results of the monitoring surveys were published in various formats up to 1983. Much of the information on the first 10 years of PSP monitoring to 1977 was summarised by Ayres and Cullum (1978). No monitoring reports were published between 1984 and 1986, although the monitoring programme was continued and internal reports were produced. The results of the 1987 and 1988-1989 PSP programmes were included in the reports on monitoring and surveillance of non-radioactive contaminants in the aquatic environment for the years 1984-1987 and 1988-1989 (MAFF, 1990, 1991).

A review which updates and supersedes the report of Ayres and Cullum (1978), summarising the pattern of PSP detected during the 22 years of DFR's monitoring programme, is in the course of preparation.

This report presents data from the 1990 monitoring programme, during which levels of toxins higher than anything seen since 1968 were recorded. Some details of the sampling programme conducted on the east coast of Scotland by the Scottish Office Agriculture and Fisheries Department (SOAFD) are also included.

1.2 The 1990 sampling programme

As in previous years, sample boxes and packing materials for mussel samples were sent to Environmental Health Departments (EHDs) in late March 1990. Sampling sites comprised:

(a) the north-east coast of England;
(b) Berwick Harbour;
(c) the east coast of Scotland.

The EHDs concerned were those of selected District Councils on the north-east coast of England, from Scarborough north to the Scottish border. These were sites which had been used for most of the 22 years of DFR's monitoring programme and were largely chosen as a result of early experience with the patterns with PSP in bivalves (Figure 1). North of the Scottish border, on behalf of SOAFD, the Scottish Sea Fisheries Inspectorate collected mussel samples from three sites on the Scottish east coast at two-weekly intervals.

As a result of the high levels of PSP detected off north-eastern England in late May by the routine monitoring, an expanded programme of sampling was undertaken from that area, from a large number of other sites around the coasts of England and Wales, and from the east coast of Scotland. This widening of the monitoring plan also included molluscs other than mussels and crustaceans. Following the expansion of the sampling programme in England and Wales in early June, the routine and expanded monitoring programme in Scotland became the responsibility of MAFF's Torry Research Station (TRS) in Aberdeen, acting on behalf of SOAFD. All east coast (England and Scotland) and all MAFF area (England and Wales) PSP monitoring samples for 1990 are reported here.

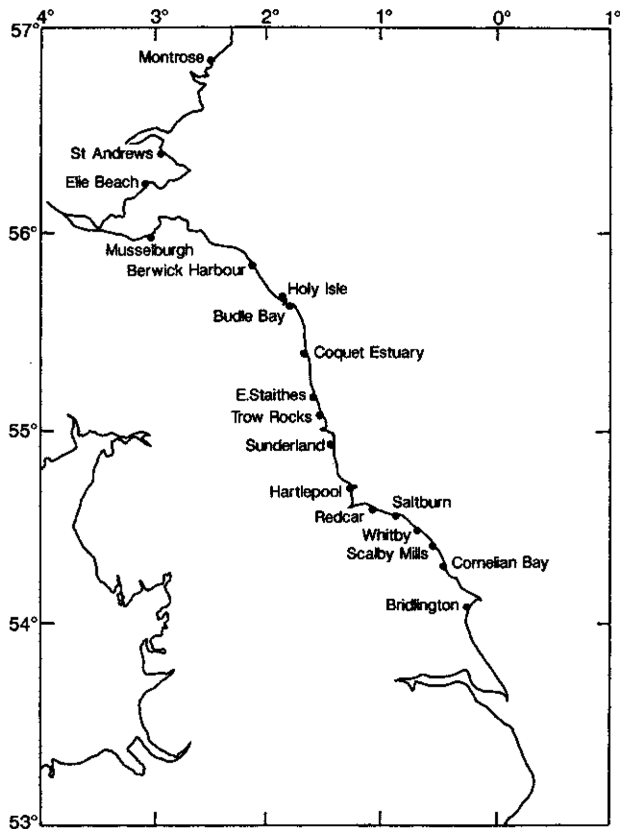


Figure 1. Stations sampled during routine monitoring for PSP in mussels on the north-east coast of England

The data from Scottish sites will be published more fully elsewhere.

1.3 Materials and methods

1.3.1 Sampling and analysis

Routine samples were collected at low tide on Monday or Tuesday of each week and dispatched by post to FDL at Weymouth. These samples consisted of about 100 g (roughly 20) whole mussels (*Mytilus edulis* L.). The mussels were opened and 50 g of meat and shell liquor were harvested and weighed.

Samples within the expanded sampling programme were collected and dispatched to FDL or TRS by the most convenient transport route, including special courier services where necessary. Scallop samples were collected from chartered fishing vessels.

All samples were processed within 36 h of receipt at FDL, most within 18 h. The analytical procedure employed was the standard mouse bioassay (Greenburg and Hunt, 1985).

Samples of species other than mussels were processed in a similar manner to mussels. Oyster samples consisted of 50 g shucked whole meats Scallops were opened and the different tissues dissected out dependant on the objective of the particular sample. The different tissues were either muscle (M), gonad (G) and viscera and mantle (V), or edible (E) (consisting of M+G) and non-edible (N) components, pooling animals in groups where necessary to obtain sufficient weight of tissue for processing. Crustacean samples consisted of whole pooled animals (shrimps and *Nephrops*), digestive glands of individual (or pooled) animals (crab and lobster) and/or white meat (crab), dependant on the reason (i.e. the specific problem being addressed) for which the sample was taken.

An equivalent volume of dilute HCl (0.18N) to the weight of tissue was added, the pH checked and, if necessary, adjusted with 5N HCl or 0.18N NaOH to lie between pH 2.0 and 4.0, the whole transferred to the jar of a Waring blender (New Hartford, Conn.) and blended thoroughly. After blending, the sample was heated to 100°C for 5 minutes, then cooled and the pH rechecked and, if necessary, adjusted as before.

The presence of PSP toxins in the clear supernatant was then assessed by the standard mouse test. Female white mice (of about 20 g in weight) were injected (by the intraperitoneal route) with 1.0 ml of the supernatant. Two mice were used in each test. The mice were then observed for 30 minutes to detect behavioural differences and to determine time of death. The level of toxin present in a sample was determined from standard calibration tables (Greenburg and Hunt, 1985) of survival time and mouse weight and expressed in terms of mouse units (mu). This abbreviation used in this report refers to mouse units per 100 g of tissue (saxitoxin equivalent 80 µg/100 g^{-1} tissue). One mouse unit of saxitoxin will kill a 20 g mouse in 15 minutes.

1.3.2 'Action' levels and procedures

A standard 'action' level of 400 mu for PSP toxins has been operated over many years in the UK. This is identical to the 'action' level which is used in the USA and Canada. When the threshold level of 400 mu is exceeded, shellfish are regarded as being unsafe for human consumption. The testing laboratory then informs the Department of Health (DoH) Food Safety Division, the Environmental Health Departments (EHDs) of the District Councils involved, plus the District Office of the Sea Fisheries Inspectorate, and Marine Environment Protection and Fisheries Divisions of MAFF. The DoH and the local EHD take action to post warning signs on the affected coastline and to publicise the risks from consumption of local shellfish.

1.4 Results

1.4.1 Molluscs

(a) Mussels - north-east coast of England and east coast of Scotland (Table 1): Sampling commenced in the week ending 13 April. First signs of PSP toxin were detected in samples collected in the week ending 18 May, when toxin was present at Whitby (218 mu), Redcar (245 mu), Trow Rocks, south Tyneside (518 mu), Cresswell (210 mu) and the Coquet Estuary (209 mu). The Trow Rocks sample at 518 mu exceeded the 'action' level of 400 mu and the DoH and the local EHD were informed accordingly.

The samples collected the following week (i.e. those samples available for processing on Thursday 24 May) were tested on the morning of 25 May, when the Trow Rocks sample was found to contain 19 881 mu, a level of PSP toxin not reached in British waters since 1968. Additionally, levels of above the 'action' level were detected in samples from Berwick Harbour (903 mu), the Coquet Estuary (1 119 mu), Hartlepool (909 mu) and Whitby (848 mu), whilst its presence at St Andrews (250 mu), Elie Beach (338 mu), Holy Island (386 mu), Saltburn (255 mu) and Scalby Mills (300 mu) showed that it had become very widespread in less than 14 days.

By 1 June, PSP was present in mussels from all sites between Saltburn and Elie Beach (except Musselburgh) and levels at Trow Rocks (Figure 2(a-b) had fallen markedly, but were still high (3 217 mu). Mussels from Sunderland contained 7 178 mu. By the week ending 15 June, toxin was essentially confined to the area between Coquet estuary and south Tyneside, and the last sample above the 'action' level (807 mu) was detected at East Staithes in the week ending 22 June. North of the Scottish Border, in the routine sampling area, the final trace of toxin was

Table 1. Levels of PSP (mu) in mussels (Mytilus edulis), sampled during routine monitoring of the north-east coast of England and the east coast of Scotland

Week Ending	13-Apr	20-Apr	27-Apr	04-May	11-May	18-May	25-May	01-Jun	08-Jun
Montrose	0		0				0	0	
St Andrews							250		
Elie Beach	0			0			338	328	
Musselburgh		0		0				0	
Berwick Harbour						0	903	464	
Holy Island							386	599	525
Budle Bay			0				0	776	394
Coquet Estuary		0	0			209	1119	3124	536
E Staithes		0		0				*4733*	*2080*
Trow Rocks	0		0	0	0	518	19881	3217	1600
Sunderland					0			7178	435
Hartlepool						0		1972	
Redcar	0		0	0	0	245	909	542	392
Saltburn	0		0	0	0	0	255	372	329
Whitby	0		0	0	0	218	848		0
Scalby Mills	0		0	0			300	0	0
Cornelian Bay	0		0	0		0		0	0
Bridlington								0	0

Week Ending	15-Jun	22-Jun	29-Jun	6-Jul	13-Jul	20-Jul	27-Jul	3-Aug	10-Aug	17-Aug
Montrose	0	0	0	0	0	0	0	0	0	0
St Andrews		0	0	214	0	0		0		0
Elie Beach	0	0		0	0		0	0		0
Musselburgh	0		0		0		0	0		
Berwick Harbour			0		0		0	0		
Holy Island	0	0								
Budle Bay			0		0			0		
Coquet Estuary	231	0	0	0	0	0	0			
E Staithes	616	807	0		0		0			
Trow Rocks	*1020*		*298*	*364*	*0*	276	*0*	*0*		
Sunderland		0			207		0			
Hartlepool	0									
Redcar	0	200	0	0	0	0				
Saltburn	218	0	0	0	0	0				
Whitby	0		0	0			0	0	0	
Scalby Mills	0	0	0	0		0	0	0	0	
Cornelian Bay	0	0	0	0	0		0	0		
Bridlington	0									

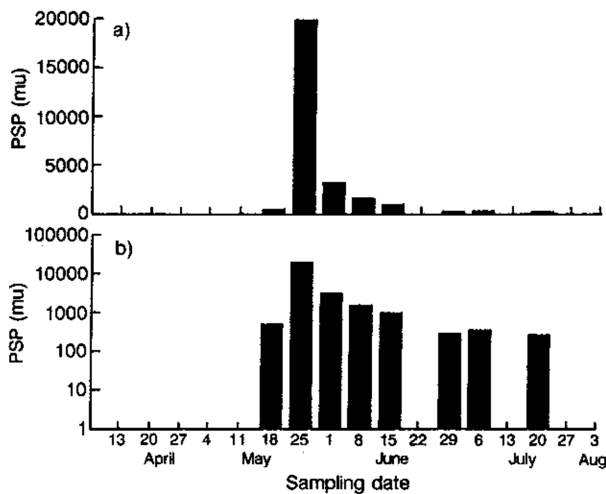


Figure 2. ***Weekly levels of PSP detected in mussels at Trow Rocks: (a) arithmetic plot and (b) log scale***

detected at St Andrews on 6 July but essentially, on the Fife coast, the toxicity problem was confined to the end of May and early June.

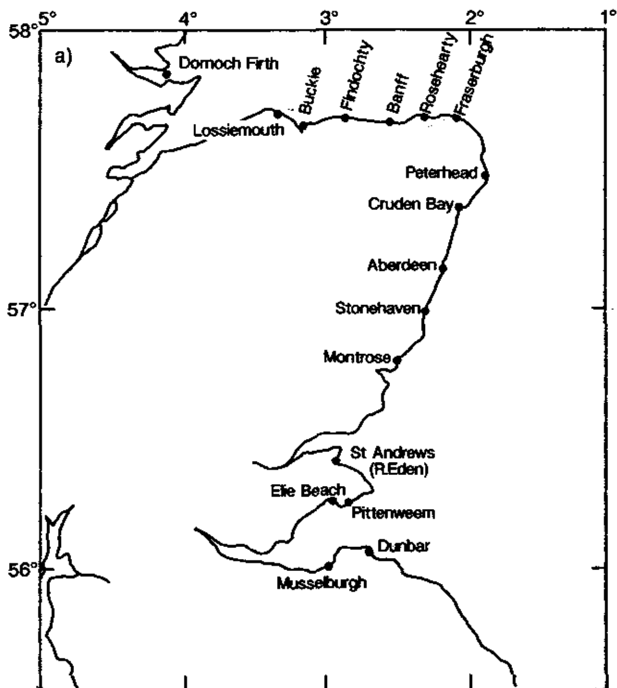
In Scottish waters, an extension of sampling points north along the east coast from Montrose (Table 2, Figure 3(a)) identified further areas of toxic contamination, but not at the high levels found further south.

(b) Extended survey - England and Wales (Figure 3(b)): In the week following the detection of serious levels of toxin in mussels on the north-east coast, a series of 'precautionary' samples was taken around the coasts of England and Wales. The majority of these samples were of mussels but, where appropriate to local bivalve fisheries, oysters and cockles were also sampled. This sampling was repeated in the week ending 6 July. Details of sites sampled are shown in Figure 3(b). No evidence of PSP-type neurotoxin was detected in any of these samples.

Table 2. Levels of PSP (mu) in mussels, sampled during routine monitoring of the east coast of Scotland

Week ending	13-Apr	20-Apr	27-Apr	04-May	11-May	18-May	25-May	01-Jun	08-Jun
Dornoch Firth								0	0
Burghead/Lossiemouth									
Buckie									
Findochty									
Banff									
Rosehearty									
Fraserburgh									
Peterhead									
Cruden Bay									
Aberdeen									
Stonehaven									
Montrose	0		0				0	0	
St Andrews							250		
Pittenweem									
Elie Beach	0			0			338	328	
Musselburgh		0		0				0	
Dunbar								0	
St Abbs/Eyemouth									

Week ending	15-Jun	22-Jun	29-Jun	6-Jul	13-Jul	20-Jul	27-Jul	3-Aug	10-Aug	17-Aug
Dornoch Firth		0	0	0	0		0	0		0
Burghead/Lossiemouth		270/252			0					
Buckie		0		0	0					
Findochty				0						
Banff		428/813	198	260/314		0	0			
Rosehearty				357	256	248			0	
Fraserburgh			325				181			
Peterhead			0							
Cruden Bay		360	250							
Aberdeen	280	192	0				0			
Stonehaven		0								
Montrose	0	0	0	0	0	0	0	0	0	0
St Andrews		0	0	214	0	0		0		0
Pittenweem							0			
Elie Beach	0	0		0	0		0	0		0
Musselburgh	0		0		0		0	0		
Dunbar		0								
St Abbs/Eyemouth			0		0					



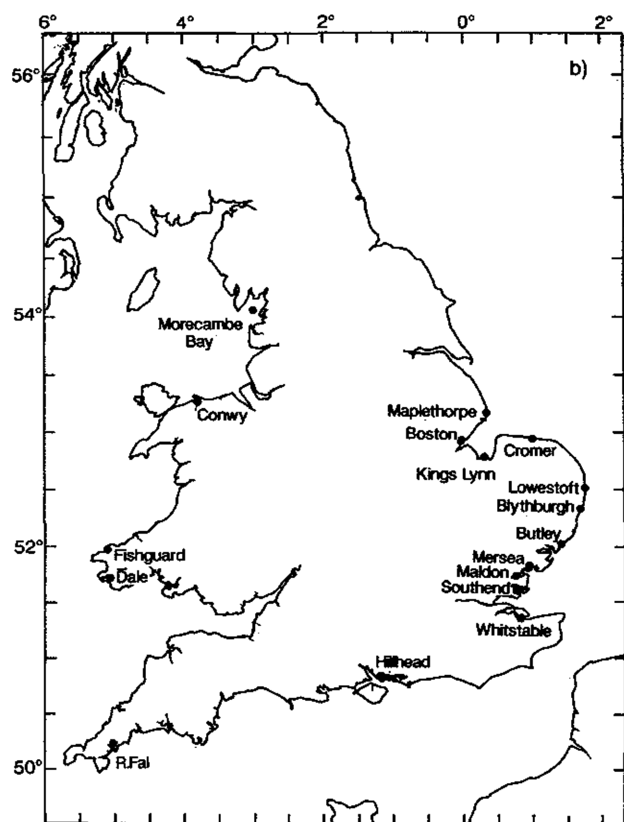


Figure 3. Stations sampled during extended monitoring for PSP in mussels: (a) in Scottish waters and (b) in English and Welsh waters

(c) Scallops - Table 3 (Figure 4(a)): Scallops are known to be a risk when mussel contamination is high (Ayres and Cullum, 1978). The levels found in mussels since 1980 were so low, however, that sampling of scallops was deemed unnecessary over that period. However, once high levels of toxin had been detected in mussels on the north-east coast, samples of scallops were also collected, principally from the scallop beds on the Farne Banks of the Northumberland coast, but also from the Flamborough Head region and from 80 miles north and 50 miles north-east of Buckie. Scallops from the Farne Banks have presented problems in the past because they have remained toxic for considerably longer than other bivalves and, in contrast to mussels, represent a significant commercial fishery for the area.

An initial sample of scallops from stocks landed at Seahouses, showed high levels of toxin. More detailed sampling later showed that levels on various beds on the Farne Bank off Seahouses had already fallen, but were still close to the 'action' level and remained so into early July. Toxin levels were measured in small pooled groups of scallop tissues. The results showed that, although PSP toxin was most frequently detected in the viscera, all tissues, including muscle and gonad, could contain toxin near to or even above the 'action' level. However, considerable variation in levels of toxin between pooled groups in the same time sample was noted. The difficulty of obtaining samples from the different scallop beds in the region by dredging made interpretation of results difficult, but it seemed that scallops on the more southerly beds (east — south-east of Seahouses and Dunstanburgh Castle) had a higher level of contamination than those on other beds. This caused a delay in lifting warnings against consumption of scallops from the Farne Banks, because a few continued to show contamination above the 'action' level. Final clearance of Farne Bank was not possible until mid-July. In the waters north-east of Buckie, some toxicity (215 mu) was evident in scallops in late July.

(d) Gastropods - (Figure 4): Samples of whelks and winkles, which are fished commercially on the north-east coast of England, were examined in June. No PSP toxin was detected in samples from any of the sites indicated in Figures 4(a-b).

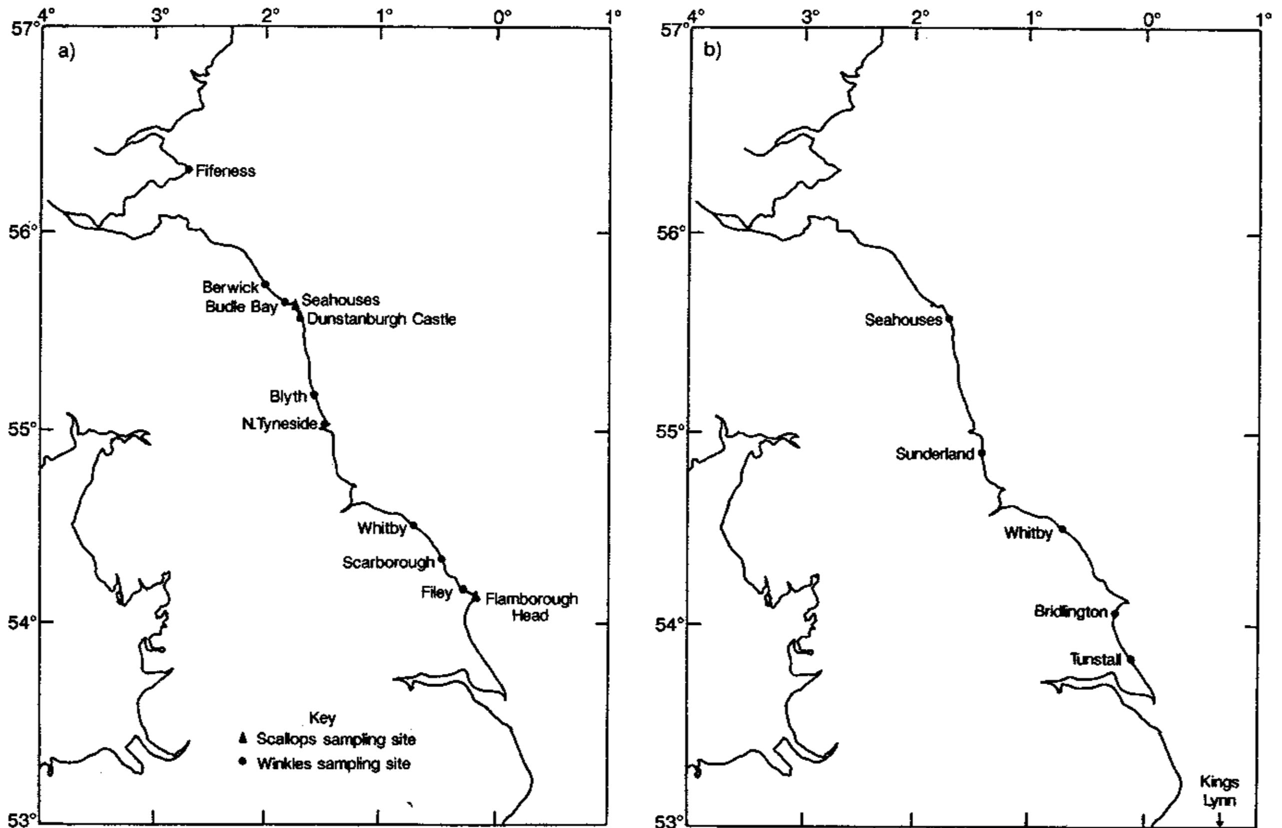


Figure 4. ***Stations sampled during routine monitoring for PSP on the north-east coast of England: (a) in scallops and winkles and (b) in whelks***

Table 3. ***Levels of PSP (mu) in scallops (Pecten maximus) from the north-east coast of England and the east coast of Scotland***

	Location	Number of pools	Number in pool	Tissue(1)	Number positive	PSP(mu)(2)
31-May	off Seahouses	1	10		1	1284
		1	10		1	1299
11-Jun	off Flamborough Head	1	2		1	365
14-Jun	Seahouses	12	3	E	10	316/229/190/212/266/0/230/241/213/296/0/221
		12	3	R	11	288/315/235/250/296/341/249/325/343/277/0/214
18-Jun	Seahouses	8	5	**	1	198
19-Jun	8 miles E of Seahouses	5	3	G	2	303/0/276/0/0
		5	3	M	1	240/0/0/0/0
		5	3	V	5	277/296/303/236/217
19-Jun	9 miles E of Dunstanburgh Castle	5	3	G	1	0/0/0/0/213
		5	3	M	2	220/0/196/0/0
		5	3	V	5	226/272/294/216/240
19-Jun	5.5 miles E of Dunstanburgh Castle	5	3	G	1	0/0/0/0/450
		5	3	M	3	0/0/222/272/203
		5	3	V	5	292/407/242/365/354
19-Jun	8 miles E of Seahouses	13	1		6	216/245/243/243/218/202
	9 miles E of Dunstanburgh Castle	14	1		5	253/200/215/250/213
	5.5 miles E of Dunstanburgh Castle	15	1		0	
28-Jun	8.5 miles E of Dunstanburgh Castle	20	2		5	250/203/236/217/203
	5 miles E of Dunstanburgh Castle	21	2		15	385/368/336/261/464/230/256/309/346/455/271/ 269/241/367/300
	9 miles ESE of Dunstanburgh Castle	23	2		12	227/210/220/205/212/187/207/265/204/258/194/217
	8 miles E of Seahouses	21	2		9	192/214/253/211/296/277/240/233/218
	9 miles E of Seahouses	22	2		4	233/216/204/229
28-Jun	8 miles E of Seahouses	15	2	**	1	211
11-Jul	10.5 miles E of Dunstanburgh Castle	20	2		7	215/207/210/210/206/234/203
	9 miles E of Seahouses	19	2		7	203/203/191/188/195/209/187
	9 miles ESE of Seahouses	19	2		12	193/201/192/168/213/275/188/203/229/203/208/212
	5.5 miles ESE of Dunstanburgh Castle	19	2		19	265/260/224/333/234/255/339/314/329/273/255/308/ 305/237/307/303/276/323/306
17-Jul	80 miles N of Buckie	1	5		0	
25-Jul	50 miles NE of Buckie	1	5		1	215

(1) Where 'Tissue' column is blank, whole animal tested, otherwise E=edible tissues, R=rest, G=gonad, M=muscle, V=viscera
(2) Where separate tissues are shown, negative samples are indicated to enable relationship between PSP levels in different tissues to be seen. Otherwise, negative samples are not given
*** Sample of Queen scallops*

1.4.2 Crustaceans

Bivalve molluscs traditionally have been regarded as presenting the significant hazard to consumers from PSP intoxication. It is possible, however, that some crustaceans may become secondarily intoxicated either directly by consumption of algae or their debris or by consumption of contaminated bivalves or zooplankton. Certainly, death of seabirds in the Farne Islands in 1968 and 1975 was attributed to secondary PSP intoxication in birds feeding on sand eels, which had presumably also derived their toxin secondarily through the food chain (Coulson *et al.*, 1968). When the extremely rapid rise in contamination of mussels was detected on the north-east coast, it was clear that, at these high levels of toxin, the risks to consumers from secondary intoxication in crustaceans could not be ignored. Warnings were issued accordingly and arrangements were made for the sampling of crustaceans from affected areas and elsewhere (Figure 5).

(a) Edible crabs - Table 4: PSP toxins were detected in samples of crabs from the Fife coast, southward to Bridlington. Levels above the 'action' limit were not found, however, and the prevalence of contaminated animals was low. A sample of crabs from Cromer in Norfolk, in week ending 1 June, also showed toxin in one sample of digestive gland pooled from two animals. A further sample of crabs from that site one week later proved to be negative. By mid-June, it was

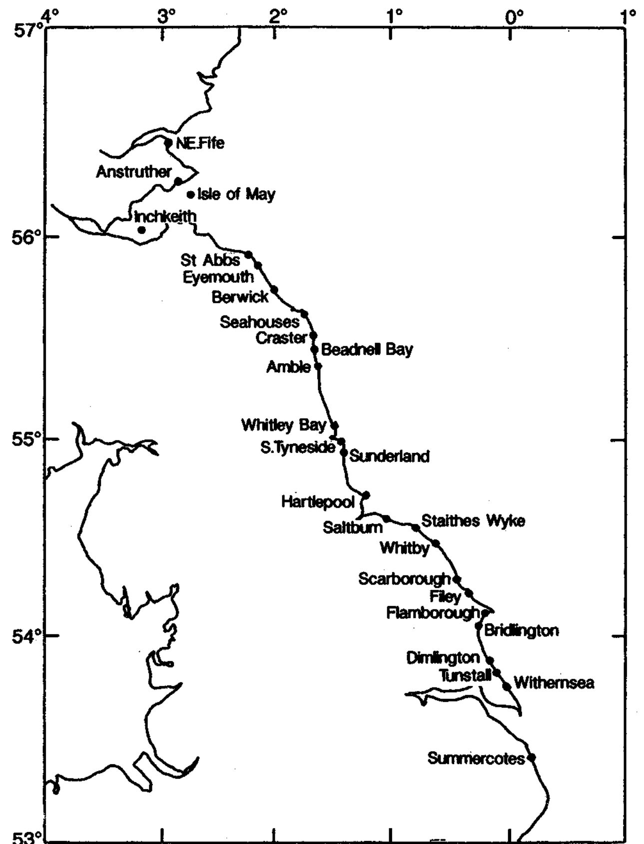


Figure 5. ***Stations sampled during routine monitoring for PSP in crustaceans on the north-east coast of England***

Table 4. Levels of PSP (mu) in crabs from the north-east coast of England

Week ending	01-Jun			08-Jun		
Site	Number	+ve	PSP	Number	+ve	PSP
NE Fife	4	1	197			
Anstruther	2	1	317			
Port Seton	4	0				
Eyemouth	4	0				
Berwick	4	0		25	0	
Seahouses	3	1	257			
Craster	1	1	243			
Beadnell Bay				20	2	265/229
Amble	2	0		30	0	
Whitley Bay				20	1	260
S Tyneside	2	0				
Sunderland				26	0	
Hartlepool				9	2	198/299
Saltburn	1	0		5	0	
Staithes Wyke				34	3	244/210/255
Whitby	1	0				
Scarborough	17	0				
Filey	4	0				
Flamborough	3	0				
Bridlington	4	0		25	1(2)	233
Tunstall	2	0				
Withernsea	9	0				
Dimlington	10	0				
Summercote	4	0				
Cromer	10	1(2)	211	25	0	
Mersea Creeks	4	0				

In many cases samples had to be pooled, either to provide sufficient tissue for analysis, or to improve handling speed. Numbers in pooled samples are indicated in brackets in the +ve column

clear that, although crabs on the north-east coast were intoxicated with PSP, presumably secondarily, the levels of toxin presented no risks to consumers from the consumption of such crabs. The DoH was thus able to lift the warning against consumption of crabs from the north-east coast.

(b) Lobster, *Nephrops* and Shrimps - Table 5: Although the initial warning against consumption of all shellfish included lobster, *Nephrops* and shrimp, samples collected from the fisheries on the north-east coast (Table 5), showed that PSP was present in lobsters, but that it was observed mainly in the digestive glands which are not eaten in the UK. It was therefore possible to lift warnings against consumption of these species and concentrate the resouces of sampling and analysis on species which presented a more evident hazard.

1.5 Discussion

Monitoring on the north-east coast of England and the east coast of Scotland resulted in the detection of the highest levels of neurotoxin in shellfish for 22 years. Additionally, the event also led to an assessment of the risk to consumers, from consumption of crustaceans. Toxin was detected in a number of samples of crustaceans, but the levels did not exceed the 'action' limit used for mussels. No outbreak of PSP in humans was reported.

1.6 Chemical analysis of PSP toxins

1.6.1 Introduction

The mouse test continues to be the only generally accepted assay for PSP toxin because of its rapid presentation of the 'sum' of toxicity of any sample. There is an obvious need, however, to replace this test as rapidly as possible with a method which offers equal or better protection to the consumer and avoids the use of live animals. HPLC methods (Sullivan and Wekell, 1986) are widely viewed as possible alternatives to the mouse test.

The mouse bioassay and HPLC analyses were both used on extracts from samples collected during the routine monitoring programme in the years 1987-1989. However, the concentrations of PSP toxins in mussels in those years were generally low and results could not be compared over a wide range of toxicity. In 1990, when shellfish contamination was high on the north-east coast of England and elsewhere, the method could be fully evaluated.

Table 5. Levels of PSP (mu) in lobsters, Nephrops and shrimps from the north-east coast of England

a) Lobsters

Week ending	01-Jun			08-Jun		
Site	Number	+ve	PSP	Number	+ve	PSP
Berwick				3	1	213
Amble	1	0		10	1(2)	257/273
Beadnell Bay				5	0	
Whitley Bay				6	0	
Sunderland	1	0		10	1(2)	229/208
Hartlepool				10	4	228/260/192/192
Saltburn	1	1	227	6	1(2)	290
Staithes Wyke				12	0	
Whitby	1	0				
Scarborough				7	0	
Bridlington	2	0				
Withernsea	1	1	206			

In many cases samples had to be pooled, either to provide sufficient tissue for analysis, or to improve handling speed , numbers in pooled samples are indicated in brackets in the +ve column

b) *Nephrops* : samples of 5 animals

Week ending	01-Jun		
Site	Number	+ve	PSP
Lossiemouth	1	0	
Isle of May	1	1	193
St Abbs Head	1	0	
Inchkeith	3	1	304
Sunderland	1	0	

c) Shrimp : Sunderland samples of 10 animals, PSP 0

1.6.2 Materials and methods

Levels of PSP toxins were determined by a standard mouse bioassay as described in sub-section 1.3.1 above. Concentrations were expressed in mouse units per 100 g of shellfish tissue (400 mouse units are equivalent to 80 µg of saxitoxin per 100 g of tissue). Sub-samples of shellfish extract were taken and stored at -20°C prior to HPLC analysis.

The HPLC method was identical to that described by Sullivan and Wekell (1986), in which the toxins were separated by ion interaction chromatography and detected by fluorescence spectrometry, following post-column oxidation with alkaline periodate.

Shellfish extracts were thawed and proteins removed by ultrafiltration prior to analysis by HPLC. Standardisation of the method was carried out using a commercially-available saxitoxin standard and other standards obtained from the US Food and Drug Administration (FDA) in Seattle. These consisted of a mixture of 10 toxins (C1, C2, B1, B2, GTX1, GTX2, GTX3, GTX4, NEO and STX) at known concentrations. A standard was analysed after each batch of 5 samples, in order to maintain good calibration and to monitor adequate separation of toxins.

The potency of individual toxins was calculated from reference tables provided by the US FDA, and the contribution of each toxin to the total toxicity was calculated and expressed as saxitoxin equivalents.

1.6.3 Results

Approximately 1 000 samples were assayed by the mouse test during the 1990 sampling programme and, at its height, more than 50 assays were undertaken daily. HPLC analysis of the samples began in January 1991, and are not yet complete. The results presented here represent analytical data available at the time of publication of this report.

Figure 6 shows the relationship between the values obtained by bioassay and HPLC for selected mussel samples. The correlation between the two data sets is reasonable ($r^2 = 0.79$). However, for data close to the 'action' limit of 400 mouse units, the correlation is poor ($r^2 = 0.28$), principally due to two (apparently false) positive values given by the HPLC technique.

Analyses for a limited number of mussel samples, carried out for each of the years 1987-1990, have shown a similar distribution of toxins, with C1/C2, GTX1 and GTX3 predominating, and with lower amounts of GTX2, GTX4, NEO and STX. B2 toxin was abundant in some samples, but few contained toxins of the other B series.

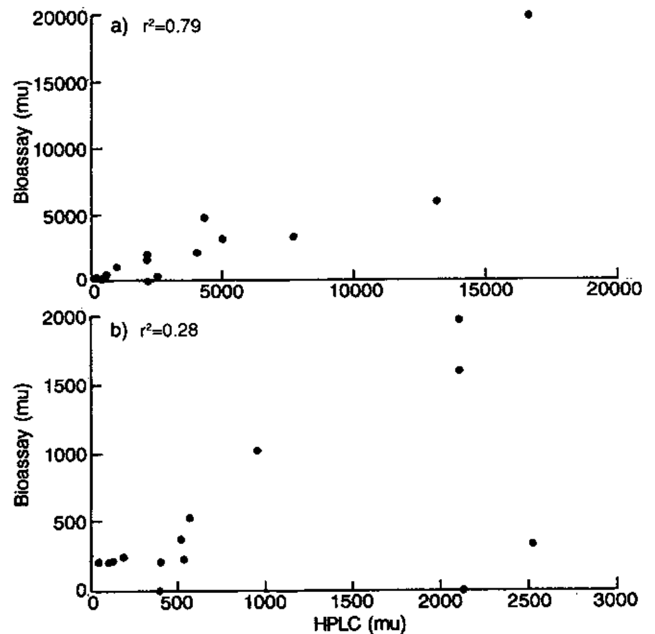


Figure 6. A comparison of mussel toxicity by mouse assay and HPLC: (a) all mussels sampled and (b) mussels with toxicities of up to 2000 mouse units

Temporal differences in the composition of toxins for a site at the centre of the toxic event, is shown in Figure 7. No clear succession of specific toxins was evident during the bloom. Well-defined spatial differences were not observed, and considerable variations in the composition of toxins within sample extracts were observed spatially, temporally, and for different species sampled at the same time in the same locality.

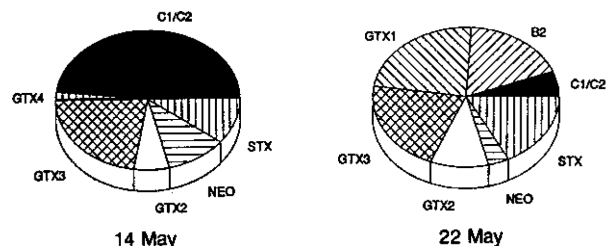


Figure 7. Temporal differences in the composition of toxins in M. edulis from Trow Rocks, south Tyneside, north-east England

Preliminary comparative results, obtained from the HPLC and the mouse bioassay for scallops and lobsters, are shown in Figures 8(a-b). Concentrations of toxins (mu) in the crustaceans were generally low, but values determined by HPLC were markedly higher. It is clear from the limited data available, either that the toxins in the animals are sequestered in some way and

are not toxic to mice, or that some of the peaks identified in the chromatograms are not in fact toxins; the latter explanation seems more likely.

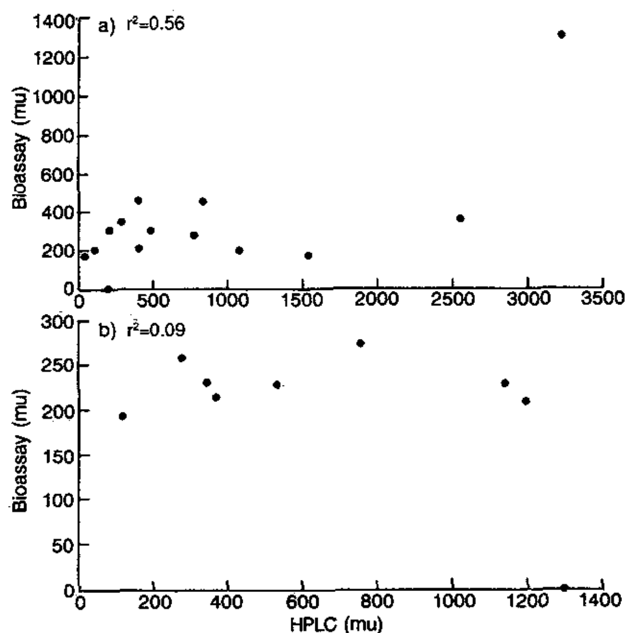


Figure 8. A comparison of toxicities of (a) scallops and (b) lobster by mouse assay and HPLC

Figures 9(a-d) show the relationship between the toxin peaks C1/C2 and GTX2 and the results of the mouse bioassay for mussels and lobsters. The concentration of GTX2 increases with increasing toxicity in both mussels and lobsters, whereas C1/C2 concentrations are largely independent of toxicity in lobsters. The result could be explained if a non-toxic fluorescent compound co-eluted with the C toxins, and indicates that further development of the method would be required before the HPLC technique could be used reliably for species other than mussels.

1.6.4 Discussion

The HPLC technique currently allows analysis of up to 20 samples per day for each HPLC unit, plus associated work on AQC materials, including standards and blanks. Although the autosampler is run overnight, a higher throughput of samples cannot be achieved because of the large amount of time required to maintain the analyser at optimal working efficiency. Three HPLC units would have been required to cope with the number of samples needing analysis during a bloom of the size seen in the 1990 event.

The results of HPLC analyses of mussel tissues broadly agree with results generated by mouse bioassay, and the technique provides specific information concerning the toxins present. However, the technique still suffers from the absence of standards for all plausible toxins and possibly from minor technical demands of the analytical approach. The most obvious problem, however, is the chemistry of the toxins in tissues and during extraction.

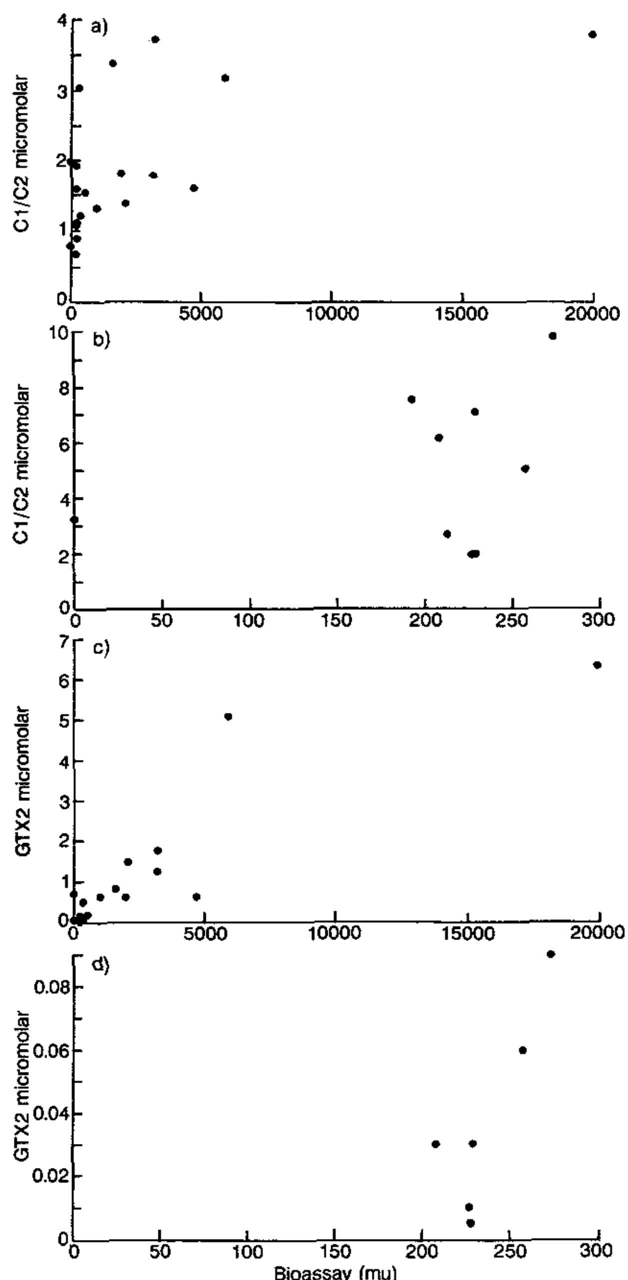


Figure 9. C1/C2 and GTX2 concentrations plotted against results of mouse assays for mussels (a) and (b), and for lobsters (c) and (d)

1.7 Chemical analysis of DSP toxins

1.7.1 Introduction

Although diarrhetic shellfish poisoning (DSP) is not recorded specifically in England and Wales, it seems likely that it does occur in a wild form. Methods for the detection of DSP toxin were therefore evaluated alongside the PSP toxin.

1.7.2 Materials and methods

The method for analysis of DSP toxins is essentially the same as that described by Lee *et al.* (1987). Shellfish tissues were homogenised in methanol and water, washed with hexane and extracted with chloroform. The extracts were dried and the toxins derivatised with 9-anthryldiazomethane. The fluorescent derivatives were then dissolved in hexane/chloroform, cleaned up on a silica 'sep-pak' column and introduced to the C18 HPLC column in methanol. Samples were run isocratically in 70% acetonitrile, 25% water and 5% methanol. Fluorescence was detected at an exitation wavelength of 365 nm and at an emission wavelength of 412 nm.

1.7.3 Results

Mussel samples were collected from eastern England during late June 1989. All samples proved negative. The exercise will be repeated for sites in later surveys.

1.8 Investigation of other algal blooms

Reports of extensive mortalities of marine animals, principally bivalves and lugworm on the coast of south-west Wales between Fishguard and Swansea, were investigated. The mortalities occurred between late August and mid-September, at first in the western area around the Pembroke peninsula and subsequently eastward reaching the Burry Inlet and Swansea Bay in mid-September. These mortalities were reported to be associated with the presence of a markedly visible algal bloom in the water which was identified by the National Rivers Authority , Wales as including *Gymnodinium* spp. at up to 10 000 cells ml^{-1}.

Samples of mussels were collected from Solva, Fishguard Harbour, Dale (Milford Haven) and Llanelli and tested for the presence of neurotoxins by the mouse method, no evidence of PSP was found.

Subsequently, mortalities extended eastwards into Swansea Bay and reports were received (Olive, pers. comm.) of large numbers of lugworm (*Arenicola marina*) behaving aberrantly and dying on the mud surface, together with extensive mortalities of cockles (*Cerastoderma edule*). These mortalities were initially attributed to heat prostration, post-reproductive die-back and possible anoxia.

Further samples consisting of cockles and lugworm from Llanelli and Swansea Bay were collected and tested for PSP with negative results.

The simple explanation for these mortalities of benthic organisms is that they derive from anoxia following collapse of a non-toxic algal bloom.

MARINE FISH DISEASES MONITORING PROGRAMME

2. THE MONITORING PROGRAMME FOR DETERMINATION AND SURVEILLANCE OF DISEASE IN MARINE FISH

2.1 Historical background

Through its marine fisheries laboratories and Fisheries Inspectorate offices, MAFF maintains *ad hoc* surveillance of diseases and anomalies in fish stocks and landed fish. Systematic investigations into the status of fish diseases and their possible relationships with contaminants in the marine environment have been undertaken by staff of the Weymouth Laboratory for the past 10 years (Bucke and Feist, 1990). The first dedicated marine fish disease investigation by DFR staff was conducted in 1972 (Shelton and Wilson, 1973) and, in common with all the early studies, was broadly based involving several fish species and a wide variety of disease conditions and parasites (Bucke *et al.*, 1983). Many of the early studies concentrated on waste disposal sites in the Thames Estuary and Liverpool Bay (Bucke *et al.*, 1983(a), (b)). However, because the fish were sampled randomly and inconsistently, these early results were rather subjective and unsuitable for monitoring purposes. To overcome this problem, which was common to many of the studies conducted by other laboratories, both in the UK and abroad, standardised sampling and recording procedures have been adopted (ICES, 1989). [These procedures are internationally accepted and are used in the various surveys conducted collaboratively e.g. under the auspices of the International Council for the Exploration of the Sea (ICES) and as a component of the North Sea Task Force Monitoring Master Plan; (NSTF, 1990)].

Consequently, the approach taken by FDL in the past few years has been to integrate their monitoring plans with existing DFR cruise programmes, e.g. annual groundfish surveys and those around waste disposal sites. The disease results from these cruises have been regularly presented via reports and papers to ICES and other bodies (Bucke *et al.*, 1983(a), (b); Bucke and Feist, 1984; Bucke and Nicholson, 1987; Brander *et al.*, 1988; Bucke and Stokes, 1989; Bucke and Feist, 1990) and in MAFF's own Aquatic Environment Monitoring Reports (MAFF, 1990, 1991).

2.2 Materials and methods

In 1990, data were collected from three field studies made on board DFR's research ships, and the results from a routine survey of fish landed by one of MAFF's District Fisheries Inspectorate Offices, are also included. The results from the field studies will also be used to complement the UK North Sea Task Force Monitoring Master Plan 1990 (NSTF, 1990).

2.2.1 *Areas monitored*

Three areas were monitored during 1990 (Figure 10):

(i) the Dogger Bank and waste disposal areas of the north-east coast (CORYSTES Cruise 6a/ 90), 5-11 May, 1990) (Figure 11);

(ii) the North Sea annual groundfish area (see Figure 13) (CIROLANA Cruise 8/90, 15 August-17 September, 1990); and

(iii) the Channel and south-west North Sea (Figure 10) (CORYSTES Cruise 11/90, 18-22 October, 1990).

2.2.2 *Sampling methods*

The recommended programme for marine fish diseases (ICES, 1989) was followed as closely as possible. Using a standardised Granton trawl, fitted with a small mesh cod-end liner, a minimum of 1 x 1 h and up to 5 x 1 h tows, were made on each station. The dab (*Limanda limanda*), was the fish species targetted for specific disease investigations; other commercial fish species were also examined, but less intensively. The aim was to examine, where possible, 100 dab 15-19 cm in length (GpI); and 100 of 20-24 cm in length (GpII), and 50 >25 cm in length (GpIII), from the whole sample or sub-sample in each haul. In the case of groundfish surveys, only one haul per station was possible.

To obtain the above samples, the trawl catches were sorted and the dab were sampled or randomly sub-sampled where large catches occurred. The samples of dab were weighed, individually measured for length

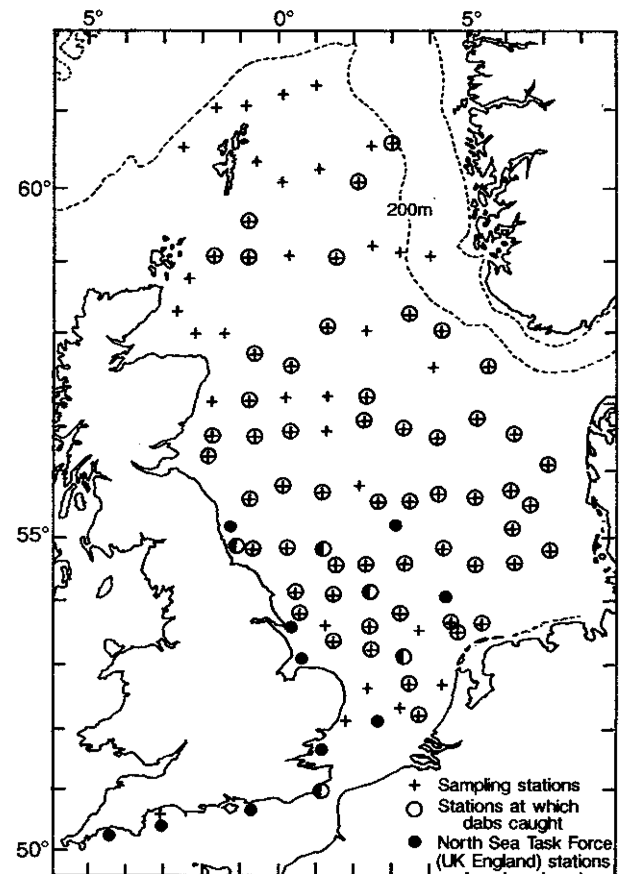


Figure 10. Stations sampled for fish diseases during routine monitoring on MAFF cruises in the North Sea and English Channel during 1990

and distributed into the size-length groups for visual examination for signs of disease. Otoliths were sampled at each station (2 per sex per centimetre length-size throughout the three groups) to assess the age-structure of the populations. Station numbers and co-ordinates were recorded.

For the groundfish survey, (CIROLANA 8/90) additional data for dab, including total catch numbers, weight, condition factor (all by sex), as well as station depth, trawl shooting and hauling co-ordinates, were included in the results.

2.2.3 *Recording of gross pathological changes in dab*

Lymphocystis, skin ulcerations, epidermal hyperplasia/ papilloma, as well as other anomalies involving abnormal pigmentation and lipoma, were recorded. Additionally, from GpIII fish, livers were examined for gross changes, including the presence of liver nodules >2 mm in diameter. If less than 50 dab >25 cm were available, the size-length group below were sampled until sufficient numbers had been examined. On the groundfish survey, all livers from fish >20 cm were examined for gross changes.

2.2.4 *Organs sampled for histological examination and assessment of stress in dab*

For confirmatory diagnosis, tissues and organs (including livers from dab) exhibiting gross lesions were sampled from each station and processed for histological examination. For comparative measurements of stress in dab, spleens from 10 fish were sampled from each of 13 selected stations in the southern North Sea. Perls' Prussian blue reaction was applied to histological sections of these spleens to demonstrate the presence of haemosiderin - a breakdown product of blood cells, which is increased in conditions of stress. Using an 'Olympus Cue-3' Image Analyser, the amount of haemosiderin in each section of spleen was quantitatively measured.

2.2.5 *Recording of gross pathological changes in other fish species*

Although the dab was the principal 'target' species for monitoring of fish diseases, less intensive examination of other commercial fish species in the catch was made as a matter of routine (when the opportunity arose). Records included the presence of skin ulcers, skeletal anomalies and other gross lesions from a number of fish species, including cod, haddock, plaice and whiting, when they were available. Histological examinations were made from tissues of individual fish which showed external signs of disease, in order to confirm or refute diagnosis. In other cases, histology was performed simply for research purposes.

2.2.6 *Fish disease returns from MAFF's Fisheries Inspectorate*

District Inspectorate Officers make random inspections of fish landed at ports and markets and unusual examples of fish diseases or anomalies are reported to FDL. If these cases are thought to be significant, arrangements are made for regular returns to be submitted to FDL for overall assessment and possible future evaluation in the field.

2.3 Results

2.3.1 *RV CORYSTES Cruise 6a, 5-11 May 1990*

Figure 11 shows the sampling sites and Figure 12 the mean length of dabs assessed by sex and by area, respectively. The inshore areas 3 and 4 (stations 79-81 and 90-94, respectively) are the waste disposal sites, and 1 and 2 (stations 58-61 and 67-70, respectively) the control areas. Overall disease results are shown in Table 6. No sample was obtained from area 3. Catches of dab in the other areas were poor and sufficient samples were only obtained by pooling catches from several hauls. Disease prevalence rates between areas 1 and 2 were similar, with the exception of epidermal hyperplasia/papilloma and ulcer prevalence in GpI fish (Table 6). Direct comparison with area 4 was difficult as the number of fish examined was reduced, especially GpIII fish. The length distribution data appeared to indicate that both male and female dab caught off the River Tees were faster growing and typically larger than dab from the Dogger Bank stations (areas 1 and 2) (Figure 11). The overall results are similar to those from previous years (MAFF, 1991).

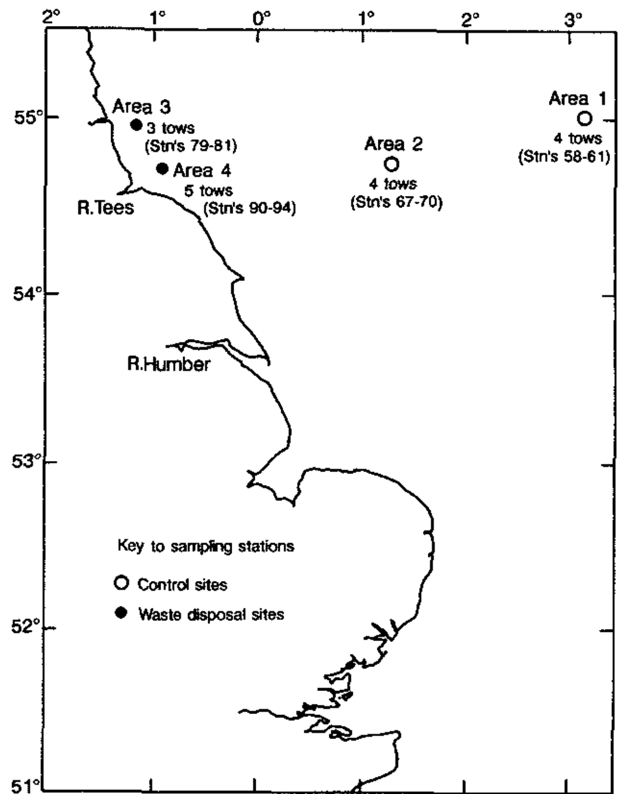


Figure 11. ***Stations sampled for diseases of dabs during routine monitoring on the north-east coast of England in May 1990***

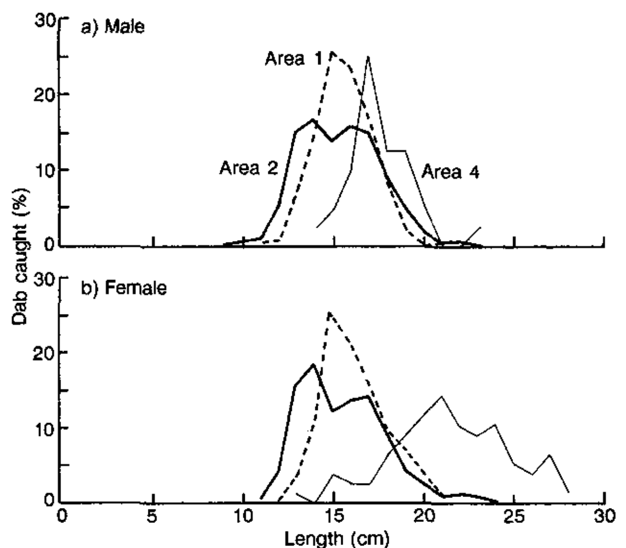


Figure 12. ***Length distribution of dabs in Areas 1-4 (see Figure 11): (a) males and (b) females***

Table 6. Diseases in dab from the north - east coast of England, May 1990 (see Figure 11)

Station number	ICES rectangle	n	Group	M	F	Disease			L/N	% Value	Overall epidermal disease % value	Overall liver nodule % value
						L	H	U				
58-61	39F3	245	1	51	49	3	5	0	-	8	10	13.3
			2	24	76	9	2	10	-	21		
			3	3	42	1	0	4	6	22.2		
67-70	38F1	249	1	55	45	1	0	10	-	11	4	14.2
			2	35	65	2	0	6	-	8		
			3	0	49	1	1	5	7	28.5		
79-81	39E8	n/s	n/s	n/s	n/s	n/s	n/s	n/s	n/s	n/s	n/s	n/s
90-94		148	1	44	23	7	2	1	-	14.9	16.2	5.6
			2	11	52	11	1	7	-	30.2		
			3	0	18	0	1	2	1	n/a		

Key: n/s=no sample; n/a=not applicable; n=total number examined; M=male; F=female; L=lymphocystis; H=epidermal hyperplasia/papilloma; U=skin ulcers; L/N=liver nodules>2mm diameter

2.3.2 RV CIROLANA Cruise 8, 15 August-17 September, 1990

On the 1990 annual groundfish survey it was possible to assess the prevalence of disease in over 10 500 dab sampled from 63 stations, covering much of the North Sea (Figure 13). The catch data for dab are shown in Table 7. It was only practical to make a one-hour tow on each station. Thus, it was not always possible to obtain the minimum sample recommended by ICES (ICES, 1989). Results shown in Table 8 indicate close similarities with monitoring results from previous years (MAFF, 1991). The highest prevalence of disease in dab was again recorded at stations off the Firth of Forth and on the Dogger Bank (Figure 14).

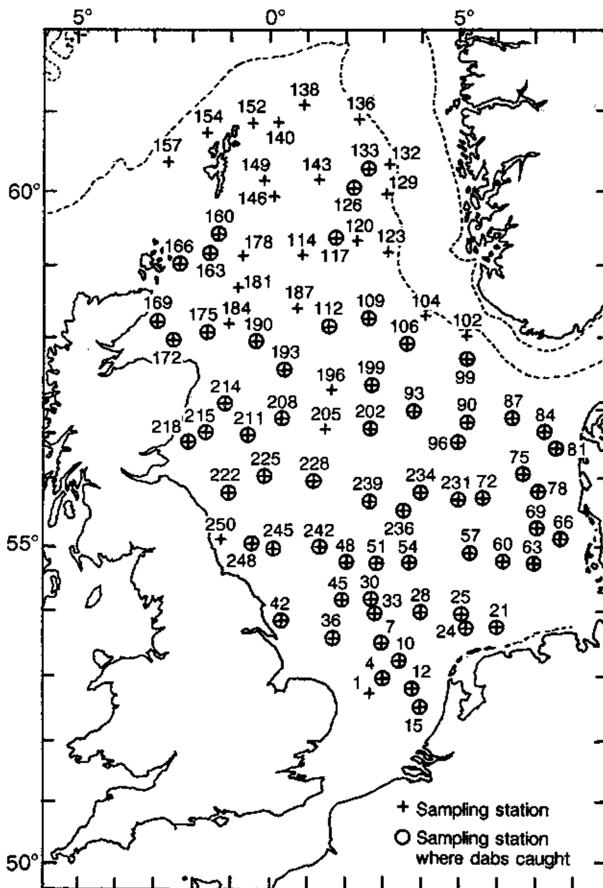


Figure 13. **Stations sampled for diseases of dabs on the North Sea annual groundfish survey**

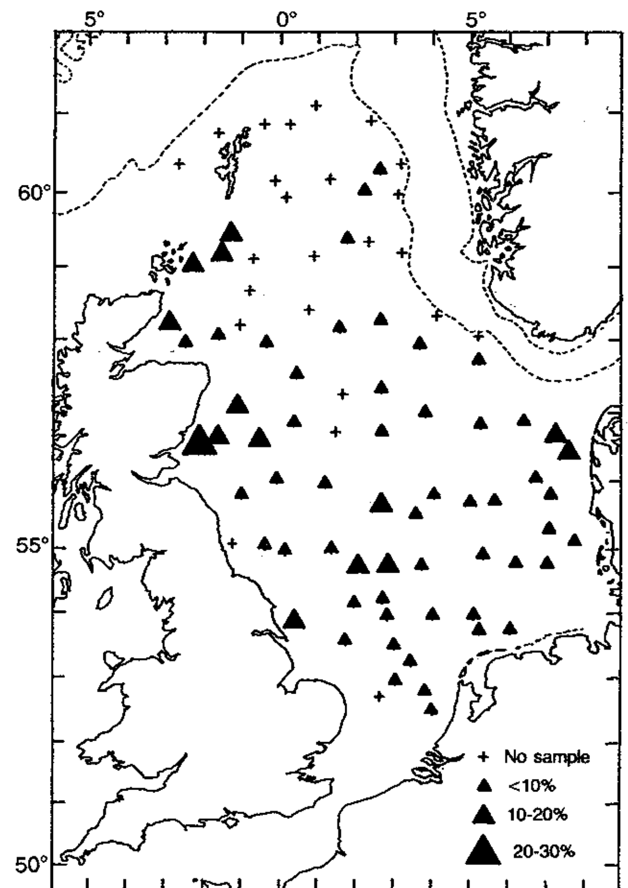


Figure 14 **Overall external disease prevalence (% values) sampled from the North Sea. Diseases included lymphocystis, epidermal ulcers and epidermal hyperplasia/papilloma**

Table 7. Catch data for dabs sampled for disease monitoring in the North Sea groundfish survey in August - September 1990 (see Figure 14)

Station number	ICES rectangle	Latitude (N)	Longitude	Depth (m)		Total catch		Total weight (kg)		Condition factor		1st length size of dab in each trawl (cm)		Last length size of dab in each trawl (cm)	
				Shot	Haul	M	F	M	F	M	F	M	F	M	F
4	34F2	52.50.9	2.43.1E	37	33	327	443	17.19	28.10	0.0097	0.0100	4.0	11.0	26.0	32.0
7	35F2	53.24.2	2.41.0E	30	30	2062	1505	64.23	54.75	0.0100	0.0100	3.0	4.0	28.0	36.0
10	35F3	53.6.7	3.7.5E	30	33	389	427	19.27	25.53	0.0101	0.0101	3.0	4.0	28.0	31.0
12	34F3	52.43.4	3.24.9E	28	28	201	273	7.04	12.14	0.0098	0.0096	10.0	11.0	25.0	30.0
15	33F3	52.25.2	3.36.4E	29	29	71	77	3.75	3.03	0.0094	0.0103	9.0	10.0	25.0	29.0
21	36F5	53.41.1	5.22.4E	31	31	1094	1121	58.81	61.65	0.0102	0.0094	6.0	6.0	24.0	31.0
24	36F4	53.37.0	4.40.9E	29	34	2642	2273	84.97	84.90	0.0096	0.0102	4.0	5.0	27.0	33.0
25	36F4	53.48.9	4.33.5E	40	40	2304	2123	116.03	114.88	0.0102	0.0100	10.0	10.0	25.0	30.0
28	36F3	53.52.0	3.36.9E	33	41	626	605	25.60	33.60	0.0088	0.0102	11.0	7.0	30.0	29.0
30	37F2	54.4.4	2.27.2E	71	67	152	277	6.83	12.84	0.0095	0.0101	5.0	12.0	20.0	21.0
33	36F1	53.50.4	2.32.9E	40	33	284	1249	20.95	128.60	0.0096	0.0099	5.0	15.0	30.0	36.0
36	35F1	53.26.2	1.34.1E	27	26	147	290	5.79	18.77	0.0097	0.0100	4.0	6.0	24.0	33.0
42	36F1	53.58.4	0.44.6E	51	51	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S
45	37F1	54.2.7	1.46.7E	75	45	568	905	34.57	68.73	0.0096	0.0096	3.0	4.0	24.0	31.0
48	38F1	54.38.2	1.52.2E	25	27	1016	1026	44.41	81.08	0.0092	0.0102	5.0	12.0	26.0	31.0
51	38F2	54.36.0	2.33.9E	22	21	977	351	75.66	21.58	0.0106	0.0105	6.0	12.0	26.0	31.0
54	38F3	54.38.6	3.20.2E	40	40	1793	2456	109.04	133.14	0.0097	0.0095	5.0	13.0	21.0	28.0
57	38F4	54.45.3	4.47.3E	45	43	958	913	40.32	41.95	0.0097	0.0095	10.0	10.0	22.0	28.0
60	38F5	54.38.0	5.33.2E	43	43	2037	1597	97.69	84.80	0.0101	0.0101	3.0	4.0	24.0	27.0
63	38F6	54.34.5	6.21.1E	40	39	1742	2982	64.92	110.42	0.0097	0.0095	4.0	4.0	24.0	30.0
66	38F7	54.56.9	7.11.6E	29	32	2626	1152	191.52	79.38	0.0105	0.0098	4.0	5.0	26.0	31.0
69	39F6	55.4.4	6.35.6E	41	45	624	540	18.50	28.21	0.0100	0.0103	3.0	4.0	27.0	29.0
72	40F5	55.33.8	5.4.2E	45	45	2094	1715	86.17	80.86	0.0098	0.0099	10.0	10.0	21.0	23.0
75	40F6	55.55.3	6.1.6E	44	47	3059	3534	131.10	154.75	0.0092	0.0086	9.0	9.0	23.0	25.0
78	40F6	55.38.6	6.42.7E	36	35	2127	2152	67.50	95.50	0.0097	0.0086	4.0	5.0	25.0	33.0
81	41F7	56.17.7	7.7.8E	33	33	2460	2216	69.97	88.33	0.0098	0.0102	4.0	4.0	26.0	30.0
84	42F6	56.35.5	6.54.0E	36	38	379	593	16.97	37.66	0.0096	0.0095	8.0	8.0	27.0	28.0
87	42F5	56.47.2	5.45.7E	55	53	570	907	27.56	57.69	0.0101	0.0095	10.0	11.0	27.0	28.0
90	42F4	56.43.6	4.44.0E	60	59	772	1062	25.29	41.69	0.0090	0.0090	7.0	7.0	19.0	29.0
93	42F3	56.53.6	3.28.9E	69	66	994	899	35.33	43.56	0.0096	0.0094	10.0	10.0	27.0	25.0
96	42F4	56.49.4	4.6.1E	55	55	925	932	47.60	38.79	0.0101	0.0087	8.0	10.0	19.0	21.0
99	44F4	57.43.8	4.41.1E	92	100	49	80	1.71	3.73	0.0086	0.0083	10.0	14.0	19.0	23.0
106	44F3	57.56.7	3.15.6E	79	69	492	325	15.20	11.02	0.0088	0.0085	12.0	12.0	24.0	24.0
109	45F2	58.18.4	2.25.2E	73	77	433	793	14.35	22.60	0.0089	0.0092	12.0	13.0	17.0	18.0
112	45F1	58.12.5	1.27.3E	108	108	42	30	2.65	1.32	0.0093	0.0091	14.0	16.0	18.0	22.0
117	47F1	59.28.4	1.38.9E	120	123	32	33	1.35	2.04	0.0089	0.0089	9.0	10.0	20.0	22.0
126	49F2	60.14.3	2.5.9E	105	115	88	54	2.31	4.90	0.0087	0.0094	10.0	9.0	23.0	29.0
133	50F2	60.32.0	2.40.7E	110	113	54	84	2.55	5.51	0.0094	0.0093	10.0	12.0	22.0	24.0
160	48E8	59.33.0	1.4.1W	127	117	49	43	3.14	3.68	0.0092	0.0093	15.0	16.0	22.0	28.0
163	47E8	59.15.7	1.13.9W	114	114	26	14	2.26	0.78	0.0160	0.0089	15.0	16.0	20.0	21.0
166	47E8	59.5.6	1.54.3W	77	70	20	31	0.99	2.20	0.0098	0.0092	14.0	15.0	20.0	28.0
169	45E7	58.17.3	2.50.2W	49	48	958	1381	39.06	51.11	0.0094	0.0093	11.0	10.0	21.0	27.0
172	45E7	58.17.9	2.31.0W	75	70	33	49	1.16	2.69	0.0092	0.0090	13.0	13.0	17.0	24.0
175	45E8	58.4.6	1.15.6W	111	108	52	57	3.56	2.39	0.0089	0.0095	14.0	14.0	19.0	28.0
190	44E9	57.55.7	0.9.4W	100	101	106	119	5.41	5.05	0.0089	0.0089	13.0	13.0	19.0	22.0
193	44F0	57.32.7	0.29.3W	88	86	9	22	0.51	0.79	0.0088	0.0089	14.0	16.0	17.0	23.0
199	43F2	57.17.3	2.27.5E	80	81	162	283	6.56	13.5	0.0093	0.0090	14.0	13.0	19.0	22.0
202	42F2	56.39.4	2.27.1E	73	75	530	317	20.92	10.51	0.0089	0.0089	13.0	12.0	18.0	20.0
208	42F0	56.47.0	0.22.6E	83	79	190	253	8.77	9.52	0.0085	0.0085	14.0	13.0	18.0	24.0
211	42E9	56.32.7	0.22.2W	73	76	966	698	51.44	38.3	0.0096	0.0094	15.0	14.0	22.0	26.0
214	42E9	56.59.9	0.55.9W	73	74	2123	417	11.91	31.58	0.0084	0.0088	14.0	15.0	23.0	34.0
215	42E8	56.34.8	1.21.6W	70	63	803	929	53.8	54.08	0.0095	0.0094	14.0	15.0	27.0	30.0
218	41E8	56.27.0	1.44.9W	49	43	1296	1485	143.09	262.2	0.0103	0.0100	11.0	10.0	32.0	39.0
222	40E9	55.40.4	0.48.0W	85	93	408	231	23.35	15.11	0.0093	0.0090	14.0	15.0	25.0	24.0
225	40F0	55.55.2	0.1.6E	90	85	152	119	8.39	5.6	0.0088	0.0086	13.0	14.0	21.0	22.0
228	40F1	55.50.4	1.8.0E	76	84	447	929	17.79	32.63	0.0090	0.0089	13.0	13.0	19.0	24.0
231	40F0	55.32.9	4.31.1E	32	33	218	695	9.74	50.51	0.0101	0.0102	11.0	11.0	24.0	31.0
234	40F3	55.39.8	3.35.6E	45	44	2061	1710	99.28	90.41	0.0098	0.0091	14.0	14.0	25.0	31.0
236	39F3	55.24.7	3.13.7E	30	30	2766	2122	128.83	121.38	0.0098	0.0093	14.0	14.0	29.0	33.0
239	40F2	55.31.5	2.24.6E	55	68	1875	1345	74.12	59.93	0.0087	0.0088	14.0	14.0	24.0	26.0
242	38F1	54.52.0	1.16.6E	36	37	3781	1614	156.29	64.9	0.0091	0.0088	7.0	6.0	23.0	28.0
245	38F0	54.48.0	0.11.8E	77	80	154	312	6.1	14.39	0.0088	0.0084	13.0	13.0	21.0	24.0
248	38E9	54.54.8	0.18.1W	73	76	247	188	9.64	8.59	0.0092	0.0093	12.0	11.0	20.0	23.0

KEY: N/S = No sample; M = Male; F = Female

Table 8. Diseases sampled in dab in the North Sea groundfish survey in August-September 1990

Station number	ICES rectangle	n	Group	M	F	Diseases			L/N	% Value	Overall epidermal disease % value	Overall liver nodule % value
						L	H	U				
4	34F2	188	1	56	70	0	0	2	-	1.6		
			2	11	28	0	0	2	0	5.1	5.3	0
			3	1	22	1	1	4	0	n/a		
7	35F2	105	1	50	44	0	0	1	-	1.1		
			2	2	4	0	0	0	0	0	0.95	0
			3	4	1	0	0	0	0	0		
10	35F3	187	1	53	54	0	0	-	0.9	0.9		
			2	25	31	0	0	0	5.4	5.4	6.4	0
			3	5	19	1	1	6	0	n/a		
12	34F3	154	1	57	74	0	0	0	-	0		
			2	1	17	0	0	0	0	0	0	0
			3	1	4	0	0	0	0	0		
15	33F3	74	1	39	20	0	0	-	1.7	1.7		
			2	4	4	0	0	0	0	0	1.4	0
			3	1	6	0	0	0	0	0		
21	35F5	195	1	74	55	0	0	-	0	0		
			2	16	35	0	0	1	1	4	1.5	7.6
			3	0	15	2	0	0	4	n/a		
24	36F4	185	1	60	53	0	0	0	-	0		
			2	6	30	0	0	0	2	5.6	1.08	6.9
			3	2	34	0	0	2	3	13.9		
25	36F4	205	1	76	61	0	0	0	-	0		
			2	6	41	0	0	0	0	0	0.5	1.5
			3	1	20	0	0	1	1	n/a		
28	36F3	206	1	76	71	2	0	2	-	2.6		
			2	10	28	0	2	2	2	15.9	5.3	7.1
			3	2	16	0	0	3	3	n/a		
30	37F2	158	1	94	58	4	0	3	-	4.6		
			2	3	3	1	0	1	0	n/a	5.7	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
33	36F2	356	1	73	9	5	1	3	-	9		
			2	46	6	0	1	0	2	5.4	7	3.9
			3	6	195	3	2	10	8	11.5		
36	35F1	183	1	59	61	0	0	2	-	1.7		
			2	2	39	0	1	1	0	4.8	4.3	0
			3	0	22	0	0	4	0	n/a		
42	36F0	231	1	68	32	2	2	8	-	12		
			2	40	60	4	3	9	1	17	15.6	3.1
			3	0	31	1	2	5	3	35.5		
45	37F1	289	1	63	75	0	0	4	-	2.9		
			2	32	87	2	0	4	1	5.9	5.2	1.9
			3	0	32	4	0	1	2	21.8		
48	38F1	246	1	60	53	3	0	2	-	4.4		
			2	6	45	2	0	6	1	17.7	9.3	9.8
			3	3	79	2	0	8	13	27.6		
51	38F2	243	1	37	93	1	0	6	-	5.4		
			2	9	54	3	2	4	6	23.8	8.6	13.2
			3	1	49	0	1	4	9	28		
54	38F3	174	1	88	59	2	0	5	-	4.8		
			2	8	14	1	0	1	0	9	5.7	3.7
			3	0	5	0	0	1	1	n/a		
57	38F4	174	1	83	63	2	1	2	-	3.4		
			2	3	24	1	0	2	1	14.8	4.6	7.4
			3	0	1	0	0	0	0	0		
60	38F5	199	1	89	65	4	0	0	-	2.6		
			2	8	29	0	1	1	0	5.4	4	0
			3	0	8	2	0	0	0	n/a		
63	38F6	171	1	92	52	2	0	1	-	2.1		
			2	10	12	0	0	0	2	9.1	1.8	8.3
			3	0	5	0	0	0	1	n/a		
66	38F7	276	1	36	60	0	0	4	-	4.2		
			2	26	92	5	0	6	4	12.7	7.6	3.3
			3	5	57	2	2	2	2	12.8		
69	39F6	146	1	48	53	3	0	1	-	4		
			2	8	24	0	0	0	1	3.1	3.4	2.2
			3	1	12	0	0	1	0	n/a		

Table 8. Continued

Station number	ICES rectangle	n	Group	M	F	Diseases			L/N	% Value	Overall epidermal disease % value	Overall liver nodule % value
						L	H	U				
72	40F5	165	1	84	73	4	2	1	-	4.2		
			2	2	6	2	0	0	0	n/a	5.5	0
			3	0	0	n/s	n/s	n/s	n/s	n/s		
75	40F6	214	1	73	72	6	1	2	-	6.2		
			2	10	56	2	1	2	1	9	6.5	1.5
			3	0	3	0	0	0	0	0		
78	40F6	251	1	63	51	5	1	0	-	5.3		
			2	11	77	4	0	3	1	9	7.6	0.7
			3	2	47	1	1	4	0	12.2		
81	41F7	267	1	54	29	0	2	3	-	6		
			2	21	89	2	1	8	0	10	12.3	0
			3	5	69	3	0	14	0	23		
84	42F6	188	1	30	53	3	1	6	-	12		
			2	19	55	6	0	3	0	12.2	11.1	0.9
			3	5	26	1	0	1	1	9.6		
87	42F5	231	1	59	78	2	0	0	-	1.6		
			2	8	58	4	1	3	0	12.1	4.3	0
			3	2	26	0	0	0	0	0		
90	42F4	170	1	60	95	3	0	1	-	2.3		
			2	0	12	2	0	0	0	n/a	3.5	0
			3	0	3	0	0	0	0	0		
93	42F3	182	1	73	85	6	0	1	-	4.2		
			2	1	18	0	0	1	0	5.3	4.9	0
			3	1	4	1	0	0	0	n/a		
96	42F4	157	1	53	44	3	0	1	-	4.1		
			2	4	30	1	1	1	0	8.7	6.4	0
			3	0	26	2	0	1	0	11.5		
99	44F4	107	1	37	63	1	0	0	-	1		
			2	0	7	0	0	0	0	0	0.9	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
106	44F3	96	1	37	55	4	0	1	-	5.4		
			2	1	3	0	0	0	0	0	5.2	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
109	45F2	67	1	22	45	2	2	0	-	6		
			2	0	0	n/s	n/s	n/s	n/s	n/s	6	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
112	45F1	67	1	28	30	1	0	0	-	1.7		
			2	0	9	0	0	0	0	0	1.5	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
117	47F1	90	1	25	54	4	0	0	-	5.1		
			2	2	7	0	0	0	0	0	4.4	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
126	49F2	108	1	30	58	1	0	0	-	1.1		
			2	5	12	2	0	0	0	n/a	2.8	n/s
			3	0	3	0	0	0	0	0		
133	50F2	102	1	39	57	1	0	0	-	1		
			2	4	2	3	0	0	0	n/a	3.3	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
160	48E8	92	1	36	17	2	0	0	-	3.8		
			2	13	24	6	0	2	0	21.6	10.8	0
			3	0	2	n/s	n/s	n/s	n/s	n/s		
163	47E8	40	1	25	11	0	2	1	-	8.4		
			2	1	3	0	0	0	0	0	7.5	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
166	47E8	50	1	18	22	1	1	0	-	5		
			2	1	7	0	0	0	0	0	4	n/s
			3	0	2	n/s	n/s	n/s	n/s	n/s		
169	47E7	124	1	53	40	2	3	4	-	9.9		
			2	3	17	1	0	2	2	n/a	12.9	9.7
			3	0	11	2	1	1	1	n/a		
172	45E7	66	1	21	39	0	0	0	-	0		
			2	0	6	0	0	0	0	0	0	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
175	45E8	102	1	51	40	2	1	0	-	3.3		
			2	0	9	0	0	0	0	0	2.9	n/s
			3	0	2	0	0	0	0	0		

Table 8. Continued

Station number	ICES rectangle	n	Group	M	F	Diseases L	H	U	L/N	% Value	Overall epidermal disease % value	Overall liver nodule % value
190	44E9	218	1	113	101	10	5	2	-	7.9		
			2	0	4	0	0	0	0	0	7.8	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
193	44F0	28	1	19	8	2	0	0	-	7.4		
			2	0	1	0	0	0	0	0	7.1	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
199	43F2	161	1	52	100	8	1	0	-	6		
			2	0	9	0	0	0	0	0	5.6	0
			3	0	0	n/s	n/s	n/s	n/s	n/s		
202	42F2	134	1	42	90	5	0	0	-	3.8		
			2	0	2	0	0	1	0	n/a	4.5	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
208	42F0	153	1	83	66	10	0	0	-	6.7		
			2	0	4	0	0	0	0	0	6.5	n/s
			3	0	0	n/s	n/s	n/s	n/s	n/s		
211	42E9	162	1	84	56	7	1	7	-	10.7		
			2	7	14	1	0	1	1	n/a	10.5	4.8
			3	0	1	0	0	0	0	0		
214	42E9	218	1	47	71	7	0	7	-	11.8		
			2	14	63	4	1	6	2	16.9	13.8	2
			3	0	23	1	1	3	0	21.6		
215	42E8	234	1	86	64	8	0	3	-	5.3		
			2	21	46	2	2	3	1	12	8.1	3.2
			3	5	12	0	0	1	2	n/a		
218	42E8	383	1	32	25	3	0	10	-	22.8		
			2	61	42	5	1	18	0	23.4	36.9	4
			3	31	172	12	3	82	8	51.7		
222	40E9	191	1	97	45	3	0	3	-	4.2		
			2	17	31	1	2	0	2	10.4	4.7	4.1
			3	1	0	0	0	0	0	0		
225	40F0	127	1	53	58	4	2	2	-	7.2		
			2	2	14	1	0	0	1	n/a	7.1	n/a
			3	0	0	n/s	n/s	n/s	n/s	n/s		
228	40F1	137	1	87	48	3	1	1	-	3.6		
			2	0	2	0	1	0	0	n/a	4.4	n/a
			3	0	0	n/s	n/s	n/s	n/s	n/s		
231	40F4	174	1	23	76	3	0	2	-	5		
			2	7	68	2	2	5	1	13.4	8	1.3
			3	0	0	n/s	n/s	n/s	n/s	n/s		
234	40F3	230	1	93	68	8	1	2	-	6.8		
			2	6	36	0	2	1	0	6.6	7.8	0
			3	1	23	2	0	2	0	16.6		
236	39F3	272	1	64	57	3	0	2	0	5		
			2	15	80	5	0	3	-	12.7	8.5	2
			3	4	52	1	0	6	1	14.3		
239	40F2	150	1	47	74	7	1	1	-	7.4		
			2	4	22	0	0	3	0	11.5	10	0
			3	0	3	0	1	2	0	n/a		
242	38F1	142	1	53	63	5	1	3	-	7.9		
			2	6	13	0	0	0	1	n/a	6.3	n/a
			3	0	7	0	0	0	0	0		
245	38F0	154	1	48	95	7	1	0	-	5.5		
			2	0	11	2	0	0	1	n/a	6.4	n/a
			3	0	0	n/s	n/s	n/s	n/s	n/s		
248	38E9	122	1	53	57	0	0	5	-	5.4		
			2	3	9	0	1	0	0	n/a	5.7	n/a
			3	0	0	n/s	n/s	n/s	n/s	n/s		

KEY: n/s=no sample ;n/a=not applicable; n=total sampled; M=male; F=female; Group 1=15-19cm length; Group 2=20-24cm length; Group 3=>25cm length;L=lymphocystis; H=epidermal hyperplasia/papilloma; U=skin ulcers; L/N=liver nodules >2mm dia (Groups 2&3)

Disease anomalies, other than those specified by ICES, were also recorded. A 'green skin' pigmentation characteristic appeared to be present at stations in the southern North Sea and, at some stations, the prevalence was as high as 10.8% (Figure 15). The cause of this condition is unknown, but under investigation.

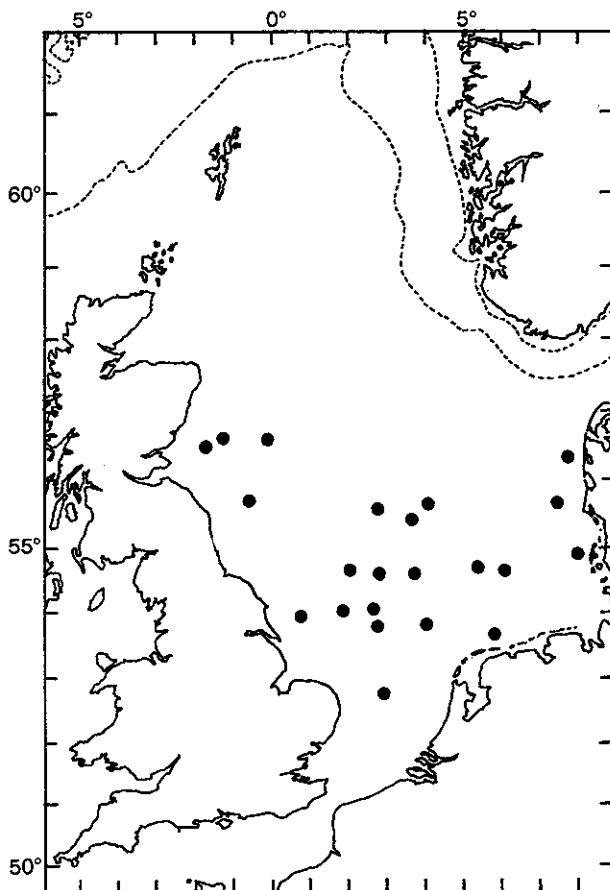


Figure 15 ***North Sea stations where cases of 'green-skin' pigmentation on dab were observed***

Orange-pigmentation of lesions in muscles of dab, from stations in the northern North Sea, was another interesting anomaly (Figure 16). Although the numbers of fish affected were insignificant, this condition, which is associated with inflammatory processes in the surrounding muscle tissues, has been tentatively ascribed to a lipoma (McVicar *et al.*, 1988). High levels of infestations of *Glugea stephani* (Microsporidea) were noted in dab sampled at stations off the Dutch coast (Figure 16), which confirmed earlier reports by van Banning (1987). In fish which were most seriously affected, the sporulating organisms had formed xenomas. Where these occurred in the intestines, occlusion of the luminal passage would probably lead to death of the fish.

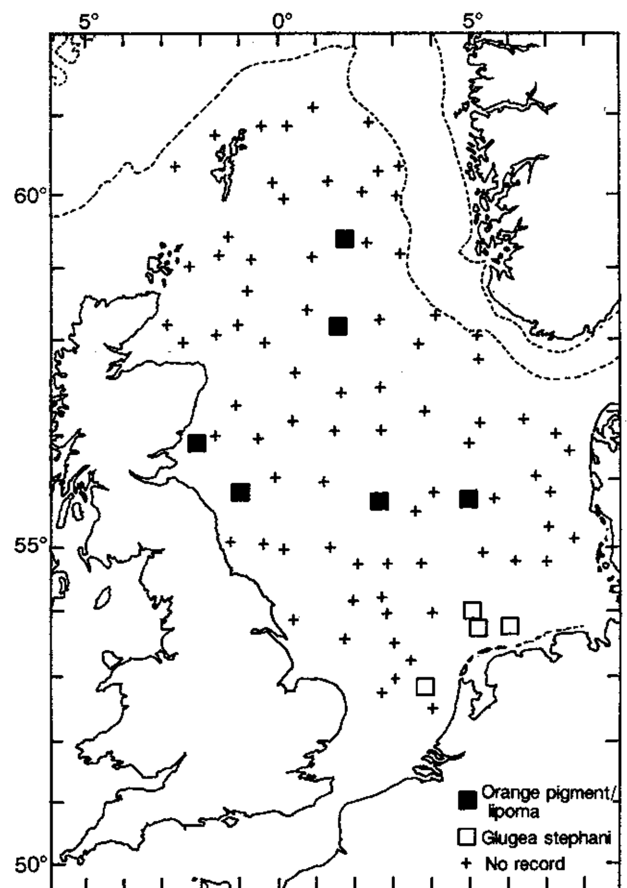


Figure 16. ***Stations where 'orange pigmented dermal lesions' and extensive infestations of the Microsporidean, Glugea stephani were observed in dabs during routine monitoring in the North Sea***

Figure 17 shows the distribution of accumulations of haemosiderin in splenic melano-macrophage centres in dab sampled at stations in the southern North Sea. From these quantitatively-assessed results, it appeared that accumulations of haemosiderin in splenic melano-macrophage centres were higher in dab sampled from Dogger Bank stations than in dab from other stations.

The prevalence of disease and physical anomalies in other fish species was low and was not significant for monitoring purposes, apart from the occurrence of vertebral compression in haddock (*Melanogrammus aeglefinus*) which was observed at stations where significant numbers of this species could be caught (Figure 18). Deformities in haddock are monitored more fully in SOAFD programmes (ICES, 1988).

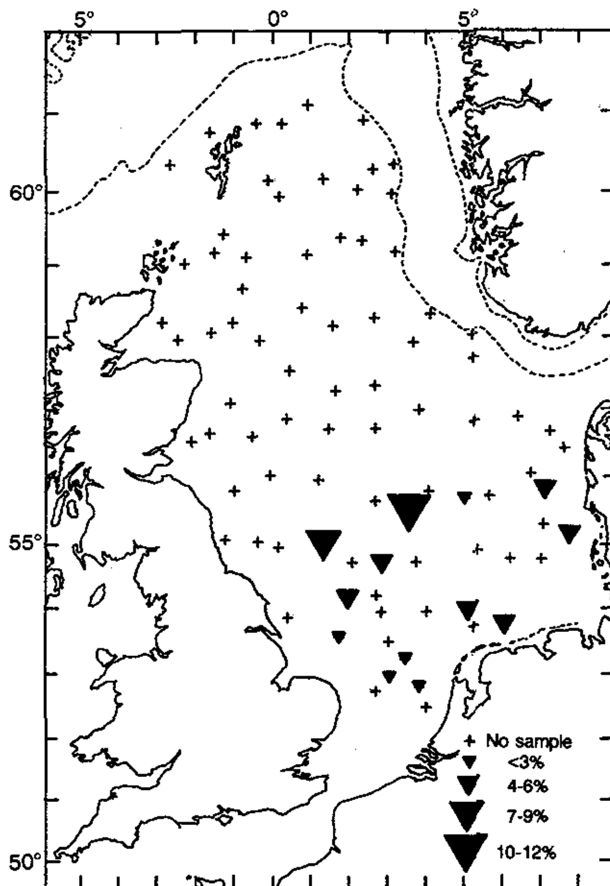


Figure 17. ***Stations sampled for assessment of stress in dabs during routine monitoring in the southern North Sea. The symbols represent mean levels of accumulation of haemosiderin in splenic melano-macrophage centres in dabs (10 per station) measured using image analysis quantification***

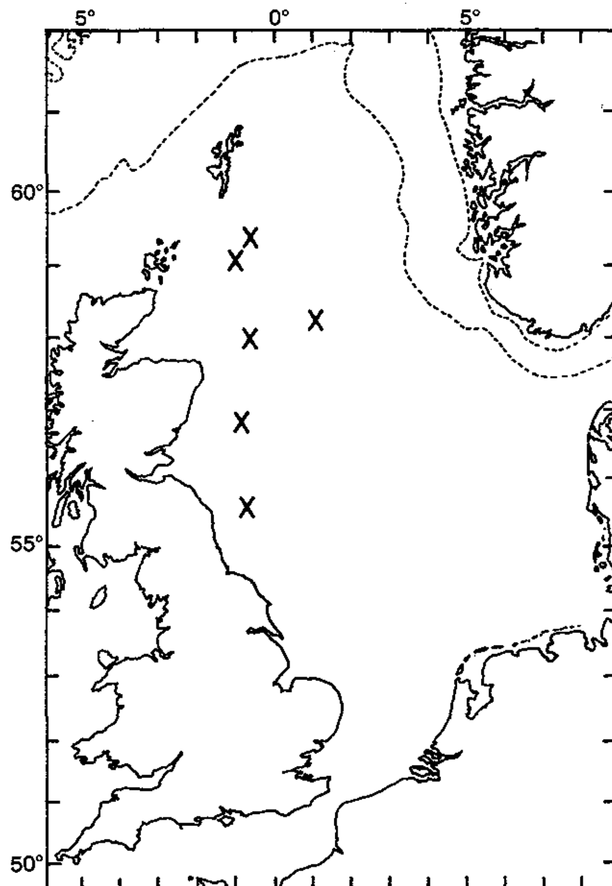


Figure 18. ***Stations where vertebral compressions were observed in haddock during routine monitoring in the North Sea***

2.3.3 RV CORYSTES Cruise 11, 18-22 October 1990

The English Channel and south-western North Sea are areas which have been recently included in the monitoring programme. Stations sampled were those recommended in the North Sea Task Force Monitoring Programme (NSTF, 1990) (Figure 10). Table 9 shows the catch data results for this cruise. Stations within ICES Rectangle 30F0 were the only ones to yield sufficient dab for monitoring purposes. Figure 19 shows the combined length-size profiles of dab from stations 68-71 (see Figure 10); age data were not available for this report. Table 10 shows the results of disease prevalence in dab from the above stations. Only 3 dab from the combined hauls from stations 68-71 were noted to have the 'green pigmented' syndrome. In comparison with results for stations in the North Sea, disease prevalence was low in ICES Rectangle 30F0.

Small samples of cod (*Gadus morhua*), comprising 48

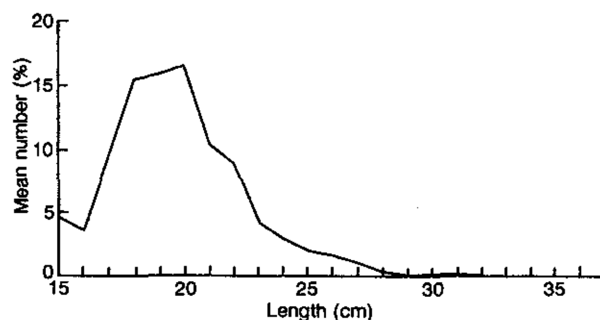


Figure 19. ***Mean length distribution of dab, sampled from 4 hauls from ICES Rectangle 30F0 (see Figure 10), during routine monitoring in the North Sea***

Table 9. Catch data for dabs sampled in the English Channel and S.W. North Sea in October 1990, (See Figure 10)

Station number	NSTF station	ICES rectangle	Depth (m)	Total catch	Total weight (kg)	Total weight (kg) Sub-sample	GP I No. (Unsexed)	Weight (kg)	GP II No. (Unsexed)	Weight (kg)	GP III No. (Unsexed)	Weight (kg)
54-56	73	30E9		N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S
63-67	72	29E9		N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S
68	69	30F0	15	846	233.0	116.5	195	14.0	147	16.3	21	4.8
69	69	30F0	17	801	209.0	69.8	162	10.8	90	10.1	15	4.3
70	69	30F0	22	753	214.0	71.4	113	8.3	123	13.0	19	4.5
71	69	30F0	20	861	216.0	72.0	138	9.4	137	14.8	17	4.0
72-77	19	32F1		N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S
78	25	33F2		N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S
79-81	-	33F1		N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S	N/S

N/S = No samples

Table 10. Disease in dabs sampled in the English Channel and S W North Sea in October 1990 (see Figure 10)

Station No	ICES rectangle	n	No. sampled in each group						Diseases														All epi-dermal disease % values	Total liver nodules % values
			GP I		GP II		GP III		GP I				GP II					GP III						
			M	F	M	F	M	F	L	H	U	% values	L	H	U	L/N	% values	L	H	U	L/N	% values		
54-56	30E9	N/S																						
63-67	29E8	N/S																						
68	30F0	201	93	29	59	41	7	13	0	0	0	0.0	3	1	0	1	6.5	0	0	1	0	5.0	2.5	2.0
69	30F0	207	68	32	42	47	1	17	1	0	0	1.0	0	0	0	1	3.1	0	1	1	0	11.0	1.4	2.0
70	30F0	219	82	18	59	41	7	12	1	0	1	2.0	0	0	0	0	0.0	1	0	0	0	5.2	1.4	0.0
71	30F0	219	79	21	63	37	1	18	0	1	0	1.0	1	0	1	0	2.0	0	0	0	0	0.0	1.4	0.0
72-77	32F1	N/S																						
78	33F2	N/S																						
79-81	33F1	N/S																						

Key: n=total sample examined; N/S=No Sample; M= Male; F= Female; L= Lymphocystes;H=Epidermal hyperplasia/papilloma; U=Epidermal ulcer; L/N= Liver nodule > 2 mm diameter

fish overall from all stations, were examined for diseases. Those seen included visceral granulomatosis and skin ulceration in ICES Rectangles 32F1, 33F1 (2 out of 2 from each site), and 33F2 (1 out of 10). All fish examined were >50 cm length size. The presence of visceral granulomatosis in cod has been reported previously in these areas. It is considered to be of infectious aetiology and unrelated to pollution (van Banning, 1987; Bucke, 1989).

2.3.4 Monitoring catch landed at ports on the north-east coast of England

A small number of cod (49 - 99 cm length, average 67.6 cm) with ulcer syndrome (synonymous with dermal necrosis, Bucke, 1989) were reported during routine biological sampling from ports in north-east England (the actual numbers of affected fish landed were unknown, because routine monitoring at ports does not require each individual fish to be examined). The condition, which has been investigated by us since 1979, continues to be found in cod, predominantly in large fish, landed at a small number of ports in north-east England (Bucke, 1989). No aetiology has been ascribed to this condition, but landing returns have indicated a localised pool of affected fish exists (Figure 20) and that specimens arrive at low, but regular, intervals at ports throughout the year. No other significant diseases in landed fish were reported to FDL during 1990.

Figure 20. Cod skin necrosis 'hotspots' data from monthly returns 1988-90

3. THE MONITORING PROGRAMME FOR CONTROL OF DISEASE IN FARMED STOCKS AND ITS TRANSMISSION TO WILD STOCKS

3.1 Introduction

Statutory controls, to prevent the introduction and spread of fish disease in Great Britain, have existed for more than 50 years. These are derived from the Diseases of Fish Act (1937) (Great Britain - Parliament, 1937) as amended by the Diseases of Fish Act (1983) (Great Britain - Parliament, 1983). The controls relate to salmon, trout and other freshwater fish and operate to prevent the introduction and spread of disease. They prohibit the importation of live salmonids into Great Britain, except from Northern Ireland, and imports of salmonid ova are permitted only under licence with supporting health certification. Similar licensing controls also apply to the importation of other types of freshwater fish and their ova which are likely to be released into the wild. The Acts also give the regulatory authorities powers to control the spread of serious 'notifiable' diseases. The following diseases are the only ones which at present come into this category:

- viral haemorrhagic septicaemia (VHS);
- infectious haematopoietic necrosis (IHN);
- infectious salmon anaemia (ISA);
- gyrodactyliasis caused by *Gyrodactylus salaris;*
- bacterial kidney disease (BKD);
- infectious pancreatic necrosis (IPN);
- whirling disease (WD);
- enteric redmouth (ERM) (Scotland only);
- furunculosis in salmon; and
- spring viraemia of carp (SVC).

Apart from SVC, these are diseases of trout and salmon and the effects are much more pronounced under intensive farming conditions. The viral diseases VHS and IHN are by far the most serious of the diseases on this list but both of them are absent from British waters as are gyrodactyliasis and ISA.

Marine shellfish are regulated under the Sea Fisheries (Shellfish) Act (1967) (Great Britain - Parliament, 1967). The Act controls the depositing of molluscan shellfish and lobsters in coastal waters. The mollusc controls operate on shellfish of UK or imported origin and cover the bivalve pests, Mytilicola, Crepidula and American tingle and a range of diseases such as *Bonamia, Marteilia* and Haplosporidia. Controls on lobsters were specifically designed to prevent the spread of gaffkaemia. This is a bacterial septicaemic condition caused by *Aerococcus viridans,* commonly imported in North American lobsters and which may be transmitted to our native stocks.

FDL has responsibility for fulfilling the Department's statutory inspection and diagnostic duties in England and Wales under these Acts. Inspectors visit farm, river and large open water sites to examine fish or shellfish where notifiable or other serious diseases are suspected and samples are taken for laboratory testing. This inspection, together with programmes for monitoring certain other serious diseases, provides for an assessment of the incidence, prevalence and significance of diseases of fish and shellfish generally in England and Wales. This information provides a scientific basis, *inter alia,* for governmental policies and the application of statutory measures for such diseases.

When notifiable diseases have been confirmed by testing, or are suspected, an Inspector has the powers to enter sites and prohibit or regulate the movement of live fish, ova and foodstuffs into and out of the designated infected area and to regulate the removal and disposal of dead and dying fish. Inspection and laboratory testing of cultivated and wild stocks of fish and shellfish are also undertaken for health certification to facilitate exports.

Since 1985, under the Registration of Fish Farming and Shellfish Farming Businesses Order (1985) (Great Britain - Parliament, 1985), it has been a legal requirement that all fish and shellfish farming businesses in England and Wales register with MAFF in order to assist efforts to prevent the spread of disease. There are currently some 569 registered fish farms and 225 registered shellfish farms. Inspectors are charged with checking the registration details and movement records which must be kept by such registered businesses.

3.2 Materials and methods

3.2.1 Sampling methods

The way in which samples are selected is important, whether the samples are being taken for monitoring or for investigating the cause of outbreaks of clinical disease. Prior to sampling, an assessment of the site and husbandry conditions which prevail may be needed, as well as an inspection of the fish or shellfish present. For monitoring purposes, sites are generally sampled to a statistical standard which provides for a 95% confidence level of detecting a 2% incidence of disease (Oassinder and Wedemeyer, 1973). This means that, in practice, 150 animals are sampled on site from across the population. With 'wild' populations, which are at lower densities and where sample size is limited, sampling to give a 95% confidence level of detecting disease at the 10% level is acceptable (Oassinder and Wedemeyer, 1973). Where overt clinical disease is manifested, sampling will normally be limited to 5-10 animals.

3.2.2 Laboratory tests

(a) ***Virology***: Testing of fish for viruses depends primarily on the isolation of the virus through passage in tissue culture, but an enzyme-linked immunosorbent assay (ELISA) for early identification of viral antigen is used in cases of clinical disease. For isolation of disease organisms, viscera (liver, kidney, spleen and pyloric caeca for salmonids; liver, spleen, kidney and brain for cyprinids) are pooled from 5 fish and homogenised with sterile sand and balanced salt solution (Hank's or Earl's) using a pestle and mortar. To eliminate contamination, the homogenates are diluted with a high-level balanced salt solution (containing the antibiotics penicillin at 1 000 i.u. ml^{-1}, streptomycin at 1 000 mg ml^{-1} and mycostatin at 50 i.u. ml^{-1}). Cell cultures are inoculated with extracts prepared from pooled homogenates and incubated for one week before a repeat passage and a further week's incubation. The type of cell culture used is dictated by the virus under investigation (Table 11). Passage of a virus through a tissue culture is normally accompanied by cytopathological effects (CPE) and confirmation of the identity of a virus causing CPE is achieved by means of a serum neutralisation test or ELISA (Way and Dixon, 1988).

Table 11. The susceptibility of some important viral fish pathogens to cell culture

Cell line	Fish viruses			
	IPN	VHS	IHN	SVC
BF	√	√	√	
FHM		√	√	√
CHSE		√	√	

(b) ***Bacteriology***: In testing for *Renibacterium salmoninarum*, the causative agent of BKD, sterile swabs are used to take fresh kidney samples which are plated onto specific kidney disease medium (Austin *et al.*, 1983). Plates are incubated at 15°C for up to 6 weeks and examined weekly. Confirmatory tests for the organism include an immuno-fluorescent antibody test (Bullock and Stuckey, 1975), the API enzyme test (Austin *et al.*, 1983), and the co-agglutination test (Dixon, 1987).

Tests for *Aerococcus viridans*, the causative agent of gaffkaemia, are based on isolation of the organism from haemolymph samples in presumptive gaffkaemia broth or directly on brain-heart infusion and blood agar plates following incubation at 28°C for 3-5 days (Stewart *et al.*, 1966). Confirmation of any presumptive gaffkaemia organisms which exhibit the typical tetrad form, are Gram-negative, and catalase-negative is obtained using a co-agglutination test.

(c) ***Parasitology***: Tests for WD are based on the examination of Giemsa-stained histological sections of cranial cartilage and bone for evidence of spores of *Myxobolus cerebralis*, the causative agent.

Tests for the oyster parasite *Bonamia ostreae* also rely on histological examination. A 5 mm steak through the gills and digestive tract is fixed in Davidson's seawater fixative (Shaw and Battle, 1957). Sections are stained by Gomori's one-step trichrome method (Drury and Wallington, 1973).

3.3 Field investigations under the Diseases of Fish Acts (1937 and 1983)

The fish disease testing in 1990 included: (a) tests on fish from sites where notifiable or serious disease was suspected; (b) the annual monitoring of trout hatcheries and salmon sites for IPN and salmon sites for WD; and (c) re-tests on sites already designated for SVC and BKD prior to 1990. Previous trends in the incidence of the major diseases of fish and methods of control have been reported by Hudson and Holliman (1985).

3.3.1 Investigations into the cause of disease outbreaks in fish

In 1990, 110 cases of fish mortality were investigated, including 62 where SVC was suspected but was not confirmed. IPN was confirmed from 6 rainbow trout farms, 3 of which had hatcheries. Two of those with hatcheries were designated as infected areas under the Diseases of Fish Acts, and the third undertook a stock

clearance and disinfection programme. The hot, dry summer was accompanied by high levels of mortality on salmon and trout farms in which Proliferative Kidney Disease (PKD), furunculosis and ERM were implicated.

3.3.2 Investigations to monitor for specific diseases of fish

(a) ***Infectious pancreatic necrosis***: A programme of monitoring of trout hatcheries and salmon sites for IPN was instigated following a relaxation in policy in 1984, concerning the control of fish movements from sites designated for this disease. All registered sites holding salmon and 10 of the major trout hatcheries are now tested annually, and the selection of trout sites is varied each year. In 1990, monitoring undertaken at the 10 rainbow trout hatcheries (each producing more than one million fry), and at all 22 registered sites holding salmon, all proved negative.

(b) ***Whirling disease***: Monitoring for WD was also instigated following the 1984 policy changes, and currently annual routine testing is limited to registered sites holding salmon of an age suitable for testing. In 1990, monitoring for WD was undertaken on the 19 registered salmon sites holding parr and smolts of a suitable age. All tests proved negative.

3.3.3 Re-tests for specific diseases of fish on designated sites

(a) ***Bacterial kidney disease***: Sites where BKD is confirmed are designated as diseased and controls are imposed on movement of stock. At present, three sites are designated as having this disease and annual re-tests are undertaken to reconfirm the 'disease status' of the sites. Bi-annual re-tests on two sites designated as having BKD (one in Cumbria, the other in Hampshire) were negative in 1990. Tests on a third (in Devon) were not needed as the site had been non-operational for a third year running.

(b) ***Spring viraemia of carp***: Following outbreaks of SVC in 1988 and early 1989, some 40 sites were found to be infected and had restrictions imposed on movement of stock. Eighteen of these have now undertaken clearance and/or disinfection. The 22 shown in Figure 21, which remain designated as SVC sites, are generally large open waters which are used as fisheries but where clearance and disinfection is not possible. Re-tests on fish from these waters, taken in spring 1989, failed to show evidence of infection. Though 150 fish samples are normally required for monitoring purposes, because of the lower population levels in these waters and the difficulty in netting, the target sample size was reduced to 30 fish. Even so, some samples still

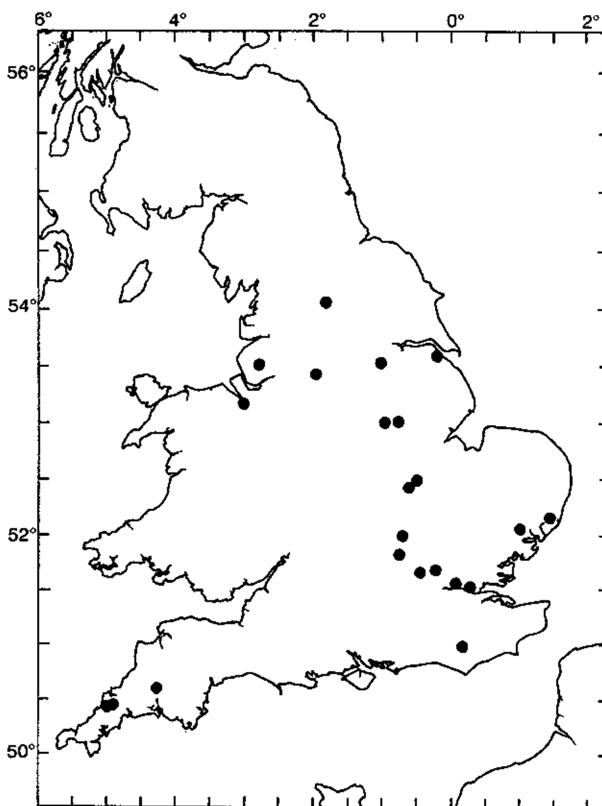


Figure 21. The distribution of sites designated as infected with SVC in England and Wales

fell below this requirement and the fish included in the sample were not always of the most sensitive indicator species or of a suitable age class. Details of the fish sampled in 1990 are shown in Table 12. Of the 22 sites, 20 were sampled and all gave negative results for SVC virus. The remaining sites included a private ornamental garden pond and a site on which only trout remained.

3.4 Field investigations under the Sea Fisheries (Shellfish) Act (1967)

Under this Act, the coast of England and Wales is divided into 27 'Control of Deposit' areas, as shown in Figure 22, and transfer of molluscan shellfish into and between these areas for deposit is controlled by licensing. Shellfish testing, under this Act, includes examinations of shellfish stocks where serious disease is suspected and the tracing of contacts when disease has been confirmed. Annual monitoring of *Ostrea edulis* stocks for *Bonamia* is also undertaken in coastal areas with a past history of disease, to establish the levels of infection and the implications for both wild and farmed stocks and to permit control of its spread by licensing movement of stock. Details of the impact and spread of *Bonamia* in the UK has recently been published (Hudson and Hill, 1991).

Table 12. Numbers and species of fish examined in SVC tests during 1990

Site	FCP	MRC	GHO	FCC	FGF	FRO	FRD	FGN	FTE	CUB	FBM	FBK	FPE	FPI	FPP	Total
Abram Flash	2	0	0	15	0	5	0	0	6	0	0	0	5	1	0	34
Bestwood duck ponds	0	0	0	4	0	11	0	0	2	0	0	0	5	8	0	30
Shotton Lake	0	0	0	0	0	28	0	0	0	0	2	0	1	0	0	31
Deen Park Lake	10	2	0	2	0	0	14	0	0	0	0	0	2	0	0	30
Carevick Lake	10	0	0	0	0	0	19	0	1	0	0	0	0	0	0	30
High Wycombe	2	0	0	1	0	4	0	0	4	0	0	0	9	10	0	30
Marley Tiles Angling Club	5	0	0	0	1	4	2	0	6	0	0	0	1	0	0	19
Orchard Close Lake	2	0	0	0	0	16	0	0	1	0	3	3	5	0	0	30
Grimsby (Garden pond)	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Romiley Angling Club	6	0	0	3	0	4	2	5	2	0	0	0	5	0	0	27
Sapphire Lakes	3	0	0	0	0	10	0	0	1	0	10	0	10	0	0	34
Sibton Lake	9	2	0	3	0	0	5	0	6	0	5	0	0	0	0	30
Toneham Pond	3	3	1	0	0	25	0	0	1	0	0	0	0	0	0	33
Trevella Lake	2	0	0	0	0	18	0	0	0	0	10	0	0	0	0	30
Tykeswater	0	1	0	10	0	5	0	0	5	2	2	0	5	2	0	32
Unity Lake	5	3	0	6	0	0	2	0	4	0	0	0	0	0	0	20
Unilever Research Laboratory, Shambrook	1	0	0	27	0	0	0	0	2	0	0	0	0	0	0	30
Warren Farm Lakes	5	0	0	5	0	0	5	0	5	0	0	0	0	0	0	20
Western Turnville	5	3	0	0	0	5	0	0	6	0	1	0	5	5	0	30
White House Lake (only trout remaining)	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Winsdon Farm Lake	0	2	0	0	0	14	0	0	3	0	11	0	0	0	0	30
Winton Fish Farm	8	3	0	0	0	15	1	0	0	0	0	0	2	0	1	30

FCP = Common carp; FRO = Roach; FBM = Bream; MRC = Mirror carp; FRD = Rudd; FBK = Bleak; GHO = Ghost koi; FGN = Gudgeon; FPE = Perch; FCC = Crucian carp; FTE = Tench; FPI = Pike; FGF = Goldfish; CUB = Chub; FPP = Zander

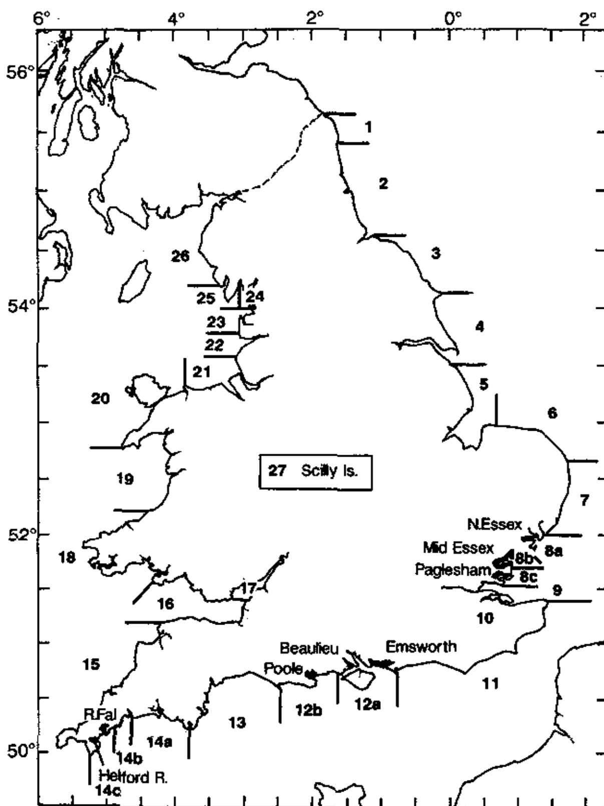


Figure 22. 'Control of Deposit' areas in England and Wales. Sites infected with Bonamia are labelled

3.4.1 Investigations to monitor for specific diseases of shellfish

(a) ***Bonamia***: Some 44 samples, comprising 5 280 *O. edulis*, were examined during 1990 and details of the results are given in Table 13. No new locations were found to be infected. Results from farmed stocks relaid in infected areas 8a, 8b and 12b under MAFF guidelines (Hudson and Hill, 1991) were promising with infection levels usually remaining below the 10% level; above which immediate clearance of the beds is recommended.

Wild stocks in the Solent Fishery have continued to remain negative for *Bonamia* with no evidence of spread of infection from infected stocks in the mouth of the Beaulieu river or Emsworth. However, the impact of the disease in wild stocks in parts of Poole Harbour and the Fal was found to be much more serious both at the population and individual animal level. Infection levels in wild Fal oysters from Maggoty Bank, East Bank and Parsons Bank rose from 9%, 9% and 7% in 1989 to 20%, 11% and 15.3%, respectively, in 1990. It was also evident during fishing for the oysters in the Fal in autumn 1990 that few 3-year and older oysters remained and the high numbers of fresh dead oysters (clean shell) suggest substantial losses have occurred.

Table 13. Numbers of Ostrea edulis tested for Bonamia during 1990

Area (see Figure 22)		Site	Results					
			No. of Oysters	No. positive	No. negative	Average infection rate (%)	Min (%)	Max (%)
8a	Walton	2 cultivation sites	300	2	0	15.5	10	21
8b	Blackwater	4 cultivation sites	550	3	1	4	0	12
9	N. Kent	1 cultivation site	89	0	1	0	0	0
12a	Solent: Lepe)		450	0	3	0	0	0
	Calshot)		150	0	1	0	0	0
	Chilling)		300	0	2	0	0	0
	Stanswood)		300	0	2	0	0	0
	Osbourne)		150	0	1	0	0	0
	Browndown)		150	0	1	0	0	0
	Pennington)	15 natural beds	150	0	1	0	0	0
	Newtown)		150	0	1	0	0	0
	Chichester)		54	0	1	0	0	0
	Bosham)		43	0	1	0	0	0
	Thorney)		135	0	1	0	0	0
	Portsmouth)		150	0	1	0	0	0
	Langstone)		150	0	1	0	0	0
	Beaulieu)		82	1	0	4	4	4
	Emsworth)		152	1	2	2	0	5
12b	Poole: Holes Bay	1 natural bed	8	1	0	38	38	38
		4 cultivation sites	364	3	1	4	0	5
	Weymouth	1 cultivation site	150	0	1	0	0	0
14b	Fal: East Bank)		450	3	0	11.3	6	15
	Parsons Bank)	4 natural beds	450	3	0	15.3	5	27
	Maggoty Bank)		150	1	0	20	20	20
	Tolverne)		150	1	0	21	21	21
18	Milford Haven	1 cultivation site	12	0	1	0	0	0
25	Morecambe	1 cultivation site	41	0	1	0	0	0

Following a number of years of poor natural spat falls of *O. edulis*, low levels of spat settlement were recorded in the Essex creeks in 1988, 1989 and 1990. These oysters have settled adjacent to or within relaying areas and may not have been dredged out at the end of the season as recommended in the MAFF guidelines because of their size or position. This could result in the build-up of *Bonamia* infection during the longer-term exposure of this wild stock and in early infection and losses in stock relaid for one season (as the guidelines recommend) in future years.

3.4.2 Tests for gaffkaemia

One case of gaffkaemia caused by *Aerococcus viridans* was confirmed in native lobsters from a holding site in north-west Wales in 1990. Losses exceeding 10% per day were reported and the owner opted for clearance and disinfection under MAFF supervision. Tentative links with North American imports could only be established through packaging materials previously used for North American imports. Tests undertaken on wild stocks taken from waters adjacent to the holding unit and subjected to stress testing failed to show evidence of infection.

3.4.3 Tests for crayfish plague

Reports of crayfish mortalities during 1990 led to the confirmation of 4 outbreaks of crayfish plague caused by *Aphanomyces astaci*, 3 of which were in new water catchments. Using isolation methods described by Alderman and Polglase (1986), a further plague outbreak was confirmed in the Mells River in the Bristol Avon catchment where plague has been present since 1981. Newly-infected catchments in 1990 were the Nene (River Ise at SP 845880), the Wye (River Arrow at SP 390590), and the Upper Severn (River Camlad at SO 275970).

The reference to proprietary products in this report should not be construed as an official endorsement of these products, nor is any criticism implied of similar products which have not been mentioned.

4. REFERENCES

ALDERMAN, D.J. and POLGLASE, J.L., 1986. *Aphanomyces astaci*: isolation and culture. J. Fish Dis., *9(5)*: 367-379.

AUSTIN, B., EMBLEY, T.M. AND GOODFELLOW, M., 1983. Selective isolation of *Renibacterium salmoninarum*. FEMS Microbiol. Lett., *17*: 111-114.

AYRES, P.A. AND CULLUM, M., 1978. Paralytic shellfish poisoning: an account of investigations into mussel toxicity in England 1968-77. Fish. Res. Tech. Rep., MAFF Direct. Fish. Res., Lowestoft, *(40)*: 1-24.

BRANDER, K.M., COLEBROOK, J.M., REES, I.S., PARKER, M.M. AND BUCKE, D., 1988. General biology and fisheries of the Irish Sea. Coop. Res. Rep., int. Coun. Explor. Sea, *(155)*: 25-60.

BUCKE, D., 1989. Observations on visceral granulomatosis and dermal necrosis in populations of North Sea cod *(Gadus morhua)*. ICES CM 1989/E:17, 9pp. (mimeo).

BUCKE, D. AND FEIST, S.W., 1984. Histological changes in livers of dab, *Limanda limanda*. ICES CM 1984/F:8, 8pp. (mimeo).

BUCKE, D. AND FEIST, S.W., 1990. Marine fish diseases: an overview of 10 years' investigations by MAFF (UK). ICES CM 1990/E:12, 9pp. (mimeo).

BUCKE, D. AND NICHOLSON, M.D., 1987. Fish disease investigations in the Irish Sea. ICES CM 1987/ E:19, 12pp. (mimeo).

BUCKE D. AND STOKES K., 1989. Fish disease monitoring on the 1987 North Sea groundfish survey. ICES CM 1989/E:7, 10pp. (mimeo).

BUCKE, D., NORTON, M.G. AND ROLFE, M.S., 1983(a). The field assessment of effects of dumping wastes at sea. II. Epidermal lesions and abnormalities of fish in the outer Thames estuary. Fish. Res. Tech. Rep., MAFF Direct. Fish. Res., Lowestoft, *(72)*: 1-16.

BUCKE, D., FEIST, S.W. AND ROLFE, M.S., 1983(b). Fish disease studies in Liverpool Bay and the north-east Irish Sea. ICES CM 1983/E:15, 6pp. (mimeo).

BULLOCK, G.L. AND STUCKEY, H.M., 1975. Fluorescent antibody identification and detection of the Corynebacterium causing kidney disease of salmonids. J. Fish. Res. Bd Can., *32*: 2224-2227.

COULSON, J.C., POTTS, G.R., DEANS, I.R. AND FRASER, S.M., 1968. Exceptional mortality of shags and other sea-birds caused by paralytic shellfish poisoning. Brit. Birds, *61*: 381-404.

DIXON, P.F., 1987. Comparison of serological techniques for the identification of *Renibacterium salmoninarum*. J. Appl. Ichthyol., *3*: 131-138.

DRURY, R.A.B. AND WALLINGTON, E.A., 1973. 'Carleton's Histological Technique', 4th edn. Oxford University Press, London. 432 pp.

GREAT BRITAIN - PARLIAMENT, 1937. Diseases of Fish Act 1937. Her Majesty's Stationery Office, London. llpp.

GREAT BRITAIN - PARLIAMENT, 1967. The Sea Fisheries (Shellfish) Act 1967. Her Majesty's Stationery Office, London. 20pp.

GREAT BRITAIN - PARLAIMENT, 1983. Diseases of Fish Act 1983. Her Majesty's Stationery Office, London. 16pp.

GREAT BRITAIN - PARLAIMENT, 1985. The Registration of Fish Farming and Shellfish Farming Businesses Order 1985. Her Majesty's Stationery Office, London. 7pp. Statutory Instrument 1985 No.1391.

GREENBURGH, A.E. AND HUNT, D.A., 1985. Laboratory Procedures for the Examination of Seawater and Shellfish. 5th edn. American Public Health Association, Washington D C. 144pp.

HUDSON, E.B. AND HILL, B.J., 1991. Impact and spread of bonamiasis in the UK. Aquaculture, *93*: 279-285.

HUDSON, E.B. AND HOLLIMAN, A., 1985. Salmonid fish farming in England and Wales. The major diseases of fish - trends in incidence and methods of control. State Vet. J.,*39*: 91-109.

ICES, 1988. Report of the ICES Working Group on Pathology and Diseases of Marine Organisms. ICES CM 1988/F:37, 96pp. (mimeo).

ICES, 1989. Methodology of fish disease surveys. Report of an ICES sea-going workshop held on U/F ARGOS, 16-23 April, 1988. Coop. Res. Rep., int. Coun. Explor. Sea., *(166)*: 1-43.

LEE, J.S., YANAGI, T., KENMA, R. AND YASUMOTO, T., 1987. Fluorometric determinations of diarrhetic shellfish poisons by high-performance liquid chromatography. Agric. Biol. Chem., *51*: 877-881.

MAFF, 1990. Monitoring and surveillance of non-radioactive contaminants in the aquatic environment, 1984-1987. Aquat. Environ. Monit. Rep., MAFF Direct. Fish. Res., Lowestoft, *(22)*: 1-60.

MAFF, 1991. Monitoring and surveillance of non-radioactive contaminants in the aquatic environment, 1988-1989. Aquat. Environ. Monit. Rep., MAFF Direct. Fish. Res.,Lowestoft, *(26)*: 1-90.

McCollum, J.P.K., Pearson, R.C.M., Ingham, H.R., Wood, P.C. and Dewar, H.A., 1968. An epidemic of mussel poisoning in north-east England. The Lancet, Oct. 5, 767-770.

McVicar, A.H., Bruno, D.W. and Fraser, C.O., 1988. Fish diseases in the North Sea in relation to sewage sludge dumping. Mar. Pollut. Bull., *19*: 169-173.

NSTF, 1990. North Sea Task Force Monitoring Master Plan 1990-91; Department of the Environment, London. 37pp. (North Sea Environment Report (*3*)).

Oasinder, F.J. and Wedemeyer, G., 1973. Computer programme for sample sizes required to determine disease incidence in fish populations. J. Fish. Res. Bd Can., *30*: 1383-1384.

Shaw, B.L. and Battle, H.I., 1957. The gross and microscopic anatomy of the digestive tract of the oyster, *Crassostrea virginica* (Gmelin). Can. J. Zool., *35*: 325-347.

Shelton, R. G. J. and Wilson, K. W., 1973. On the occurrence of cymphocystes, with notes on other pathological conditions in the flatfish stocks of the north-east Irish Sea. Aquaculture, (*2*): 395-410.

Stewart, J.E., Cornick, J.W., Spears, D.I. and Mcleese, D.W., 1966. Incidence of *Gaffkya homari* in natural lobster (*Homarus americanus*) populations of the Atlantic region of Canada. J. Fish. Res. Bd Can., *23*: 1325-1330.

Sullivan, J.J. and Wekell, M.M., 1986. The application of high pressure liquid chromotography in a paralytic shellfish poisoning monitoring program. pp357-371. *In*: D.E. Kramer and J. Liston (Eds.) 'Seafood quality determinations', Proceedings of an International Symposium, 10-14 November 1985. University of Alaska, Sea Grant College Program, Anchorage.

Van Banning, P., 1987. Long-term recording of some diseases using general fishery research surveys in the south-east part of the North Sea. Dis. Aquat. Org., *3*: 1-11.

Way, K. and Dixon, P.F., 1988. Rapid detection of VHS and IHN viruses by the enzyme linked immunosorbent assay (ELISA). J. Appl. Ichthyol., *4*: 182-189.

Wood, P.C. and Mason, J., 1969. Paralytic shellfish poisoning: a short account of an outbreak occurring on the north-east coast of Britain in May 1968. ICES C.M. 1969/K:25, 4pp. (mimeo).

ANNEX 1. Areas of monitoring mentioned in the text and staff responsible for the projects

Biotoxins

Dr D J Alderman

W Doré

Mrs K Henshilwood

Dr G Barker

A Hancock

Dr M Waldock (Burnham-on-Crouch)

Marine Fish Diseases

D Bucke

S W Feist

Miss S Swaine

P B Moore (Sea Fisheries Inspectorate Office, North Shields)

Statutory Monitoring

E B Hudson

D McGregor

A E Howard

Dr C J Rodgers

Dr B P Wood

Mrs A Forrest

Mrs C Crane

Ministry of Agriculture, Fisheries and Food
Directorate of Fisheries Research
Fisheries Laboratory
Lowestoft
Suffolk
NR33 OHT
England

AQUATIC ENVIRONMENT MONITORING REPORT Number 35