CENTRE FOR ENVIRONMENT, FISHERIES AND
AQUACULTURE SCIENCE

AQUATIC ENVIRONMENT MONITORING REPORT
Number 57

Monitoring of the quality of the marine environment, 2002-2003

LOWESTOFT
2005

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Sci. Ser., Aquat. Environ. Monit. Rep., CEFAS, Lowestoft, (57): 64pp.

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FOREWORD

Aquatic Environment Monitoring Report No. 57 collects together work carried out in 2002-03 by CEFAS scientists in support of our monitoring and surveillance duties (see overleaf for the background to the work). The information presented covers both environmental surveillance at offshore and coastal sites and site-specific work carried out in support of risk assessments and regulatory procedures. Some of the science reported here forms part of wider efforts to integrate data from Departments and Agencies in the UK to provide a comprehensive picture of the quality of the marine environment via the UK National Marine Monitoring Programme (NMMP). Other components are unique to CEFAS due to our requirement to understand ecosystem response resulting from potential pressures from deposit, extraction and discharge activities.

The strategy for the NMMP is described in publications commissioned by the Marine Environment Monitoring Group (MEMG). The programme seeks to develop time trend data for a limited number of sites around the UK and this work is augmented by special surveys of compounds likely to pose specific risks, or for which few data exist. The programme manual, known as the Green Book, is available in downloadable format from the Scottish Environmental Protection Agency website at: www.sepa.org.uk/marine

In this report, Chapter 1 describes work on radioactivity in coastal waters, and Chapter 2 a study of commonly used pharmaceutical compounds in UK estuaries. Chapter 3 describes an assessment of the endocrine-disrupting potential of produced water discharged from offshore installations. Chapter 4 outlines recent findings relating to contaminants in marine mammals, undertaken within the UK Marine Mammals Strandings Programme, which underpins UK commitments under the ASCOBANS (Agreement on the Conservation of Small Cetaceans of the Baltic and North Seas) treaty. Chapter 5 provides benchmark information on the current levels of mercury in fish from Liverpool Bay, an area of historic contamination which is monitored as a requirement of OSPAR. Chapter 6 describes the CEFAS contribution to the North Sea Benthos project, an international collaborative study. Chapter 7 concerns fish disease and pathology biomarkers, and Chapter 8 describes the use of the CALUX assay to determine the activity of compounds which exert their effects in the same manner as dioxins in UK estuaries. Chapter 9 describes the application of two of the biological effects techniques developed for the NMMP (the determination of EROD and bile metabolites) in fish. Chapter 10 assesses the data to date on PAH in sediments at intermediate and offshore NMMP stations and their suitability for the detection of time trends in concentration. Chapter 11 outlines studies of rehabilitation at aggregate extraction sites.

There is a close link between the focused research conducted at CEFAS and the regulatory processes on which we advise Government, and Chapters 12 to 17 relate to the sea disposal of dredged material. Chapter 12 summarises the activity in licensing of deposits at sea in 2002 and 2003. Chapters 13 and 14 describe the levels of PAH contamination in samples of dredged material and at selected disposal sites around England and Wales, and Chapter 15 describes levels of radioactivity in dredged materal. Chapter 16 describes the levels of TBT contamination at selected dredged material disposal sites around England and Wales. Chapter 17 is a case study which outlines monitoring approaches taken at the Rame Head disposal site off Plymouth.

During the period under report, CEFAS has also assisted Defra in the preparation of their '*Charting Progress*' report, which will be available for downloading from www.defra.gov.uk early in 2005. This report, earlier reports in this series and other publications are available in downloadable format from the CEFAS website www.cefas.co.uk

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BACKGROUND TO THE WORK

As an Executive Agency of the Department for Environment, Food and Rural Affairs (Defra), CEFAS carries out work in support of Defra's five strategic priorities, all of which underpin the overarching aim of promoting sustainable development:

- Climate change and energy
- Sustainable consumption and production
- Natural resource protection
- Sustainable rural communities
- A sustainable farming and food sector, including animal health and welfare.

Within these priorities, environment work at CEFAS is directed at research, monitoring and assessment of the impact of potentially harmful substances or activities on the quality of the marine, coastal and estuarine environments. We are involved directly in advising on UK and international legislation and in developing policy relating to management of the aquatic environment. We provide advice to Governments, enforcement agencies and policymakers throughout the world on the development and implementation of monitoring and assessment programmes and control measures.

An important component of our work is to provide advice to Defra Ministers and other Government Departments on all aspects of non-radioactive contamination of the aquatic environment. Specifically under Part II of the Food and Environment Protection Act (1985) (FEPA) (Great Britain Parliament, 1985a), Defra has the responsibility to licence and control the deposit of material to sea. Following the cessation of the disposal of sewage sludge to sea, licensed materials are predominantly sediments, derived from maintenance and capital dredging activities in coastal waters. Disposal at sea is also regulated internationally by OSPAR, and our work enables the UK to fulfil its obligations as a Contracting Party.

The CEFAS Inspectorate evaluates scientific and technical aspects of licence applications and makes regular visits to licence holders to ensure that any stipulated conditions are being met. Conducting monitoring programmes in support of risk assessments enables Defra to ensure the effectiveness of the assessment process and provides a basis for decisions on future policy for the management of marine resources. CEFAS scientists monitor the environmental conditions at marine disposal sites and compare the results with those obtained during more general monitoring studies, allowing action to be taken if unexpected impacts should occur. This also provides a feedback loop which ensures that risk assessments undertaken within the licensing process incorporate the most recent research findings.

Under the Water Resources Act (1991) (Great Britain Parliament, 1991), Defra is a statutory consultee for all discharges to controlled (tidal) waters. CEFAS scientists assess the fishery implications of applications for consent to discharge permits. Consideration is given to resources in the area, the toxicity of the effluent, local hydrographic conditions and any standards set out in national policy or EU Directives.

We also provide advice to the Department of Trade and Industry (DTI) and the Office of the Deputy Prime Minister (ODPM) concerning the control of pollution in other areas affecting the marine environment including the extraction of offshore oil and gas and marine aggregate. The statutory Offshore Chemical Notification Scheme and the Government View on the winning of aggregates, respectively, control these activities, and the regulatory regime for aggregates is presently also changing to a statutory scheme.

On Defra's behalf, CEFAS is responsible for monitoring intermediate and offshore stations within the UK National Marine Monitoring Programme (NMMP), which seeks to integrate national and international monitoring programmes for all UK agencies. Each year, we collect samples of seawater, sediment and biota for chemical analysis and deploy a number of biological effects techniques, including water and sediment bioassays and fish disease surveys. The current phase of the NMMP (phase II) is focused on the detection of long-term temporal trends in contaminant concentrations and the development and deployment of a wider range of biological effects techniques studying organism response at a variety of cellular and sub-cellular levels. The NMMP allows us to ascertain the effectiveness of regulatory measures taken to reduce the inputs of hazardous substances to UK seas. In addition, it contributes to the UK's international monitoring obligations to demonstrate compliance with various EU Directives: Dangerous Substances Directive (76/464/EEC); Shellfish Waters Directive (79/923/EEC); Shellfish Hygiene Directive (91/492/EEC); Fishery Products Directive (91/493/EEC); the Commission Decision 93/351/EEC concerning maximum mercury limits in fishery products, and similar requirements under OSPAR.

In order to ensure that the advice provided to Defra and other regulators is always based on the most up-to-date knowledge and techniques, CEFAS carries out a wide range of research and development to provide for the future needs of monitoring and surveillance programmes. For example, we have developed new and more sensitive bioassay techniques, analytical methods and unattended sampling and monitoring devices. Within these programmes we have made a number of significant contributions to environmental protection and as a consequence of our work have established a worldwide reputation in the field of aquatic environmental research. More information on our research programmes is available on the CEFAS website (www.cefas.co.uk).

GLOSSARY OF TERMS

ANOSIM	Analysis of Similarities
ASCOBANS	Agreement on the Conservation of Small Cetaceans of the Baltic and North Seas
ASG	Ammonium duodeca-molybdophosphate on silica gel
BDE	Brominated diphenyl ether
BEQUALM	Biological Effects Quality Assurance in Monitoring programme
CYP1A1	Cytochrome P4501A1
DNA	Deoxyribose Nucleic acid
DR-CALUX	Dioxin-responsive chemically-activated luciferase expression assay
EROD	Ethoxyresorufin-O-deethylase
EU	European Union
FEPA	Food and Environment Protection Act 1985
FRS MLA	Fisheries Research Services Marine Laboratory, Aberdeen
GSI	Gonado-somatic index
HBCD	Hexabromocyclododecane
ICES 7	The sum of concentrations of PCB congeners CB28, CB52, CB101, CB118, CB138, CB153 & CB180.
JMP	Joint Monitoring Programme (of OSPAR)
MDS	Multi-dimensional scaling
MEMG	Marine Environment Monitoring Group
MFO	Mixed-function oxidase
MPMMG	Marine Pollution Monitoring Management Group
NADPH	β-Nicotinamide Adenine Dinucleotide Phosphate
NMMP	UK National Marine Monitoring Programme
OSPAR	Oslo and Paris Commission
PAH	Polycyclic aromatic hydrocarbon
PCB	Polychlorinated biphenyl
PBDE	Polybrominated diphenyl ether
PRIMER	Plymouth Routines in Multivariate Ecological Research
RAP	Registry of Aquatic Pathology
TEQ	Toxic equivalent
TIE	Toxicity Identification and Evaluation
UK	United Kingdom
YES	Yeast oestrogen screening assay

SEA WATER

1. RADIOACTIVITY IN UK COASTAL WATERS

Author: David McCubbin

1.1 Introduction

Seawater surveys support international studies concerned with the quality status of coastal seas (e.g. OSPAR, 2000) and provide information which can be used to distinguish different sources of man-made radioactivity (e.g. Kershaw and Baxter, 1995). In addition, the distribution of radioactivity in seawater around the British Isles is a significant factor in determining the variation in individual exposures at coastal sites, as seafood is a major contribution to food chain doses. Therefore, a programme of surveillance of the distribution of key radionuclides is maintained using research vessels and other means of sampling.

Detailed historical data for ^{134}Cs and ^{137}Cs in seawater have been published in a series of reports so as to aid model development (Camplin and Steele, 1991; Baxter *et al*., 1992; Baxter and Camplin, 1993a-c) and have been used to derive dispersion factors for nuclear sites (Baxter and Camplin, 1994). The data have also been used to examine the long distance transport of activity to the Arctic (Kershaw *et al*., 1999) and long-term trends in Northern European seas (Povinec *et al*., 2003). In recent years (since 1994), discharges of ^{99}Tc from the British Nuclear Fuels plc (BNFL) facilities at Sellafield have increased significantly, against the overall trend observed for most other radionuclides. During 2003, discharges of ^{99}Tc from Sellafield were reduced due to a successful trial of abatement technology and are expected to reduce further in 2004 and subsequent years.

Studies of the migration behaviour of ^{99}Tc have afforded opportunities to substantiate and extend the information obtained from earlier similar studies of ^{137}Cs. The distribution of ^{99}Tc in waters around the British Isles prior to, and immediately after, the increased ^{99}Tc discharges indicated a rapid advection of ^{99}Tc within and from the Irish Sea to the north of Scotland as compared to previous estimates (Leonard *et al*., 1997a,b; McCubbin *et al*., 2002). The subsequent transport rate out of the North Sea and northwards with the Norwegian Coastal Current and West Spitsbergen Current slowed markedly, in apparent correspondence with variations in the North Atlantic Oscillation (NAO) winter index (Karcher *et al*., 2004; Kershaw *et al*., 2004).

1.2 Methods

1.2.1 Sampling

The research vessel programme on radionuclide distribution currently comprises an annual survey of the Bristol Channel together with biennial surveys of the Irish Sea and the North Sea. Large volume surface seawater samples (50 litres) are collected, using the ships pumped supply, during cruises of the CEFAS research vessels, *RV CIROLANA* and *RV CORYSTES*. In 2002, surveys of the Bristol Channel and North Sea were carried out. Additionally, for the first time, the waters of the western English Channel were included in the surveys since these may be affected by transport of radioactivity from the Irish Sea and Bristol Channel and by the legacy of past dumping in the Hurd Deep.

1.2.2 Sample analysis

Samples were filtered (0.45 µm) to separate dissolved and particulate phases. Analyses of dissolved ^{137}Cs involved pumping filtered seawater, acidified with nitric acid, through cartridges filled with ASG resin (ammonium duodeca-molybdophosphate on silica gel) to extract caesium. Analyses of ^{3}H involved double distillation of water samples under alkaline conditions and in the presence of holdback carriers to ensure chemical separation from all gravimetric and radiometric interference. Subsamples of distillate were assayed for ^{3}H using a Packard Tri-Carb 2550 TR/LL liquid scintillation counter.

1.3 Results and discussion

The results of the seawater surveys are given in Figures 1(a)-1(d), overleaf.

1.3.1 ^{137}Cs distribution

The North Sea ^{137}Cs data (Figure 1a) indicate that levels ranged from ~0.0025 Bq l^{-1} up to ~0.0079 Bq l^{-1} (i.e. 3 fold variation). The concentration pattern of ^{137}Cs was, however, rather irregular. At some sampling sites, concentrations were only marginally elevated above the 'background' level in surface Atlantic seawater (~0.002 Bq l^{-1}, Dahlgaard *et al*., 1995) due to global fallout from atmospheric testing of nuclear weapons in the 1950s and early 1960s. Enhanced concentrations were observed at other sites due to the influx of contaminated water masses originating from the Irish Sea and the Baltic. Continuing ^{137}Cs remobilisation from seabed sediments, contaminated by discharges from the Sellafield nuclear

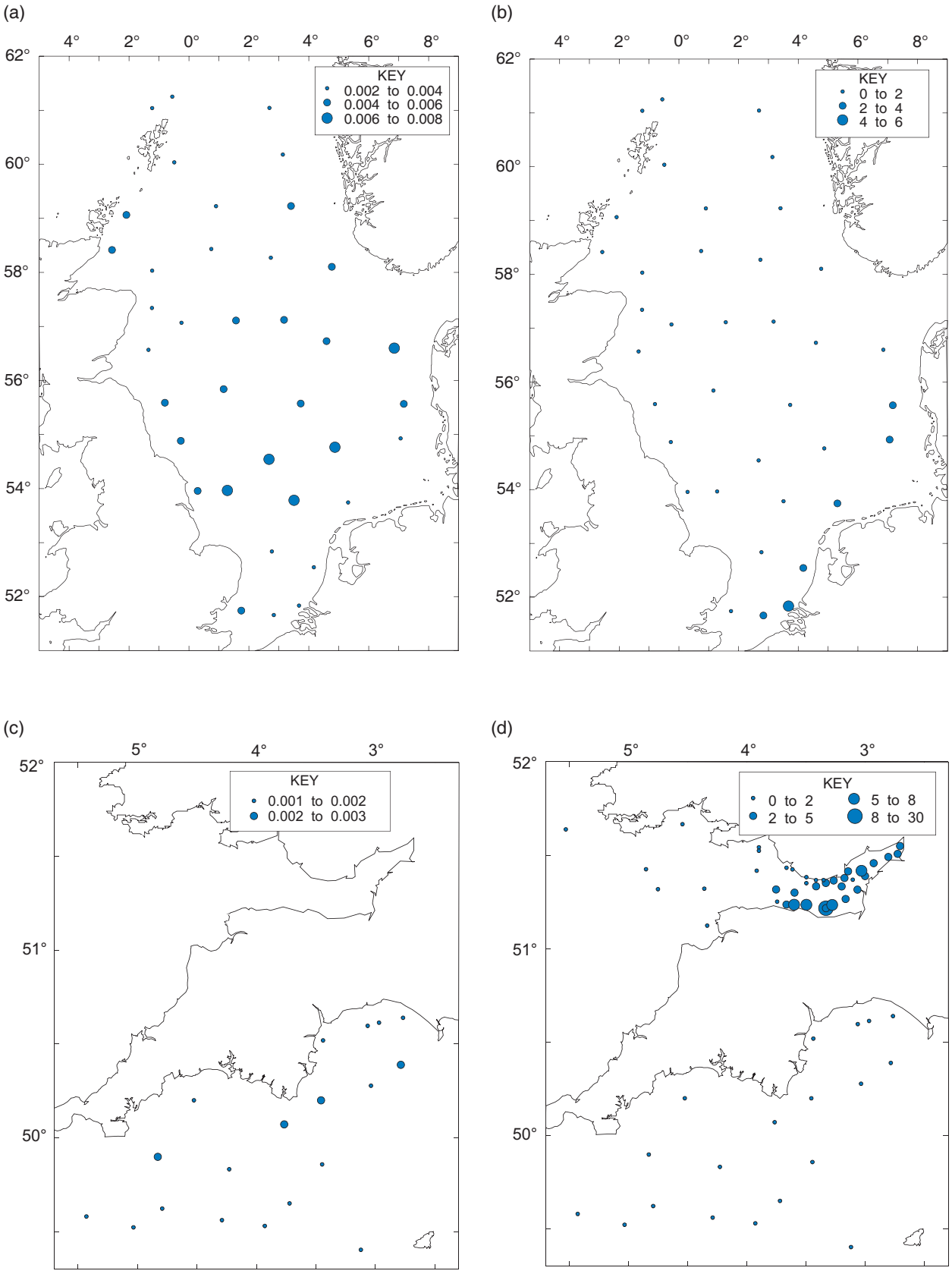

Figure 1. ***Concentrations of ^{137}Cs (Bq l^{-1}) and ^{3}H (Bq l^{-1}) in surface seawater from the UK continental shelf: a) Dissolved ^{137}Cs in the North Sea (August-September 2002), b) ^{3}H in the North Sea (August-September 2002),c) Dissolved ^{137}Cs in the western English Channel (September - October 2002), d) ^{3}H in the Bristol Channel and western English Channel (September - October 2002)***

fuels reprocessing plant in the 1970s, appears to be the predominant (~90%) source term to the water column in the Irish Sea (Leonard *et al*., 1998). This flows into the North Sea via the Scottish coastal current and southwards and eastwards along the coastal margin. En route, it gradually mixes with poor Atlantic water containing lower ^{137}Cs concentrations that provide the major water inflow into the North Sea. It exits the North Sea via the Norwegian Coastal Current. The Baltic Sea was more heavily contaminated than the North Sea by ^{137}Cs arising from the Chernobyl accident in 1986, and since the outflow from the Baltic Sea is restricted. Baltic waters continue to provide a net source of ^{137}Cs to the North Sea (Kershaw and Baxter, 1995). The ^{137}Cs distribution observed in the present survey is probably due to complex water circulation in the North Sea (North Sea Task Force, 1993).

^{137}Cs concentrations in the western English Channel ranged from 0.001-0.003 Bq l^{-1} with an average concentration of 0.0017 Bq l^{-1} (Figure 1c). These are typical of the background level in surface Atlantic seawater due to global fallout.

1.3.2 ^{3}H distribution

Levels of ^{3}H in the North Sea (Figure 1b) were below the limit of detection (ca. 2 Bq l^{-1}) over most of the survey area. However, slightly enhanced levels were apparent along some of the European coastline. These were likely to be a result of discharges from the La Hague (France) nuclear fuel reprocessing plant.

Detectable ^{3}H concentrations were observed in the Severn estuary near the points of release from the Amersham plc radiopharmaceutical plant at Cardiff and the Hinkley Point nuclear power station (Figure 1d). The greatest ^{3}H concentrations (up to ~30 Bq l^{-1}) were observed on the English side in the vicinity of the Hinkley Point nuclear power station, compared with <5 Bq l^{-1} in the vicinity of the Amersham facility at Cardiff. The impact of the ^{3}H inputs into the Severn estuary was most apparent downstream of both these points of discharge. Detectable concentrations were observed to the eastern limit of the survey area. This is to be expected given the tidal nature of the Severn estuary. Tidal current speeds generally exceed 1.5 m s^{-1} at springs and 0.75 m s^{-1} at neaps, meaning water parcels can move up to 25 km during a flood or ebb tide (Uncles, 1984). Outside of the typical tidal excursion, ^{3}H concentrations decreased rapidly with distance downstream of the points of discharge (i.e. in a westerly direction). Concentrations at the mouth of the Bristol Channel, and in the western English Channel, were below the limit of detection.

1.3.3 Other radionuclides

Concentrations of ^{99}Tc in seawater are now decreasing, following the substantial increases observed since 1994 due to increases in discharges of this radionuclide from Sellafield. The results of research cruises, involving studies of this radionuclide have been published by Leonard *et al*. (1997a and b, 2004) and McCubbin *et al*. (2002). Trends in plutonium and americium concentrations in the seawater of the Irish Sea, have been considered by Leonard *et al*. (1999). A full review of the quality status of the North Atlantic region has been published by OSPAR (2000).

2. PHARMACEUTICAL COMPOUNDS IN UK ESTUARIES

Author: Paul Roberts

2.1 Introduction

The presence of pharmaceutical compounds in the environment is a concern that has grown over recent years. Many of the more commonly used pharmaceutical groups (e.g. antibiotics), are used in quantities similar to those of many agrochemicals and other micro-organic pollutants, but they are not required to undergo the same level of testing for possible environmental effects prior to use. In addition, most pharmaceuticals are designed to be persistent so that they retain their chemical structure long enough to do their therapeutic work. This, coupled with their continual input, may enable them to remain in the environment for a significant period of time.

Questions are now being asked about the potential long-term environmental fate and effects of some pharmaceuticals. Inputs of pharmaceutical compounds into aquatic systems has led to their occurrence being reported in wastewater treatment works (WTW) effluents, rivers and lakes, and (more rarely) in groundwater (Buser *et al*., 1998a and 1999; Stan and Heberer, 1997; Ternes, 1998; Heberer *et al*., 1998; Hirsch *et al*., 1998; Stumpf *et al*., 1999; La Farre *et al*., 2001; Ollers *et al*., 2001; Kolpin *et al*., 2002). Fewer data are available for estuarine or marine waters (Buser *et al*., 1998b; Weigel *et al*., 2002).

Establishing the occurrence of pharmaceutical compounds in the marine environment is essential if the environmental risk by these compounds is to be assessed. For the purposes of this study, the pharmaceutical compounds prioritised by the UK Environment Agency (for establishing the occurrence of pharmaceuticals in STW effluents and rivers (Hilton and Thomas, 2003)) and by the Oslo and Paris Commission (OSPAR) (for the protection of the marine environment of the North East Atlantic – the OSPAR list of substances of possible concern) were selected.

2.2 Methods

Water samples (2.7 l) were collected in silanised, clean amber glass Winchester bottles, following UK National

Marine Monitoring Programme (NMMP) guidelines. The sampling sites were located on the lower reaches of the rivers Tyne, Tees, Mersey, and Thames as well as in Belfast Lough. Samples were stored at < 4°C and extracted within 48 h of collection.

2.3 Analysis

The selected pharmaceutical compounds were isolated from the samples using solid phase extraction (SPE), after the addition of a surrogate standard (^{13}C-phenacetin). Analysis was performed using reversed-phase high-performance liquid chromatography-electrospray tandem mass spectrometry (HPLC-ESI-MS/MS). The limits of detection (LOD) for the extraction and full details of the analytical method are described in detail elsewhere (Hilton and Thomas, 2003; Thomas and Hilton, 2004).

2.4 Results

The concentrations of the selected compounds determined in this study are presented in Table 1. Clotrimazole, a topical antifungal agent, was detected most frequently, being present in 59% of all samples. Clotrimazole concentrations ranged from below the LOD (1 ng l^{-1}) to 22 ng l^{-1}, with a median concentration of 7 ng l^{-1}.

The next most frequently observed compound was ibuprofen, which was detected in 50% of the samples. The concentrations ranged from below the LOD (8 ng l^{-1}) to 928 ng l^{-1}, with a median concentration of 48 ng l^{-1}. These relatively high concentrations are not surprising, as ibuprofen is already known to occur in certain UK rivers receiving STW effluent, (Ayscough *et al*., 2000; Ollers *et al*., 2001; La Farre *et al*., 2001; Kolpin *et al*., 2002; Hilton and Thomas, 2003) with a reported half-life of 50 days (Singer *et al*., 2002). Trimethoprim was the only antibiotic to be detected, and was present in 50% of all samples. Concentrations ranged from below the LOD (4 ng l^{-1}) to 569 ng l^{-1}, with a median concentration of ~5 ng l^{-1}. These concentrations of trimethoprim are consistent with those reported previously (Hirsch *et al*., 1999; Kolpin *et al*., 2002, Hilton and Thomas, 2003). Limited data are available on the environmental fate of trimethoprim, but half-lives of 20-100 days have been reported (Zuccato *et al*., 2001).

Propranolol was detected in 41% of the samples analysed, at concentrations ranging from less than the LOD (4 ng l^{-1}) to 56 ng l^{-1}, with a median concentration of 13 ng l^{-1}. Recent surveys of the occurrence of propranolol in WTW effluents showed the compound to be present at detectable concentrations in all samples collected (n = 45). These data compare well with previously reported values (Hirsch *et al*., 1996; Ternes, 1998).

Diclofenac, mefenamic acid and dextropropoxyphene were detected in 32%, 23% and 14% of the samples analysed respectively. Diclofenac was found at concentrations ranging from less than the LOD (8 ng l^{-1}) to 195 ng l^{-1}, with a median concentration below 8 ng l^{-1}. Previous studies of diclofenac occurrence reported concentrations of 6 ng l^{-1} in the estuary of the River Elbe. These measured environmental concentrations are similar to those reported in other studies (Ayscough *et al*., 2000; Ollers *et al*., 2001; La Farre *et al*., 2001). Mefenamic acid was detected in only five of the samples collected, with concentrations ranging from less than the LOD (20 ng l^{-1}) to 196 ng l^{-1}. These data are comparable with maximum and median mefenamic acid concentrations of 366 and 62 ng l^{-1} reported for samples from UK rivers (Hilton and Thomas, 2003) and approximately 10 ng l^{-1} in samples collected from Austria (Ahrer *et al*., 2001). Dextropropoxyphene was only detected in three of the samples collected at concentrations ranging from less than the LOD (8 ng l^{-1}) to 80 ng l^{-1}.

Tamoxifen was only detected in four of the samples collected. The concentrations ranged from less than the LOD (4 ng l^{-1}) to 71 ng l^{-1}. Clofibric acid was detected in only two of the samples collected at a concentration of approximately 100 ng l^{-1}. Clofibric acid has previously been reported as a contaminant in the marine environment, with reported concentrations ranging between 0.03 and 19 ng l^{-1} in samples collected from the North Sea (Buser *et al*., 1998a; Weigel *et al*., 2002).

Paracetamol, lofepramine, erythromycin, sulfamethoxazole and acetyl-sulfamethoxazole were not detected above the limits of detection of the method used.

2.5 Discussion

This initial study of UK estuaries has shown that residues of some human pharmaceutical compounds are present at measurable concentrations. Of the pharmaceutical compounds and their metabolites measured, clofibric acid, clotrimazole, dextropropoxyphene, diclofenac, ibuprofen, mefenamic acid, propranolol, tamoxifen and trimethoprim were all detected with the methods used. Clotrimazole was the most frequently detected compound, at low ng l^{-1} concentrations. Ibuprofen was detected at the highest concentration (928 ng l^{-1}). In order to fully establish the occurrence of human pharmaceuticals in the aquatic environment a survey of the levels present in sediments needs to be conducted. In addition, more aquatic data are required before the risk that they may present can be assessed.

Table 1. Concentration of selected pharmaceutical compounds in samples collected from UK estuaries

Estuary	Station	Position		Concentration (ng l^{-1})†								
		Latitude	Longitude	Mefenamic acid	Diclofenac	Propranolol	Dextro-propoxyphene	Tamoxifen	Clofibric acid	Ibuprofen	Trimethoprim	Clotrimazole
Mersey	1	53° 26.89'N	3° 1.39'W	34	195	16	<8	13	<20	325	22	6.2
	2	53° 26.17'N	3° 0.51'W	<20	<8	<4	<8	<4	<20	129	<4	<1
	3	53° 20.96'N	2° 57.24'W	39	<8	51	80	<4	<20	342	569	<1
	4	53° 27.83'N	3° 3.39'W	<20	<8	<4	<8	<4	<20	386	6.8	<1
	5	53° 24.63'N	3° 0.74'W	<20	<8	10	13	<4	<20	<8	24	<1
	6	53° 19.93'N	2° 57.14'W	<20	<8	14	<8	<4	<20	287	49	<1
Tyne	1	54° 59.183'N	1° 28.335'W	<20	90	<4	<8	<4	<20	755	46	14
	2	54° 59.182'N	1° 28.303'W	<20	40	<4	<8	<4	<20	698	<4	8.5
	3	54° 59.192'N	1° 28.125'W	<20	<8	11	33	<4	<20	<8	<4	18
	4	54° 57.304'N	1° 38.110'W	<20	<8	<4	<8	<4	<20	<8	<4	<1
	5	54° 58.05'N	1° 36.30'W	<20	<8	<4	<8	<4	<20	<8	<4	7.6
	6	54° 57.591'N	1° 33.401'W	<20	<8	<4	<8	<4	<20	<8	<4	6.8
Thames	1	51° 30.595'N	0° 5.079'E	58	57	<4	<8	<4	98.4	928	<4	<1
	2	51° 30.56'N	0° 8.76'E	104	125	20	<8	<4	<20	<8	<4	11
Belfast Lough	1	54° 38.47'N	5° 52.77'W	196	<8	45	<8	71	111	124	30	19
	2	54° 39.37'N	5° 51.33'W	<20	<8	<4	<8	<4	<20	<8	24	20
	3	54° 40.62'N	5° 47.28'W	<20	<8	56	<8	35	<20	376	32	22
Tees	1	54° 36.86'N	1° 09.04'W	<20	<8	<4	<8	<4	<20	88	<4	<1
	2	54° 36.20'N	1° 09.63'W	<20	33	<4	<8	23	<20	<8	11	7.5
	3	54° 34.81'N	1° 12.76'W	<20	<8	18	<8	<4	<20	<8	<4	3.8
	4	54° 35.10'N	1° 11.59'W	<20	191	<4	<8	<4	<20	<8	17	<1
	5	54° 35.45'N	1° 14.72'W	<20	<8	<4	<8	<4	<20	<8	<4	7.2

† *Following pharmaceutical compounds < limit of detection: Paracetamol <20 ng l^{-1}; Lofepramine <4 ng l^{-1}; Erythromycin <4 ng l^{-1}; Sulfamethoxazole <20 ng l^{-1}; Acetyl-sulfamethoxazole <20 ng l^{-1}.*

3. PRODUCED WATER STUDIES

Author: Jan Balaam

3.1 Introduction

Produced water (PW) is water which is present in the underground formation in which oil and gas are found. It rises up the wells with the oil and gas and is separated from those products on the production platforms, before being discharged to sea. Produced water represents the largest discharge from producing platforms and its volume increases as fields age. In an initial study of the effects of dissolved chemicals in this discharge stream, produced water and surface water samples were collected at increasing distances along transects away from the Statfjord C production platform in the Norwegian sector of the North Sea.

3.2 Methods and results

The samples were extracted and analysed using toxicity identification and evaluation (TIE) methods. Also known as bioassay-directed fractionation, this is a method which is used to identify compounds which are causing effects of various types on organisms. In this case, TIE analysis pointed towards alkylphenols (nonylphenol) as the main source of oestrogenic activity, a form of endocrine disruption, in PW (Thomas *et al*., 2003a).

A further study investigated offshore installations in the UK sector of the North Sea. Eighty-one PW samples were taken and tested for the presence of C_1 – C_9 alkylphenols (Balaam *et al*., in press) and also for oestrogenic potency. C_1-C_9-substituted alkylphenols are naturally produced compounds which are present in the underground oil or gas reservoir and rise to the platform dissolved in the PW and so are then discharged overboard. Comparison of these data showed little correlation between total alkylphenol content and oestrogenic potency (Thomas *et al*., 2003b; 2004a, b; Balaam *et al*., 2003). C_1-C_5 alkylphenols were identified and quantified, at concentrations ranging from 5 to 1600 ng l^{-1}. Thirty-nine different isomers were identified in total. Samples ranged from having just one alkylphenol compound identified, to having 19 different isomers, with an average of 10 isomers found per sample. Oestrogenic potency, expressed as equivalents of oestradiol, ranged from below the limit of detection (10 ng E2 equivalents l^{-1}) to 91 ng E2 equivalents l^{-1}. No alkylphenols with chain length greater than C_5 were found. This is consistent with predictions based on oil-water-rock partition coefficients (Taylor *et al*., 1997; Faksness *et al*., 2004). Aryl hydrocarbon receptor agonist potency was also characterised for samples from these 81 installations (Hurst *et al*., in preparation). Aryl hydrocarbon potency, in this case expressed as toxic equivalents of the most potent dioxin compound, 2,3,7,8-tetrachlorodibenzo-*p*-dioxin as determined by a standardised bioassay, ranged from 1 to 430 ng TCDD TEQ_{CALUX} l^{-1}.

Following this work, and in order to further characterise the potential effects of PW, a further TIE study was carried out on PW samples from 16 installations in the UK sector of the North Sea. These 16 installations were selected using data from the previous studies, based on the highest discharge volume, highest oestrogenic potency and highest aryl hydrocarbon receptor agonist potency. The samples were then tested using a range of bioassays; YES assay (for oestrogenic receptor agonist potency), CALUX assay (for aryl hydrocarbon receptor agonist potency), *Tisbe battagliai* assay (for acute toxicity), *Skeletonema costatum* assay (for algal toxicity) and oyster embryo (*Crassostrea gigas*) bioassay (for bivalve larval development). Preliminary assay data are available (Balaam *et al*., 2004; Thomas and Balaam, 2004), and the most potent sample identified using each assay will now be subjected to a TIE type study in order to elucidate the causative compounds.

BIOTA

4. CONTAMINANTS IN MARINE MAMMALS

Author: Robin Law

4.1 Introduction

At the time of the phocine distemper outbreak in seals around European coasts in 1988, concern was expressed that high contaminant burdens may be exacerbating the impact of the disease and increasing mortality. Currently, there is widespread concern that a number of persistent marine pollutants may pose a significant and global threat to the health status and viability of marine mammal populations, with persistent organohalogen contaminants such as polychlorinated biphenyls (PCBs) raising the greatest concern (see e.g. Colborn and Smolen, 1996). The UK Marine Mammals Strandings Programme is funded by Defra as one component of their commitment to the ASCOBANS (Agreement

on Conservation of Small Cetaceans in the Baltic and North Seas) treaty and other international conservation agreements. Within this programme, one strand of the studies has been directed towards investigating possible links between contaminants and infectious disease mortality. This work has concentrated on the common (harbour) porpoise (*Phocoena phocoena*) as it is the most abundant cetacean in northern European waters and is the most frequently stranded cetacean in the UK. In addition, earlier studies have demonstrated high levels of a range of contaminants in porpoises from UK waters, and also demonstrated associations between concentrations of some of these contaminants and infectious disease mortality (Bennett *et al*., 2001; Jepson *et al*., 1999).

4.2 Methods

Between July 1989 and December 2002, 1061 harbour porpoise carcasses from stranded or bycaught animals were taken for postmortem study. Detailed pathological investigations followed a standard protocol (Law, 1994), which also involved the collection of tissue samples for contaminant analysis. Blubber samples were analysed for a range of organochlorine pesticides and chlorobiphenyl (and, in later years, brominated diphenyl ether (BDE)) congeners, and liver samples for a range of trace elements and butyltin compounds, following standardised protocols and with associated quality assurance procedures.

For statistical treatment, the porpoises were divided into two groups based on the cause of death as determined during postmortem investigation: individuals diagnosed to have died due to acute physical trauma or bycatch were included in the physical trauma category (n = 175), whilst those which died due to disease caused by one or more infectious agents were assigned to the infectious disease category (n = 82). Animals for which no cause of death could be established were excluded. Full details of the procedures and of the numbers of animals assigned to each category by contaminant group are given in Jepson *et al*. (2005). A number of potentially confounding variables (including age, sex, nutritional status, region of stranding, and year of stranding) were also tested within the statistical treatment.

4.3 Results

The infectious disease group was found to have significantly higher concentrations of chlorobiphenyls (as the sum of the 25 CB congeners determined) than the physical trauma group. Mean concentrations were 27.6 and 13.6 mg kg^{-1} on a lipid basis, respectively. The summed CB congener concentrations were also recalculated on a formulation basis as equivalents of Aroclor 1254, one of the commercial PCB products which are now banned, using a factor of 3.0 x sum ICES7 CB congeners derived from other studies in fish. This was to allow comparison with a proposed threshold level of adverse health effects in marine mammals derived from experimental studies (mostly in rats and mink) of 17 mg kg^{-1} lipid (Kannan *et al*., 2000). For animals with blubber PCB concentrations below 17 mg kg^{-1} lipid (n = 103), no significant differences were observed between the two groups. However, for animals with blubber PCB concentrations above 17 mg kg^{-1} lipid (n = 153), the associations between PCB concentrations and infectious disease mortality were highly significant. Also, this association was not confounded by the other variables tested. These findings are consistent with, but do not conclusively prove, a causal relationship between PCB exposure and a predisposition to infectious disease mortality, for instance due to immunosuppression. Future studies will try to apply a probabilistic risk assessment approach (Schwacke *et al*., 2002), so as to model impacts at a population level, and allow the effects of contaminant burdens to be assessed in relation to other population threats, such as fisheries bycatch.

5. THE CONCENTRATIONS OF MERCURY IN FISH TAKEN FROM LIVERPOOL BAY, UK IN 2002

Author: Andrew Franklin

5.1 Introduction

Mercury levels in commercial fish species taken from UK coastal waters have been monitored by the Burnham Laboratory since the 1970s (e.g. Portmann, 1979). Early results indicated that the highest concentrations were found in four areas, Liverpool Bay, Morecambe Bay and Swansea Bay (all of which received discharges containing mercury from the chlor-alkali industry) and in the outer Thames Estuary where a significant input was via sewage sludge disposal. Annual monitoring of these four areas commenced in the early 1980s, following the adoption in 1980 by the Paris Commission of a mercury Environmental Quality Standard (EQS). This required that, in areas receiving significant mercury inputs, the concentration of mercury in a representative sample of fish flesh (chosen as an indicator) should not exceed 0.30 mg kg^{-1} on a wet weight basis. Monitoring soon indicated that the concentrations of mercury in Swansea Bay and the Thames Estuary were no longer elevated to a level that could potentially exceed the EQS and in 1985 it

was agreed that monitoring in these areas could cease. Mercury concentrations remained relatively high at this time in Liverpool and Morecambe Bays and results continued to be reported to OSPAR until 1994 (though at two yearly intervals from 1990), when it was agreed that the requirement for regular reporting could cease. Some re-assurance monitoring has been undertaken since that time and the results from the most recent survey of fish from Liverpool Bay, carried out in 2002, are summarised in Table 2.

Table 2. Liverpool Bay - Mercury concentrations found in fish muscle in 2002

Former OSPARCOM JMP guideline categories;-

"lower" level; <0.1 mg kg^{-1} wet weight
"medium" level ; 0.10 – 0.30 mg kg^{-1} wet weight,
"upper" level ; >0.30 mg kg^{-1} wet weight,

Species	Number of fish analysed	Mean length (cm)	Mean mercury concentration in fish muscle (mg kg^{-1} wet weight)
Cod	13	28.1	0.08
Whiting	25	29.5	0.23
Dab	50	23.8	0.25
Flounder	0*	-	-
Plaice	75	27.6	0.16
Sole	50	23.0	0.15
		Mean, all fish	**0.17**

** None caught in Liverpool Bay in 2000 or 2002*

The initial representative samples of fish flesh were taken from six species, but one of these, flounder, has become increasingly difficult to obtain in Liverpool Bay in recent years. In 2002 none were caught, despite effort on two separate cruises, so the mean mercury concentration in 2002 is for the five fish species, cod, whiting, dab, plaice and sole.

The concentration of mercury in individual fish species is now very much less than the limit of 0.50 mg kg^{-1} set in European Community Decision 93/351/EEC on maximum limits for mercury in fishery products. The overall mean concentration in the flesh of the fish sampled was 0.17 mg kg^{-1}, so reassurance can continue to be given that the mercury EQS will not be breached.

Table 3. Liverpool Bay - Time series of mean concentrations of mercury in fish flesh

Year	Concentration of mercury in mg kg^{-1} wet weight
1981	0.28
1982	0.27
1983	0.28
1984	0.31
1985	0.25
1986	0.22
1987	0.22
1988	0.21
1989	0.20
1990	0.19
1992	0.17
1994	0.17
1996	0.16
1998	0.17
2000	0.16
2002	0.17

Although the data were not collected for time trend purposes, (for which a separate study using more stringent sampling procedures has been attempted), the time-series of results obtained from Liverpool Bay over the 1981-2002 period is given in Table 3. (As flounder have been scarce or absent since the mid 1990s, results for this species have now been excluded from the whole table.)

The time-series indicates that a reduction in overall mercury fish flesh concentrations took place during the 1980s, but levels appear to have remained fairly constant, within the previous OSPAR JMP 'medium' concentration category (Table 2), since the early 1990s. This is likely due to historical contamination, as mercury inputs from the UK to the OSPAR maritime area have continued to decline (OSPAR, 2004).

6. THE BENTHIC ECOLOGY OF THE WESTERN NORTH SEA

Authors: Rebecca Smith, Hubert Rees, Jacqueline Eggleton, Claire Mason, Peter Kershaw

6.1 Introduction

During 2000-2002, 91 stations in the North Sea were sampled for benthic macrofauna and sediment (Figure 2). CEFAS work involved the re-sampling of stations off the English east coast occupied in 1986 as part of the ICES North Sea Benthos Survey (Künitzer *et al*., 1992). Samples for five of these stations were kindly contributed by the Senckenberg Institute, Germany. Work undertaken by the FRS MLA involved stratified random sampling of the northern and eastern North Sea as part of an EU project 'Managing Fisheries to Conserve Groundfish and Benthic Invertebrate Species Diversity' (MAFCONS).

In 2002, Defra funded the work-up of these samples as a contribution both to the UK '*Charting Progress*' report and to an international collaborative initiative to re-appraise the status of the North Sea benthos

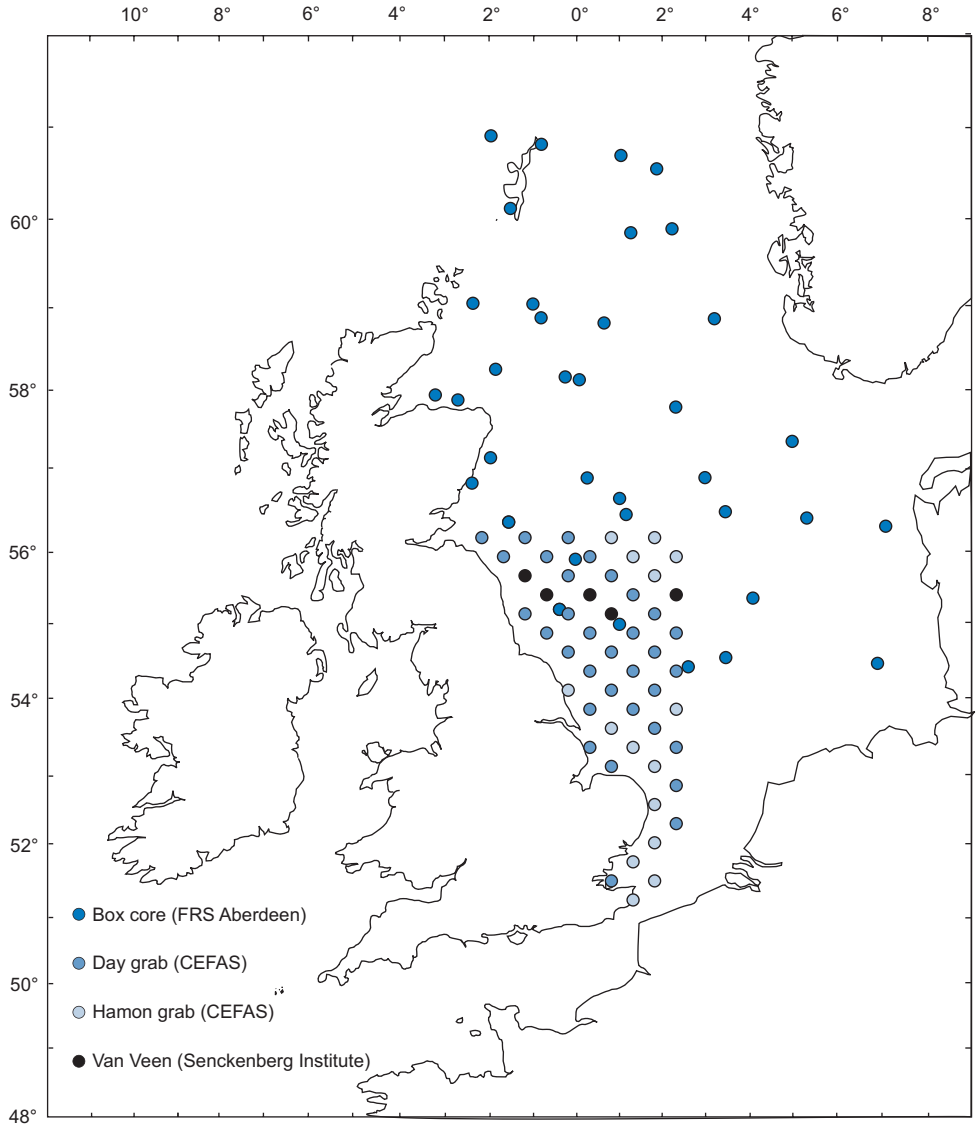

Figure 2. Locations of stations, principally in the western North Sea, sampled in the period 2000-2002

following an earlier (1986) survey, under ICES auspices (ICES, 2004). The present draft summary relates to the first of these aims.

CEFAS stations were sampled using a 0.1 m^2 Hamon grab or Day grab; Senckenberg Institute stations were sampled with a 0.1 m^2 van Veen grab; FRS MLA stations were sampled with a 0.25 m^2 box corer. A 1 mm mesh sieve was used in the processing of all benthos samples. Sediment sub-samples collected by CEFAS and FRS MLA were analysed for particle size. Additionally, sub-samples at CEFAS stations were analysed for % organic carbon and nitrogen content and the concentrations of a range of trace metals.

6.2 Summary

A range of univariate and multivariate statistical techniques were applied to the biological and environmental data. Preliminary findings are as follows:

There is no evidence of major structural change in the benthic communities of the western North Sea between 1986 and 2000. Differences in univariate measures were not significant, with the exception of measures of evenness, indicating increased dominance of common species in 2000 (e.g. *Spiophanes bombyx*).

Spatial variations in the numbers of species and densities of the benthic macrofauna are mainly accounted for by a trend towards increasingly fine substrata, increasing water depths and reduced tidal current strengths from south to north. Species-rich assemblages associated with gravelly substrata along parts of the southern English coast provide a notable exception to this general trend. Other environmental factors associated with this latitudinal gradient may be important, but are difficult to isolate due to confounding influences.

Biomass values from the 2000 survey show an increase in biomass from south to north, with polychaete worms dominating overall. Comparison of the distributions and percentage contributions of the major groups in 2000 with 1986 data indicate that the distribution of echinoderm species has moved northwards and densities are reduced in the area off north Norfolk. Mollusc biomass has also reduced in the offshore central North Sea between 1986 and 2000.

Changes in the distributions or densities of individual species are apparent between earlier (1986) and recent sampling effort and are the subject of ongoing investigation into the probable causes.

Concentrations of a range of trace metals in offshore sediments provide little indication of levels of contamination that would have an impact on the distribution and densities of benthic species at the scale of survey effort employed in the 2000 survey. This is confirmed by the absence of any correlation between contaminant levels and benthic assemblages.

The distribution patterns of the different trace metals in sediments are not consistent and comparison of the North Sea data with small-scale survey data from the NMMP. and FEPA disposal at sea survey programmes also shows few consistent patterns.

The concentration of trace metals obtained in the present survey generally exceeded Ecotoxicological Assessment Criteria levels. However, the analyses were conducted on the <63 µm fraction, representing generally <10% of the particle size range, and the mean concentrations for the whole sediment are likely to be much lower. It is not thought that these levels, when considering the probable whole sediment concentration, will have a significant impact on biota.

The absence of any 'footprint' associated with oil and gas installations (the most widely distributed anthropogenic activity, other than demersal fishing) adds weight to the view that adverse effects on the benthic macrofauna from this activity remain very localised in extent. It follows that, similarly, there is no evidence from preliminary observations of large-scale cumulative consequences arising from oil and gas exploitation, or from the activities of aggregate extraction and dredged material disposal.

The distribution of commercial beam trawling effort is confounded with latitude, and hence with natural environmental factors such as substratum type and depth, as well as with trends in the benthic macrofauna. There is no evidence of any causal relationship between the distribution of benthic assemblages and this activity, although more subtle effects cannot be dismissed and will be further explored by reference to historical information.

Next steps are the inclusion of site-specific biological survey data from the NMMP and FEPA programmes to assess, respectively, the representativeness of stations and the influence of spatial scale on evaluations of anthropogenic impacts.

Further detailed analyses of the western North Sea data to identify possible reasons for the differences in species distribution between 1986 and 2000 is also in progress with the aim of providing additional insights into the causes of these observed patterns and changes over time.

A full report of this study was published at the end of December 2004.

7. FISH PATHOLOGY AND DISEASE BIOMARKERS 2002-2003

Authors: Stephen Feist and Grant Stentiford

7.1 Introduction

The level of disease within a population provides an excellent indicator of the general health status of that population and has been used as a measure of environmental stress. As such, the study of fish diseases and liver histopathology have been included in national marine and estuarine monitoring programmes for many years (Lang and Dethlefsen, 1996). Within the UK NMMP, the disease status of dab (*Limanda limanda*) at offshore sites and flounder (*Platichthys flesus*) at inshore and estuarine sites are monitored. The externally visible diseases present in the target species include lymphocystis, epidermal hyperplasia and papilloma, acute and healing ulcerations and hyperpigmentation. Internally, the condition of the liver is recorded, especially with regard the presence of macroscopic tumours.

The aetiology (or cause) of the various diseases is not known in all cases. However, it is known that lymphocystis is caused by an iridovirus and virus-like particles have been found in fish with epidermal papillomas, but the latter association has not been demonstrated experimentally. The presence of ulcers is often associated with general stress conditions and, invariably, a diverse microbial fauna is associated with the damaged tissues. The initial causes are likely to be multifactorial, including trauma. Although the pathological features of hyperpigmentation have been established, the aetiology of the condition remains unknown. Taken together, these external disease conditions, as well as information on the parasite burdens, provide an indicator of the health of the populations sampled. Such data are used to detect long-term trends in disease prevalence and, in combination with other biomarkers, are used to provide greater confidence in the use of fish diseases as indicators of contaminant effects (Lang and Dethlefsen, 1996; Wosniok *et al*., 2000).

The presence of liver tumours (neoplasms) in flatfish are a more direct indicator of contaminant effect and have also been used for many years in environmental monitoring programmes around the world (Myers *et al*., 1987, 1992, 1998; Stein *et al*, 1990; Vethaak and Wester, 1996; Stentiford *et al*., 2003). The occurrence of pre-neoplastic lesions such as 'foci of cellular alteration' and benign and malignant tumours have been linked with exposure to organic contaminants. The current monitoring programme includes measures of genotoxic damage at the DNA level (formation of DNA adducts) and markers of genotoxin exposure (EROD) as well as levels of contaminant metabolites in the bile.

The fish disease monitoring programme also seeks to identify conditions in the target species that may be added to those used routinely. In addition, the programme also aims to provide an assessment of the health status of commercial fish species, particularly in order to identify diseases that threaten specific populations, or that render fish unsightly and unmarketable.

This section presents the data on fish disease detected in various species from the North Sea, Irish Sea and English Channel and for the first time provides an assessment of the association between disease status and geographical location. In addition, it provides information on the principal diseases that can be used to discriminate discrete sites from each other.

7.2 Materials and methods

Monitoring was undertaken as part of the integrated biological effects monitoring cruises that take place annually during the summer (June and July). In 2002 and 2003, a total of nineteen and eighteen sites respectively were assessed for external fish disease and the presence of macroscopic liver nodules. In 2002, nineteen sites and, in 2003, eighteen sites, were sampled specifically for liver pathology (For site details see Figure 3).

Sampling protocols followed those established by ICES (Bucke *et al*., 1996) for external diseases. Target species were dab and cod (*Gadus morhua*) for offshore sites and flounder at inshore locations. Where sufficient numbers of other species were caught a disease assessment was undertaken. Species sampled included plaice (*Pleuronectes platessa*), Dover sole (*Solea solea*), haddock (*Melanogrammus aeglifinus*), herring (*Clupea harengus*), whiting (*Merlangius merlangus*), turbot (*Scophthalmus maximus*), brill (*Scophthalmus rhombus*), lesser spotted dogfish (*Scyliorhinus caniculus*), and rays (*Raja clavata*). In addition, four-bearded rockling (*Rhinonemus cimbrius*) were examined for the presence of liver tumours since this species has shown this condition during previous surveys.

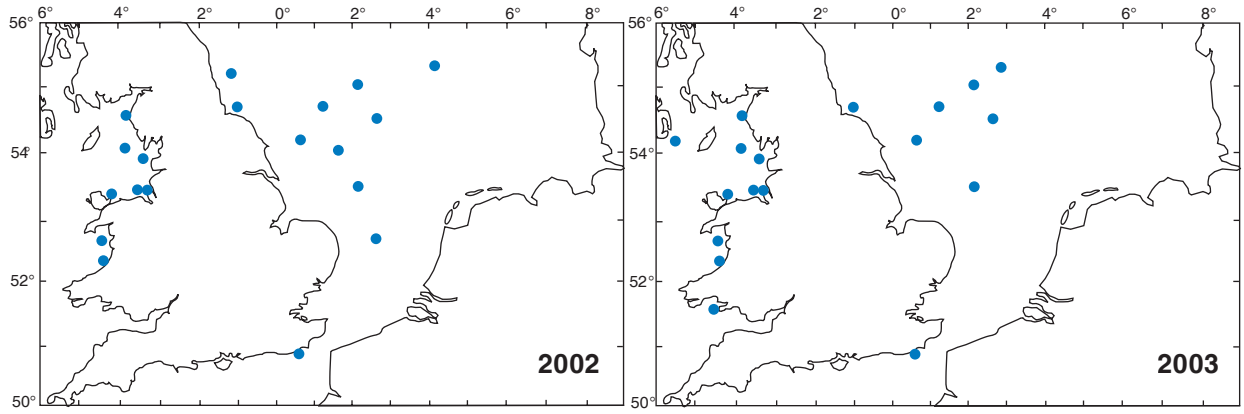

Figure 3. Locations sampled over 2002-2003

From all dab examined for external disease that harboured liver nodules greater than 2mm in diameter, a section of the liver incorporating the suspected tumour was taken for confirmatory histological analysis. Samples were fixed in neutral buffered formalin for between 24 and 48 hours and stored in 70% alcohol prior to further processing. Samples for other fish species requiring histological confirmation were treated similarly. In addition, standard sections of liver and gonad tissue were sampled from 50 dab greater than 20 cm in length for the assessment of pathological changes. At each site sampled, the first 20 fish were also sampled for other biomarkers (see Sections 10 and 11). The otoliths were also removed from each of these fish for age assessment (data not reported here). Histological methods and diagnostic criteria followed those developed by ICES and were undertaken according to the quality assurance requirements required under the Biological Effects Quality Assurance in Monitoring (BEQUALM) programme (Feist *et al*., 2004). Pathological changes to the liver are presented here under the broad categories of 1) non-specific inflammatory lesions, 2) non-neoplastic toxicopathic lesions, 3) foci of cellular alteration, 4) benign neoplasms and 5) malignant neoplasms. Fish displaying no liver pathology are reported as 'No Abnormalities Detected'.

Multivariate statistics using PRIMER software (Clarke and Warwick, 2001) were applied to the data since this approach provides increased sensitivity for the detection of differences in disease patterns between years compared to univariate analyses. Cluster analysis (Multi-dimensional scaling - MDS) was employed to identify site associations according to the similarity of the disease status in dab at those locations. Principal components analysis was used to determine the main diseases responsible for site discriminations. Fourteen sites sampled in both 2002 and 2003 were included in the PRIMER analysis of external diseases. Seventeen sites sampled in both 2002 and 2003 were included in the analysis of liver pathology. Additional analyses were undertaken using long-term data from Liverpool Bay between 1994 and 2004 in order to examine disease trends over that period.

7.3 Results

7.3.1 Dab diseases

Disease prevalence and severity data for dab sampled in 2002 and 2003 according to size group are presented in Tables 4 and 5. Overall, disease levels were broadly similar in most areas sampled to those recorded in previous years (CEFAS, 1998 and 2000). Dab captured at Flamborough and the Dogger Bank continue to exhibit higher levels of disease (including macroscopic liver tumours) than at the Rye Bay reference site and the prevalence of hyperpigmentation remains higher in dab from the North Sea, with only low levels being recorded from most Irish Sea stations. However, disease levels, including hyperpigmentation appear to be increasing at the Cardigan Bay sites.

The MDS ordination plot for 2002 clearly discriminates a group of sampling locations from the Irish Sea (sites 1, 6, 7, 10 and 14) from a separate cluster of inshore North Sea sites, Rye Bay, North Cardigan Bay and SE Isle of Man (Figure 4). This pattern appears to be driven by the higher prevalence of skin ulceration at the former sites. A more marked discrimination is apparent between these two groups and locations on the Dogger Bank and Off Flamborough. The west Dogger location being isolated with the principle component influencing this being skin hyperpigmentation.

The MDS ordination plot for 2003 reveals a similar pattern with principal components of skin ulceration and hyperpigmentation influencing the distribution (Figure 5). Comparison of the two datasets confirmed a high statistical similarity between the MDS ordination plots for 2002 and 2003 (Rho = 0.83, $p<0.01$), where a Rho value of 1 would indicate a perfect match between years. A similar approach was used to investigate changes in disease prevalence at Liverpool Bay between 1994 and 2004. MDS ordination plots showing the discrimination between years based on the principal components of external diseases and gross liver nodules is given in Figure 6.

Table 4. Summary catch data and disease prevalence in dab (Limanda limanda) by size category and disease severity on stations sampled during 2002

Station no.	Location (NMMP)	Size (cm)	M	F	No. and severity of disease cases Recorded in according to ICES Guidelines (Bucke *et al.*, 1996)																Total examined
					LY			U			EP			HYP			LN	X	ST	LP	
					1	2	3	1	2	3	1	2	3	1	2	3					
1	Inner Cardigan Bay	15-19	87	28	7	0	0	5	1	1	3	1	0	16	6	2	4	0	16	51	
	(656)	20-24	1	5	0	0	0	1	0	0	0	0	0	1	0	0	0	0	0	4	121
		25>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
100	North Cardigan Bay	15-19	106	105	1	0	0	6	1	2	2	1	0	2	1	0	0	0	11	8	
	(NA)	20-24	16	120	2	0	0	3	2	2	3	0	0	7	2	2	14	0	7	33	351
		25>	0	4	0	0	0	0	0	0	1	0	0	0	0	0	1	0	0	1	
19	Red Wharf Bay	15-19	134	66	1	0	0	9	2	0	2	0	0	1	0	0	0	0	14	6	
	(776)	20-24	27	131	3	0	0	13	5	1	5	0	0	3	1	0	6	0	2	4	386
		25>	0	28	0	0	0	5	3	2	2	0	0	0	0	0	3	0	0	0	
27	North Liverpool Bay	15-19	68	33	3	0	0	7	2	1	2	0	0	0	0	0	0	0	5	0	
	(715)	20-24	52	86	4	1	0	13	3	3	10	0	0	3	1	0	6	0	4	3	272
		25>	0	33	2	0	0	4	2	2	2	1	0	0	0	0	1	0	1	2	
44	Burbo Bight	15-19	73	32	2	0	0	9	4	1	2	0	0	0	0	0	0	0	3	3	
	(705)	20-24	27	104	2	0	0	13	4	6	5	0	0	1	0	0	9	0	4	22	269
		25>	0	33	1	0	0	5	2	6	1	0	0	1	0	0	7	0	1	9	
78	St Bees	15-19	78	25	5	0	0	6	0	0	2	0	0	0	0	0	0	0	62	25	
	(768)	20-24	47	83	3	0	0	10	2	4	3	0	0	0	0	0	10	0	41	51	256
		25>	1	22	1	0	0	4	0	0	1	0	0	0	0	0	1	0	10	15	
91	Off Morecambe Bay	15-19	126	80	5	0	0	21	6	1	1	0	1	0	0	0	0	0	24	12	
	(796)	20-24	32	200	1	0	0	21	3	2	2	0	0	0	0	0	4	0	7	50	469
		25>	0	31	0	0	0	5	0	1	0	0	0	0	0	0	0	0	1	10	
123	Rye Bay	15-19	70	33	1	0	0	2	3	0	0	0	0	1	1	0	2	0	1	19	
	(486)	20-24	42	98	2	0	0	2	2	3	0	0	0	3	0	0	1	0	4	44	264
		25>	1	20	0	0	0	0	0	0	0	0	0	0	0	0	0	0	4	10	
165	Off Humber	15-19	35	71	3	0	0	2	0	1	0	0	0	0	0	0	1	0	11	5	
	(346)	20-24	14	119	4	3	0	2	2	5	5	0	0	4	2	0	2	0	37	2	255
		25>	0	16	2	0	0	0	0	1	0	0	0	1	0	0	3	0	1	1	
90	South East Isle of Man	15-19	77	29	7	0	0	2	0	1	2	0	0	0	0	0	0	0	31	13	
	(805)	20-24	8	6	1	0	0	0	0	0	1	0	0	0	0	0	0	0	10	3	120
		25>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
168	Flamborough	15-19	51	54	1	0	0	2	0	1	4	0	0	5	1	0	1	0	24	1	
	(344)	20-24	26	75	5	1	0	1	0	1	2	0	0	16	6	1	2	0	18	0	218
		25>	2	10	2	0	0	2	0	0	1	0	0	3	1	0	1	0	0	0	
171	Tees Off	15-19	47	55	4	0	0	1	0	3	1	0	0	1	0	0	0	0	33	0	
	(295)	20-24	23	95	11	0	0	1	0	3	3	1	0	2	0	0	1	0	31	2	220
		25>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
192	Amble	15-19	65	36	4	2	0	1	0	0	2	0	0	1	0	0	1	0	48	0	
	(244)	20-24	24	26	4	0	0	1	0	0	0	0	0	1	0	0	0	0	15	0	152
		25>	0	1	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
211	North Dogger	15-19	51	49	4	0	0	4	1	5	3	1	0	8	3	0	0	0	57	0	
	(NA)	20-24	43	72	3	1	0	14	9	10	1	0	0	18	1	3	3	0	51	3	335
		25>	11	109	6	1	0	8	5	12	1	0	0	12	9	7	12	0	6	1	
215	Indefatigable Bank	15-19	34	66	0	0	0	1	2	1	0	0	0	0	0	0	4	0	13	15	
	(NA)	20-24	9	91	1	0	0	2	0	3	0	1	0	4	2	0	2	0	11	18	237
		25>	0	37	0	0	0	3	1	1	1	1	0	2	1	2	2	0	0	9	

Table 4. continued.

Station no.	Location (NMMP)	Size (cm)	M	F	No. and severity of disease cases Recorded in according to ICES Guidelines (Bucke *et al.*, 1996)																Total examined
					LY			U			EP			HYP			LN	X	ST	LP	
					1	2	3	1	2	3	1	2	3	1	2	3					
213	Hospital Ground (Central Dogger)	15-19	59	42	3	0	0	5	7	12	0	0	0	4	2	0	1	0	45	3	
	(287)	20-24	31	101	2	0	0	8	7	3	1	0	0	11	7	4	3	0	14	13	306
		25>	4	69	1	0	0	7	4	9	1	0	0	8	3	7	7	0	1	13	
206	West Dogger	15-19	59	44	6	0	0	4	0	2	1	1	0	13	4	1	1	0	61	1	
	(286)	20-24	64	92	10	3	0	8	3	14	3	2	0	32	11	11	15	0	66	2	289
		25>	0	30	1	1	0	2	1	3	0	0	0	10	1	4	5	0	2	0	
209	East Dogger	15-19	63	36	7	0	0	7	1	2	0	0	0	4	0	0	1	0	68	1	
	(NA)	20-24	42	75	5	1	0	13	13	5	2	0	0	1	0	2	4	0	18	1	283
		25>	1	66	6	1	0	11	3	2	2	0	0	3	1	0	1	0	2	0	
217	Smiths Knoll	15-19	41	48	0	0	0	1	1	0	0	0	0	0	0	0	0	0	2	1	
	(395)	20-24	17	24	0	0	0	0	0	0	1	0	0	0	0	0	0	0	0	3	142
		25>	2	10	0	0	0	0	0	0	0	0	0	1	0	0	0	0	0	1	

Key: LY = Lymphosystis
U = Epidermal ulceration
EP = Epidermal papilloma
HYP= Hyperpigmentation
LN = Macroscopic liver lesion
X = x-cell disease
ST = Stephanostomum
LP = Lepeopthierius pectoralis
AC = Acanthochondria
NM = Nematodes
GL = Glugea

Table 5. Summary catch data and disease prevalence in dab (Limanda limanda) by size category and disease severity on stations sampled during 2003

Station no.	Location (NMMP)	Size (cm)	M	F	No. and severity of disease cases Recorded in according to ICES Guidelines (Bucke *et al.*, 1996)																			Total examined
					LY			U			EP			HYP			LN	X	ST	LP	AC	NM	GL	
					1	2	3	1	2	3	1	2	3	1	2	3								
128	Burbobight	15-19	68	47	1	0	0	7	3	3	0	0	0	0	0	0	1	0	4	41	2	1	11	
	(705)	20-24	23	104	0	0	0	11	2	6	3	0	0	0	0	0	2	0	2	63	4	2	20	275
		25>	0	33	0	0	0	3	1	4	0	0	0	0	0	0	2	0	0	22	2	2	8	
70	Carmarthen Bay	15-19	21	18	0	0	0	4	1	0	0	0	0	0	0	0	0	0	0	1	0	0	0	
	(NA)	20-24	42	44	2	0	0	5	7	3	3	1	0	1	0	0	0	0	1	6	1	5	8	176
		25>	1	50	2	0	0	1	2	1	3	0	0	4	0	1	3	0	1	14	1	2	6	
93	Dundrum Bay	15-19	65	32	1	0	0	1	4	3	1	0	0	14	0	0	3	0	10	10	0	3	0	
	(815)	20-24	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	97
		25>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	
164	Flamborough	15-19	62	54	7	0	0	0	0	3	1	0	0	8	1	0	0	0	29	0	0	95	1	
	(344)	20-24	58	73	7	0	0	2	0	2	3	0	0	27	6	2	1	0	23	3	0	123	0	257
		25>	0	10	0	0	0	0	0	2	0	0	0	3	2	0	2	0	0	0	0	9	0	
152	Hospital Ground (Central Dogger)	15-19	56	44	2	0	0	6	1	10	1	1	0	13	4	2	0	0	46	5	0	7	3	
	(287)	20-24	20	80	1	0	0	7	1	12	3	1	0	25	7	1	3	0	43	9	0	21	6	229
		25>	2	27	1	0	0	2	0	4	0	0	0	11	3	2	4	0	4	8	0	9	1	
1	Indefatigable Bank	15-19	76	124	2	0	0	1	1	1	1	0	0	5	1	3	0	0	9	0	0	0	0	
	(NA)	20-24	18	148	3	0	0	4	2	4	5	0	0	14	3	6	1	0	1	2	0	10	3	435
		25>	1	68	1	0	0	1	0	6	3	1	0	12	5	2	6	0	0	3	1	21	3	
83	Inner Cardigan Bay	15-19	142	58	1	0	0	4	1	0	4	0	0	5	3	0	15	0	2	1	2	3	5	
	(656)	20-24	0	3	0	0	0	1	0	0	0	0	0	0	0	0	1	0	0	0	0	0	0	203
		25>	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	

Table 5. continued.

Station no.	Location (NMMP)	Size (cm)	M	F	No. and severity of disease cases Recorded in according to ICES Guidelines (Bucke *et al*., 1996)																		Total examined
					LY			U			EP			HYP			LN	X	ST	LP	AC	NM	GL	
					1	2	3	1	2	3	1	2	3	1	2	3								
113	Inner Liverpool Bay	15-19	56	18	1	0	0	2	1	3	1	0	0	0	0	0	0	0	16	4	0	1	9	
	(715)	20-24	49	70	1	0	0	7	2	6	2	0	0	0	0	0	9	0	9	26	6	3	14	291
		25>	2	96	4	0	0	3	0	4	1	0	0	1	0	0	11	0	3	26	4	5	9	
102	Off Morecambe Bay	15-19	57	61	1	0	0	6	2	2	1	1	0	4	0	0	0	0	40	16	0	0	0	
	(796)	20-24	10	118	1	0	0	5	5	1	3	0	0	9	0	0	0	0	16	48	0	0	7	286
		25>	0	40	0	0	0	3	3	2	2	0	0	1	0	0	1	0	0	26	0	1	8	
150	North East Dogger	15-19	62	38	5	0	0	14	4	5	0	0	0	2	0	1	0	1	37	1	4	0	0	
	(NA)	20-24	34	66	7	0	0	16	7	8	1	0	0	13	2	1	1	0	43	3	1	16	2	300
		25>	10	90	5	1	0	7	4	4	2	0	0	11	1	2	6	0	21	5	1	25	2	
148	North Dogger	15-19	57	46	5	0	0	17	3	6	3	0	0	17	3	3	1	0	56	4	0	12	0	
	(NA)	20-24	34	93	9	0	0	17	2	14	3	0	0	36	9	6	7	0	51	7	1	28	1	349
		25>	9	110	2	0	0	25	1	14	4	0	0	43	11	6	9	0	22	16	1	29	2	
174	Off Tees	15-19	50	50	7	0	0	2	0	2	1	0	0	4	3	0	0	0	46	1	0	72	1	
	(295)	20-24	18	82	7	0	0	2	0	1	2	0	0	2	0	0	1	0	35	1	1	77	0	202
		25>	0	2	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	1	0	2	0	
104	Red Wharf Bay	15-19	55	66	0	0	0	0	2	0	0	0	0	0	0	0	0	0	11	16	1	0	2	
	(776)	20-24	18	108	1	0	0	4	1	4	0	1	0	0	0	0	0	0	13	39	6	1	4	272
		25>	0	25	1	0	0	2	1	1	0	0	0	0	0	0	0	0	2	14	4	1	0	
86	Roker Park	15-19	64	83	0	0	0	6	0	1	4	1	0	1	0	1	1	0	6	9	0	4	4	
	(649)	20-24	5	66	1	0	0	3	1	2	7	0	0	3	2	0	5	0	2	9	0	3	3	220
		25>	0	2	0	0	0	0	0	0	0	0	0	0	1	0	0	0	0	1	0	0	0	
36	Rye Bay	15-19	136	64	1	0	0	5	3	1	0	0	0	2	0	0	0	0	1	0	0	0	0	
	(486)	20-24	60	140	1	0	0	8	2	2	1	0	0	2	0	1	0	0	0	15	0	2	3	450
		25>	6	44	1	0	0	5	3	1	2	0	0	6	1	0	1	0	0	6	0	6	2	
97	South East Isle of Man	15-19	137	62	3	0	0	3	0	1	1	0	0	0	0	0	0	0	62	4	0	0	0	
	(805)	20-24	25	48	0	0	0	1	4	3	3	0	0	3	2	0	0	0	3	11	0	2	2	280
		25>	0	8	0	0	0	0	0	1	1	0	0	4	0	1	0	0	0	2	0	2	0	
134	St Bees	15-19	65	38	2	0	0	0	0	3	5	0	0	0	0	0	1	0	92	40	0	1	0	
	(768)	20-24	14	19	0	0	0	2	1	1	0	0	0	1	0	0	1	0	21	9	0	0	2	148
		25>	1	11	0	0	0	0	1	1	0	0	0	0	0	0	0	0	3	5	0	0	0	
146	West Dogger	15-19	72	39	11	0	0	7	1	3	3	0	0	33	4	4	2	0	80	1	1	14	4	
	(286)	20-24	58	76	7	0	0	11	2	9	3	0	0	43	12	15	10	0	87	7	2	50	9	255
		25>	0	10	0	0	0	1	0	0	1	0	0	4	1	2	1	0	3	4	0	7	1	

Key: LY = Lymphosystis
U = Epidermal ulceration
EP = Epidermal papilloma
HYP= Hyperpigmentation
LN = Macroscopic liver lesion
X = x-cell disease
ST = Stephanostomum
LP = Lepeopthierius pectoralis
AC = Acanthochondria
NM = Nematodes
GL = Glugea

The most significant finding, that years 1994 to 1998 form a distinct group, can be explained by a major change in the sampling regime that occurred in 1998. Up until 1997, data were collected during the winter months, while in 1998 there were two cruises in winter and summer (the latter recorded as 1998b on the ordination plot) and thereafter cruises were undertaken during the summer season. During warmer temperatures it is known that the prevalence of skin ulcerations increases (Wosniok *et al*., 1999). However, other insights into changes of disease prevalence can also be discerned. The prevalence of liver nodules has increased steadily since 1998, with a similar pattern being seen for epidermal hyperplasia. Further investigations will be needed to determine the specific factors that are influencing this change.

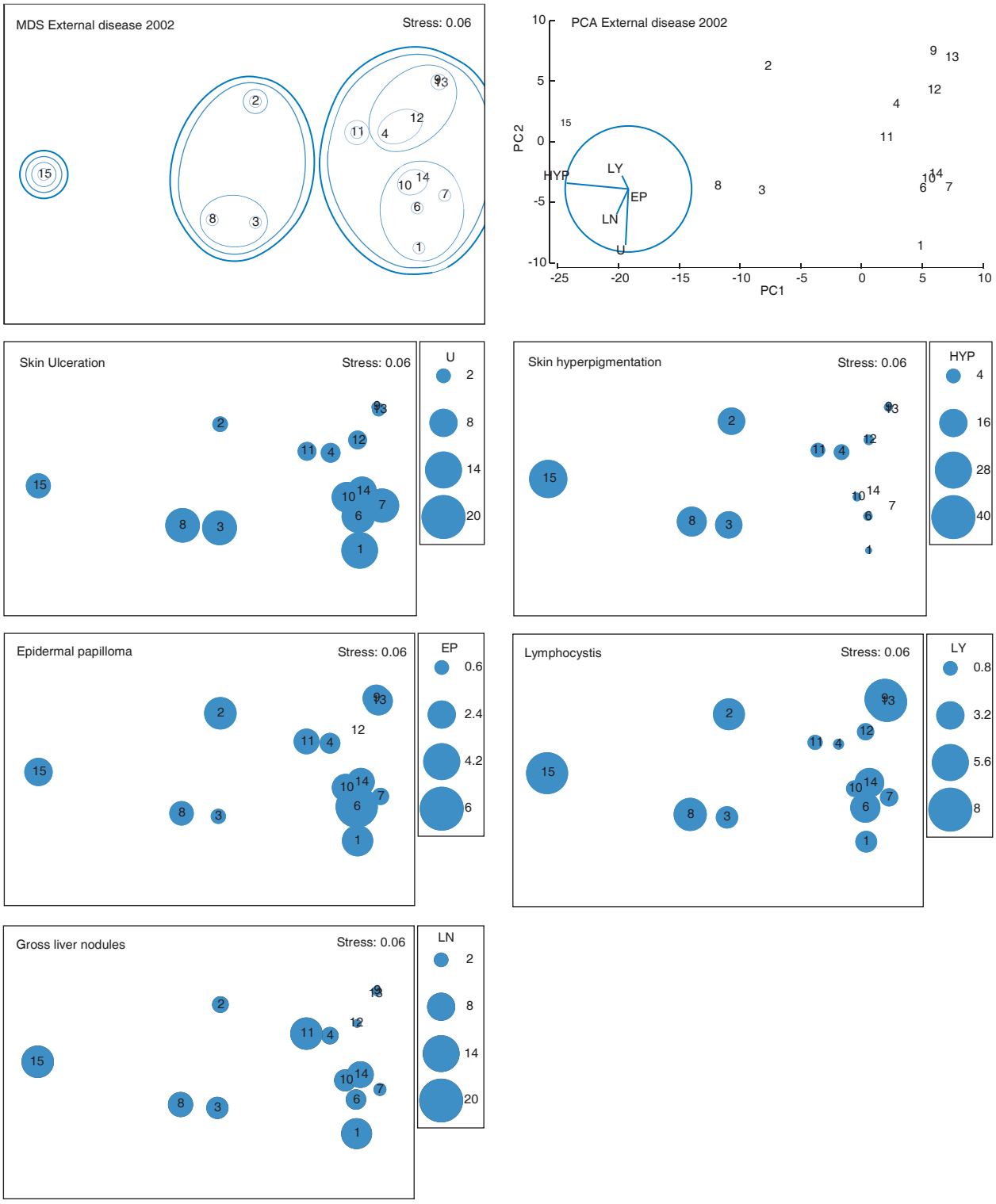

Figure 4. ***Multi-dimensional scaling, principal component analysis and individual bubble plots for externally recorded diseases from dab (Limanda limanda) captured from 14 sites as part of the NMMP during 2002. Data are expressed as average prevalence by site. Site key: 1 (Burbo Bight), 2 (Flamborough), 3 (Hospital Ground), 4 (Indefatigable Bank), 6 (Liverpool Bay), 7 (Morecambe Bay), 8 (North Dogger), 9 (Off Tees), 10 (Red Wharf Bay), 11 (North Cardigan Bay), 12 (Rye Bay), 13 (SE Isle of Man), 14 (St Bee's), 15 (West Dogger)***

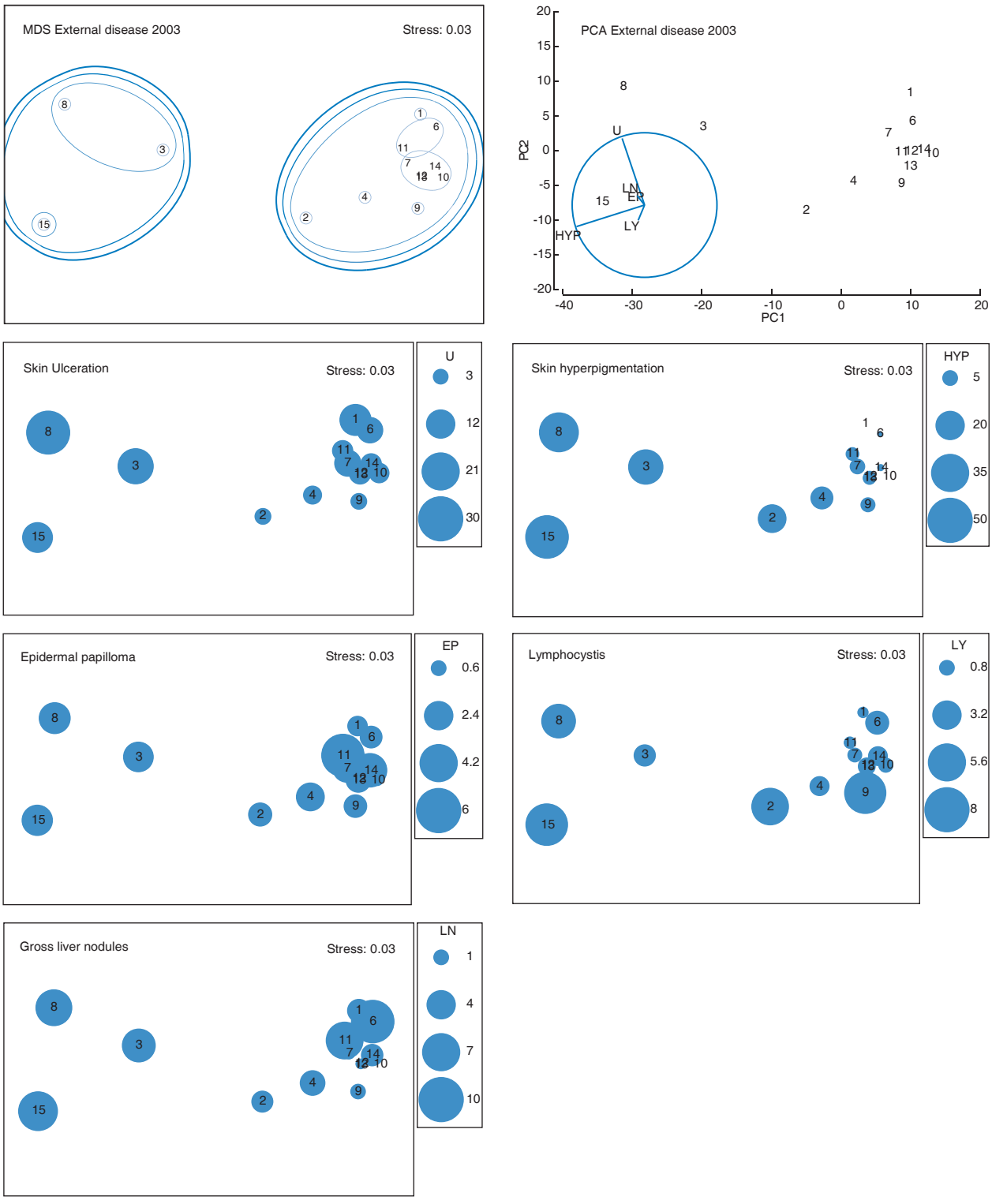

Figure 5. ***Multi-dimensional scaling, principal component analysis and individual bubble plots for externally recorded diseases from dab (Limanda limanda) captured from 14 sites as part of the NMMP during 2003. Data are expressed as average prevalence by site. Site key: 1 (Burbo Bight), 2 (Flamborough), 3 (Hospital Ground), 4 (Indefatigable Bank), 6 (Liverpool Bay), 7 (Morecambe Bay), 8 (North Dogger), 9 (Off Tees), 10 (Red Wharf Bay), 11 (North Cardigan Bay), 12 (Rye Bay), 13 (SE Isle of Man), 14 (St Bee's), 15 (West Dogger)***

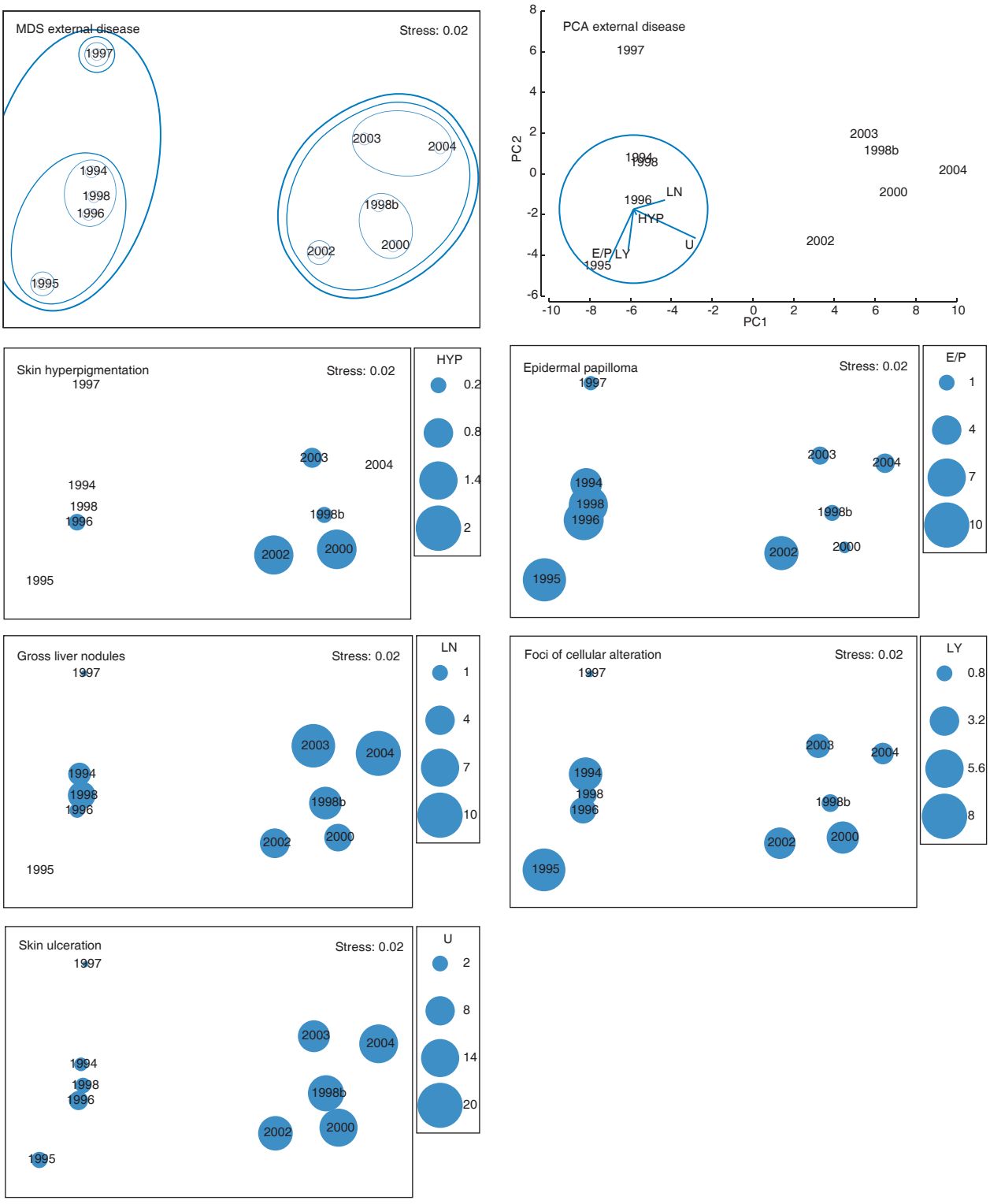

Figure 6. ***Multi dimensional scaling (MDS), principal component analysis (PCA) and individual bubble plots for externally recorded diseases from dab (Limanda limanda) captured from Liverpool Bay, Irish Sea over the period of 1994 to 2004. Samples up to 1998 were collected during the winter and samples including and following 1998b were collected during the summer***

7.3.2 Histological analysis of dab livers

Distribution of sites according to the prevalence of histopathological liver lesions shows a similar pattern between 2002 and 2003. However the statistical similarity between these was lower than that seen between the data for external disease prevalence (Rho = 0.45, $p<0.01$) (Figures 7 and 8). The distribution of sites according to increasing severity of liver lesions shows a marked shift to the right of the ordination plot. The highest prevalence of fish with malignant tumours being seen at sites 1, 8 and 20 (Inner and North Cardigan Bay and North Dogger Bank) in 2002 and sites 1, 18 and 20 (Inner Cardigan Bay, West and North Dogger Bank) in 2003. Sites are also discriminated by the prevalence of fish with no discernable pathology and non-specific pathology. For an assessment of relative levels of liver pathology, the use of Rye Bay as a reference site appears justified. However, other sites (e.g. SE Isle of Man) may also act as reference sites, since they also show low levels of liver pathology. It should also be noted that, while broad geographical sites could be discriminated on the basis of external disease (see above), similar patterns were not observed with liver pathology, since both toxicopathic lesions and neoplasms were observed at multiple sites in the Irish and North Seas.

7.3.3 Disease status of other species

General levels of disease in fish species other than dab were low (see Table 6). Apart from a case of skeletal deformity and two cases of lymphocystis in plaice from the Irish Sea in 2002 and a case of epidermal hyperplasia in whiting from the Celtic Deep in 2003, all observations were of parasitic infections. However, the numbers of each species examined were often insufficient to detect low prevalence of infections. In most cases, there was little discernable host response, the exception being infections with the gill copepod *Lernaeocera branchialis*. Infections of the gill of whiting and haddock frequently result in emaciation and anaemia, evident from extreme gill pallor. No evidence of *Ichthyophonus* was detected in herring from the North Sea in 2002. The previous epidemic of this disease in herring appears to have disappeared from the southern North Sea in recent years. In 2002 a sample of 197 Dublin Bay prawns (*Nephrops norvegicus*) was examined macroscopically for the presence of *Hematodinium*, which is known to be prevalent throughout much of the Irish Sea. All were free of the disease, though the sampling period would not have coincided with the known infection period (winter-spring) for this disease. Normal tissues and representative examples of diseased tissues and parasites from fish and shellfish specimens were taken for further processing in the laboratory and for accession to the Registry of Aquatic Pathology (RAP) reference collection held at the CEFAS Weymouth Laboratory.

7.4 Discussion

The application of multivariate statistics to fish disease data collected during 2002 and 2003 has revealed important new insights concerning the health status of dab at various locations in UK waters. Using external disease indicators, it has been possible to demonstrate clear differences in disease patterns between areas in the North and Irish Seas. Overall disease prevalence levels remain similar to previous years at several sites, with locations on the Dogger Bank showing consistently higher levels of external disease and liver pathology than other sites. The increased disease prevalence in dab from Cardigan Bay in recent years appears to have stabilised and, although fish exhibiting hyperpigmentation are recorded in the Irish Sea, the levels remain much lower than those observed at the majority of North Sea sites. The assessment of liver pathology data has also revealed clear site-to-site discrimination (with the SE Isle of Man site separated in particular in 2002) but in this case broader geographical groupings were not so apparent.

Commercial fish and other species examined during the monitoring programme did not show evidence of disease or parasitism above those recorded in previous years. Round fish such as haddock and whiting are more susceptible to the digenean parasite *Cryptocotyle* and the copepod *L. branchialis*, but only a few fish infected with these parasites showed obvious emaciation.

The analysis of liver histopathology data using multivariate statistics has shown that site discrimination is possible and that the presence of fish showing no pathology as well as those exhibiting benign and malignant tumours and non-neoplastic toxicopathic pathology are important components influencing the pattern seen in the ordination plots. However, more general groupings of sites based on geographical location, as were seen with the external disease data, were not as apparent. This is mainly due to the fact that late stage neoplasms are evident at both North and Irish Sea sites. Since several of the categories of pathology observed are thought to be related to exposure to organic compounds, further analyses incorporating chemical data as well as biomarkers of exposure are required to determine whether significant associations between these factors exist at the areas currently monitored. Fundamental to such studies will be an understanding of the behavioural biology and population genetics of the various dab populations examined.

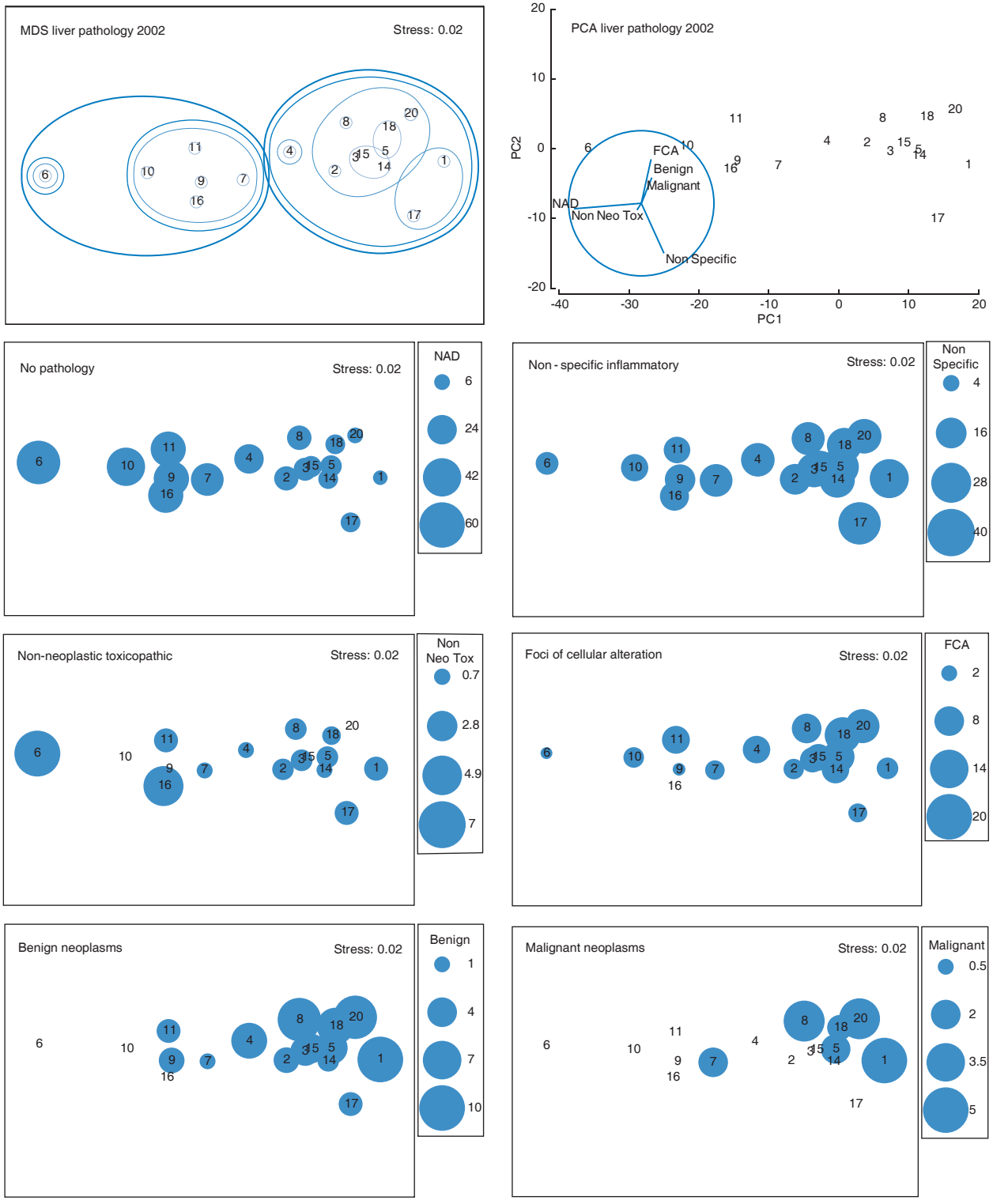

Figure 7. ***Multi dimensional scaling, principal component analysis and individual bubble plots for liver pathology from dab (Limanda limanda) captured at 17 stations as part of the NMMP during 2002. Data are expressed as averaged prevalence according to lesion categories as shown in bubble plots. Site Key: 1 (Inner Cardigan Bay), 2 (Red Wharf Bay), 3 (Liverpool Bay), 4 (Burbo Bight), 5 (St Bee's), 6 (SE Isle of Man), 7 (Morecambe Bay), 8 (North Cardigan Bay), 9 (Outer Cardigan Bay), 10 (Rye Bay), 11 (Outer Gabbard), 14 (Off Humber), 15 (Flamborough), 16 (Off Tees), 17 (Amble), 18 (West Dogger Bank), 20 (North Dogger Bank)***

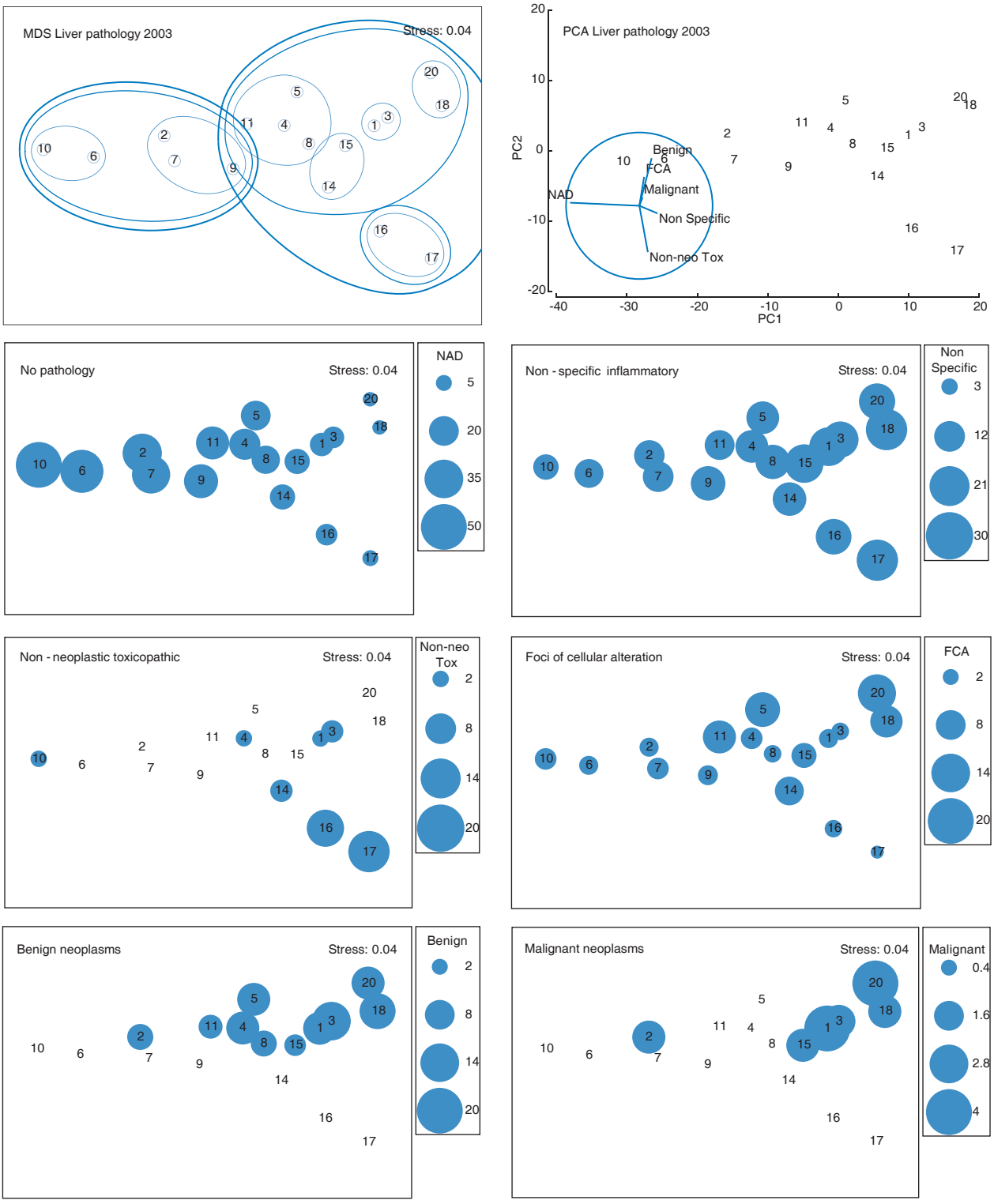

Figure 8. ***Multi dimensional scaling, principal component analysis and individual bubble plots for liver pathology from dab (Limanda limanda) captured at 17 stations as part of the NMMP during 2003. Data are expressed as averaged prevalence according to lesion categories as shown in bubble plots. Site Key: 1 (Inner Cardigan Bay), 2 (Red Wharf Bay), 3 (Liverpool Bay), 4 (Burbo Bight), 5 (St Bee's), 6 (SE Isle of Man), 7 (Morecambe Bay), 8 (North Cardigan Bay), 9 (Outer Cardigan Bay), 10 (Rye Bay), 11 (Outer Gabbard), 14 (Off Humber), 15 (Flamborough), 16 (Off Tees), 17 (Amble), 18 (West Dogger Bank), 20 (North Dogger Bank)***

Table 6. Disease status of non-target species from locations in the North Sea and Irish Sea sampled during 2002-2003

Species	No. examined	Year	Location	Parasites/Pathology (No. affected)
Haddock	18	2002	Dundrum Bay	NAD
"	23	"	Farne Deep	CR(1), LB(1), NEM(18)
"	50	2003	Celtic Deep	LB(1)
"	21	"	Dundrum Bay	CR(8), LB(1)
"	38	"	Flamborough	CR(3), LB(16), SD(1)
Plaice	1	2002	Liverpool Bay	LY
"	70	"	Morecambe Bay	LY(1), SD(1)
"	9	2003	Rye Bay	NAD
"	30	"	Carmarthen Bay	"
"	3	"	North Cardigan Bay	"
"	50	"	SE Isle of Man	LP(1)
"	100	"	Liverpool Bay	NEM(6), LP(3), GL(1)
"	100	"	Off Tees	NEM(21), LP(8), GL(3)
Whiting	50	2003	Celtic Deep	DM(24), MA(4), EH(1)
"	26	"	Dundrum Bay	CR(3), LB(1), DM(3)
"	100	"	Liverpool Bay	CR(23), LB(7), NEM(5)
"	62	"	Flamborough	CR(8), LB(1)
"	100	"	Off Tees	CR(4), LB(8), NEM(5), SD(2)
"	100	"	Farne Deep	LB(2)
"	48	"	Outer Thames	CR(1), LB(5)
Cod	7	2002	Liverpool Bay	NAD
"	30	"	Off Humber	LB(3)
"	9	2003	Flamborough	NAD
Dover sole	10	2002	Morecambe Bay	"
"	3	"	Off Humber	"
"	3	2003	North Cardigan Bay	"
Brill	1	2002	Liverpool Bay	Ulcer
"	1	"	Off Humber	NAD
"	3	2003	North Cardigan Bay	"
Thornback ray	22	2002	Liverpool Bay	Gill monogenean parasite
"	140	2002	North Cardigan Bay	NAD
Turbot	10	2003	"	"
"	5	"	Dundrum Bay	"
Dogfish	10	"	Carmarthen Bay	"
Herring	55	2002	Farne Deep	NEM(21)
Rockling	35	2003	"	NAD
"	26	"	Indefatigable Bank	"

Key: NAD = No abnormalities detected
CR = Cryptocotyle
LB = Lernaeocera branchialis
NEM = Nematodes (Anisakis spp.)
DM = Diclodophora merlangi
MA = Myxobolus aeglifini
LY = Lymphocystis
SD = Skeletal deformity (scoliosis/lordosis)
LP = Lepeophtheirus pectoralis
GL = Glugea
EH = Epidermal hyperplasia

8. THE USE OF THE DR-CALUX BIOASSAY TO DETERMINE DIOXIN-LIKE ACTIVITY IN UK ESTUARIES

Author: Mark Hurst and Jan Balaam

8.1 Introduction

Sediments from a number of UK estuaries have been tested for the presence of dioxins and dioxin-like compounds using a bio-analytical assay, DR-CALUX. The dioxin responsive-chemically activated *luciferase* expression assay uses a coupled receptor-reporter gene system and responds specifically to compounds which interact with the aryl hydrocarbon receptor. This includes dioxins and furans and other compounds with a similar mode of action. Increasing concentrations of dioxins and dioxin-like compounds cause an increase in the luminescence produced in the assay, which can be calibrated to yield a TEQ (toxic equivalent) value relative to the most active dioxin compound (2, 3, 7, 8-tetrachlorodibenzo-*p*-dioxin; TCDD).

Whole surface sediment samples were collected using a stainless steel van Veen grab, transferred to hexane-rinsed glass jars and stored frozen until analysed (Hurst *et al*., 2004). Samples were collected from 35 sites in seven UK estuaries, selected to include varying degrees of industrial, agricultural and domestic inputs (Clyde, Firth of Forth, Tyne, Tees, Thames, Southampton Water, Mersey) (Figure 9). The results of this study are given in Table 7. All samples gave TEQ values above the detection limit of the method, and the concentrations ranged from 1.0 to 106 ng kg^{-1} dry weight TCDD equivalents. Samples from the Firth of Forth, Tyne and Tees had the highest mean TEQ values, and samples from the Thames and Southampton Water the lowest. Within the estuaries showing the highest concentrations, TEQ values varied by a factor of about 3- to 8-fold. A further survey was conducted on the Tees estuary to study the variability in a single area, with five replicate samples being collected at each of four sites within the estuary (Figure 10). The results of this study are given in Table 8. The TEQ values ranged from 2.3 to 73 ng kg^{-1} dry weight, with differences in both the mean TEQ at the 4 sites (10-45 ng kg^{-1} dry weight) and in the variability at a site (less than 2x at site A and 10x at site B). The UK does not currently have an Environmental Quality Standard (EQS) or other formal risk evaluation for dioxins in sediments, although other countries (Canada, the United States of America and the Netherlands) have developed guideline values of 0.85, 2.5 and 13 ng kg^{-1} dry weight, respectively. Sediments from a number of the sites sampled in this study exceed these guideline TEQ values, and so could cause adverse effects in sensitive organisms. Comparison with the results obtained using conventional targeted chemical analysis to generate TEQ values showed that, for three of the five samples

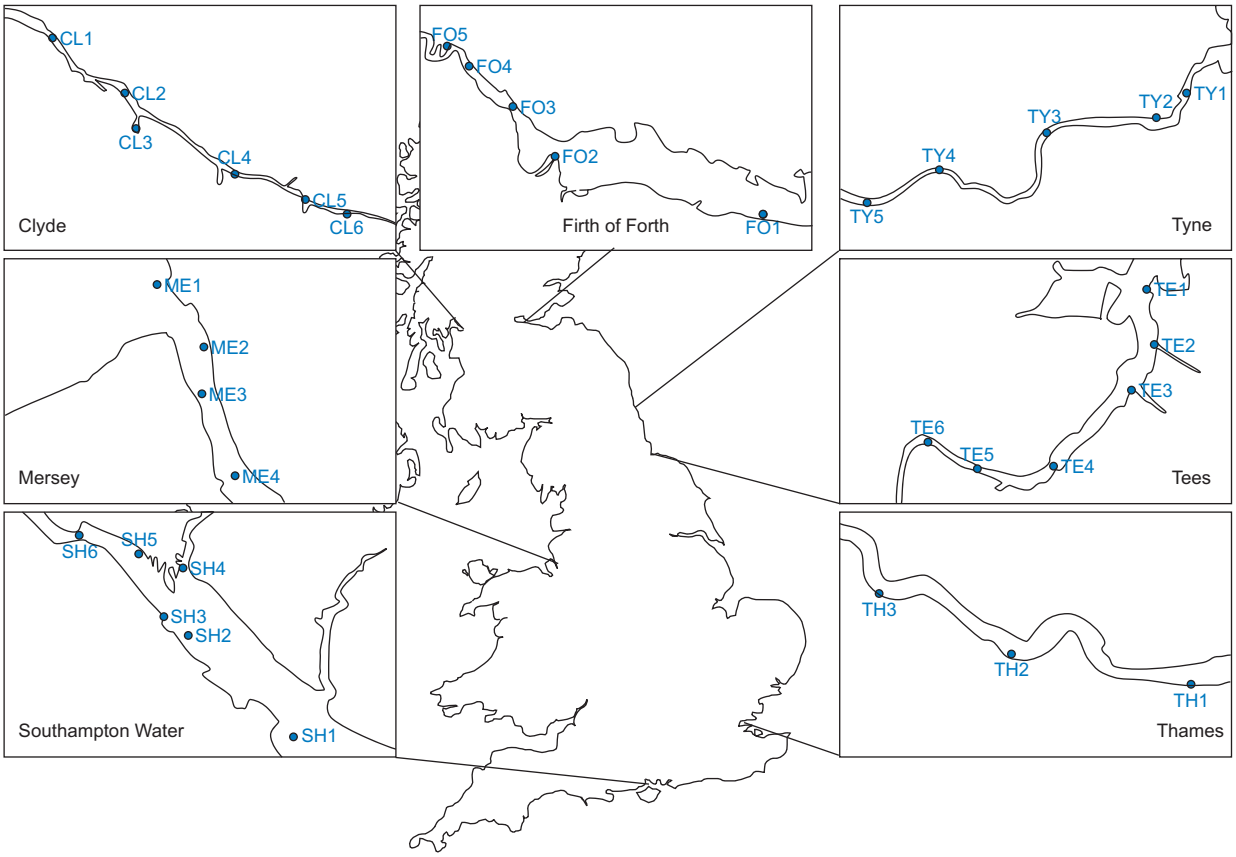

Figure 9. Sampling sites of the spatial survey of UK estuaries

Table 7. TEQ values for sediment samples produced using DR-CALUX assay

Estuary Name	Sample Site Code	ng TEQ kg^{-1} dry wt (SD)
Firth of Forth	FO1	46 (4.7)
	FO2	106 (4.3)
	FO3	19 (1.1)
	FO4	27 (0.3)
	FO5	20 (0.3)
Tees	TE1	15 (1.1)
	TE2	7.8 (0.3)
	TE3	27 (0.5)
	TE4	42 (2.3)
	TE5	88 (2.0)
	TE6	35 (1.4)
Tyne	TY1	47 (1.1)
	TY2	33 (2.8)
	TY3	32 (1.3)
	TY4	13 (0.6)
	TY5	30 (0.5)
Mersey	ME1	8.7 (0.6)
	ME2	40 (3.6)
	ME3	24 (1.9)
	ME4	19 (0.5)
Clyde	CL1	8.0 (0.01)
	CL2	16 (2.2)
	CL3	17 (1.3)
	CL4	23 (2.9)
	CL5	15 (1.1)
	CL6	25 (1.8)
Southampton Water	SH1	2.2 (0.2)
	SH2	4.5 (0.004)
	SH3	16 (0.8)
	SH4	3.0 (0.2)
	SH5	1.5 (0.1)
	SH6	16 (0.8)
Thames	TH1	1.0 (0.04)
	TH2	2.3 (0.1)
	TH3	2.3 (0.1)
Procedural Blank	BLK1	<0.3

studied, the DR-CALUX assay produced TEQ values that were three- to ten-fold higher than those generated analytically. This suggests that other compounds with dioxin-like activity, which are not included in the targeted chemical analysis, are contributing to the activity assessed using the bioassay technique. Overall, the DR-CALUX assay is considered to be a useful technique for monitoring and regulatory purposes.

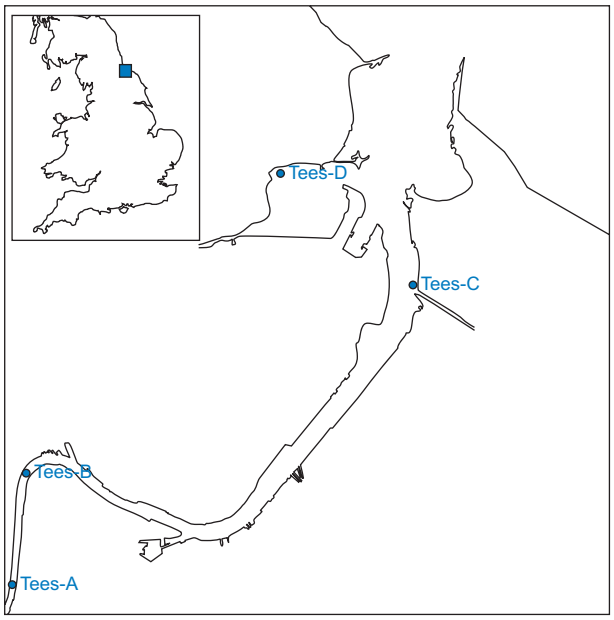

Figure 10. Map showing the sampling sites of the Tees survey

Table 8. TEQ values from the DR-CALUX assay of cleaned-up Tees sediment extracts with a 24 h exposure period

Sample Site	ng TEQ kg^{-1} dry wt (SD[a])
Tees-A1[b]	57 (2.4)
Tees-A2	33 (0.7)
Tees-A3	36 (3.2)
Tees-A4	51 (3.3)
Tees-A5	49 (3.0)
Mean Tees-A	45 (10[c])
Tees-B1	4.6 (0.3)
Tees-B2	2.3 (0.1)
Tees-B3	8.5 (0.2)
Tees-B4	12 (0.6)
Tees-B5	23 (0.5)
Mean Tees-B	10 (8.1[b])
Tees-C1	44 (0.9)
Tees-C2	73 (3.8)
Tees-C3	24 (0.9)
Tees-C4	20 (0.5)
Tees-C5	31 (0.5)
Mean Tees-C	38 (21[b])
Tees-D1	8.2 (0.2)
Tees-D2	27 (0.8)
Tees-D3	35 (0.6)
Tees-D4	19 (0.9)
Tees-D5	47 (2.1)
Mean Tees-D	27 (15[b])
Procedural Blank	<0.3

[a] *Standard deviation of 3 microplate replicates*
[b] *Standard deviation of 5 replicate sediment samples*
[c] *Site and replicate number*

9. THE USE OF BIOMARKERS IN BIOLOGICAL EFFECTS MONITORING

Authors: Paula Neall, Jennifer Rooke and Mark Kirby

9.1 Introduction

The mixed-function oxygenase (MFO) enzyme system in fish is the primary route by which many planar, organic contaminants, including PCBs and PAHs, are detoxified, and the MFO system is induced by exposure to these compounds. Ethoxyresorufin-O-deethylase (EROD) is a proven marker of MFO system induction because it is dependent on CYP1A1, the terminal component of the system.

9.2 EROD

9.2.1 Methods

The data reported here derive from the 2002 survey (research cruise *CIROLANA* 3b/02). This is the seventh consecutive year for which EROD data for Dab (*Limanda limanda*) has been collected in UK waters. (see CEFAS, 1998-2001 for earlier data). Fish were collected in July, using *RV CIROLANA*'s Granton trawl. Once on deck, target species were separated into tanks containing flowing seawater. Dissections were performed within 1 hour of capture. The liver was excised and placed in a cryovial, which was immediately placed in liquid nitrogen for storage. Notes were taken of fish condition, length, sex, gonad length and parasitism. Only fish over 10 cm were taken as samples.

Homogenate preparation

Once at the laboratory, liver samples were stored at -80°C. At the time of assay a 200 mg (±10) sliced sample of liver was homogenised with 1 ml of ice cold homogenising buffer (100 mM K_2HPO_4/KH_2PO_4 pH 7.5, 1 mM EDTA, 1 mM dithiothrietol, 150 mM KCL) using 6 strokes of a Potter-Elvehjem automatic homogeniser set at 4000 rpm. The homogenate was transferred to a polyethane Eppendorf tube and centrifuged at 10,000 g for 20 minutes at 4°C. Supernatants were removed and stored on ice.

EROD activity determination

EROD activity was determined by the standard ICES method (Stagg and McIntosh, 1998). A Perkin Elmer LS50B fluorescence spectrometer set at 535 nm excitation and 580 nm emission with a cuvette stirring function was used. Assay reagents were kept at 20°C (±1) in a water bath to maintain assay temperature. The final reaction mixture (2 ml volume) consisted of 1.96 ml assay buffer (100 mM K_2HPO_4/KH_2PO_4 pH 7.5, 100 mM KCL), 20 µl liver supernatant, 10 µl ethoxyresorufin substrate (0.2 nM in dimethylsulphoxide) and 10 µl resorufin internal standard 3.125 µM, adjusted by absorbance to allow for variation in the percentage purity of resorufin stock, in pH 8.0 phosphate buffer.

The reaction was initiated by the addition of 10 ml NADPH (0.25 mM), and fluorescence emission readings were taken at 0 and 60 seconds. EROD activity was normalised to protein content, which was determined by using a plate reader modification of the Bradford method (1976) using a bovine serum albumin standard.

9.2.2 Results

RV CIROLANA 3b/02

Dab (*Limanda limanda*) were collected from 21 sites in the North Sea and around the UK coast (See Table 9).

Table 9. Sampling Sites during RV CIROLANA 3b/02

Station No.	Location	NMP No.	Latitude	Longitude
1	Inner Cardigan Bay	656	52° 17.34'N	04° 18.14'W
19	Red Wharf Bay	776	53° 24.29'N	04° 05 53'W
27	Liverpool Bay	715	53° 27.98'N	03° 42.38'W
44	Burbo Bright	705	53° 28.31'N	03° 18.62'W
78	St Bees	769	54° 33.32'N	03° 50.05'W
90	SE Isle of Man	806	54° 03.73'N	03° 49.87'W
91	Morecambe Bay	796	53° 55.34'N	03° 23.24'W
100	N. Cardigan Bay	649	52° 41.96'N	04° 32.48'W
101	Outer Cardigan Bay		52° 23.51'N	04° 53.64'W
123	Rye Bay	486	50° 51.18'N	00° 46.98'E
153	Outer Gabbard		52° 01.20'N	02° 06.75'E
154	Wash		53° 08.00'N	00° 32.50'E
157	Inner Humber		53° 18.50'N	00° 26.14'E
165	Off Humber	346	54° 03.00'N	01° 46.61'E
168	Off Flamborough	344	54° 12.83'N	00° 36.52'E
171	Tees	294	54° 44.73'N	01° 08.29'W
192	Amble	244	55° 15.34'N	01° 14.51'W
206	West Dogger	286	54° 47.53'N	01° 16.83'E
209	East Dogger/ Bremehaven 9	283	55° 26.87'N	04° 04.98'E
211	North Dogger	284	55° 03.24'N	02° 03.34'E
213	Mid Dogger Hospital Ground		54° 33.31'N	02° 41.11'E

Table 10. Mean EROD (pM/min/mg protein) activity data for RV CIROLANA 3b/02

Station	Location	Number	All		Female		Male	
			Mean	Standard deviation	Mean	Standard deviation	Mean	Standard deviation
1	Inner Cardigan Bay	20	213	126	242	115	178	138
19	Red Wharf Bay	20	377	313	343	371	411	258
27	Liverpool Bay	20	654	461	791	549	531	349
44	Burbo Bight	20	604	518	688	711	528	271
78	St Bees	20	596	488	656	504	536	490
90	South East Isle of Man	20	564	467	808	608	364	144
91	Morecambe Bay	20	872	605	669	663	1076	492
100	N. Cardigan Bay	20	100	84	93	106	107	60
101	Outer Cardigan Bay	17	253	221	239	158	264	264
123	Rye Bay	20	93	55	121	65	66	24
153	Outer Gabbard	17	122	109	165	120	62	53
154	Wash	10	121	80	121	80	N/A	N/A
157	Inner Humber	20	151	101	183	132	119	43
157	Humber (Flounder)	21	42	35	38	23	45	46
165	Off Humber	20	395	179	374	119	416	229
168	Off Flamborough	20	436	348	593	411	279	177
171	Tees	20	322	242	355	279	290	208
192	Amble NMMP 244	20	1208	846	670	482	1692	823
206	West Dogger	20	196	152	219	167	174	142
209	East Dogger Bremerhaven 9	20	178	174	235	213	126	120
211	North Dogger	20	181	216	192	206	170	236
213	Mid Dogger Hospital Ground	20	318	224	300	192	332	256

The only sample showing mean EROD activity above 1000 pM/min/mg/protein was the site denoted Amble (NMP No. 244). (see Table 10). This sample also demonstrated a significantly ($p<0.05$) higher response in males compared to females which was not observed at any other site. Sample sites with the highest EROD levels (> 500 pM/min/mg/protein) were mainly situated in the North East of England (e.g. Amble, Tees) and the Eastern Irish Sea (e.g. Morecambe Bay, Burbo Bight). This generally reflects the pattern observed in previous years. Relatively low values (<150 pM/min/mg/protein) were found in Cardigan Bay, Rye Bay and the Wash, again, in keeping with previous results from these areas.

Table 11. Correlation (r values) analysis of EROD activity with other sample variables for RV CIROLANA 3b/02

	All	Female	Male
Length	-0.05	-0.21	0.12
Weight	-0.06	-0.19	0.09
Gonad Length	-0.01	-0.03	0.08
HSI	-0.20	-0.20	-0.19
GSI	0.00	0.00	-
Condition Factor	0.05	0.13	-0.03

Somatic data were analysed and did not appear to be a major factor influencing the EROD activity results on this occasion (see Table 11). Mean lengths ranged from 18 - 23.8 cm in females and 18.4 – 23 cm in males, while mean weights ranged from 63.2 - 138.2 g in females and 58.8 – 122 g in males. (See Table 12). Unfortunately, due to the variable abundance of target fish at the sites, it was not always possible to standardise these variables in the sampling strategy. The mean gonado-somatic index (GSI) for females across the sites varied between 0.5 and 2.1, indicating a potential difference in the mean maturation state of the dab populations at each site. There is a high female GSI at the Amble site which could explain the variation seen between males and females, because mature females are known to exhibit lower EROD levels than immature and male fish. (Lange *et al*., 1999). A difference in EROD levels between males and females may not have been observed at other sites if all fish were immature and so not subject to the influence on EROD levels normally associated with the reproductive cycle.

Table 12. Mean data for size and somatic indices of dab taken during RV CIROLANA 3b/02

Station	Location	All					Female					Male				
		Length	Weight	HSI	GSI	Conditioning factor	Length	Weight	HSI	GSI	Conditioning factor	Length	Weight	HSI	GSI	Conditioning factor
1	Inner Cardigan Bay	19.0	65.6	1.1	0.7	0.9	19.6	72.3	1.0	0.7	0.9	18.4	58.8	1.1		0.9
19	Red whalf bay	21.7	104.9	1.6	0.8	1.0	22.2	114.1	1.5	0.8	1.0	21.1	95.7	1.6		1.0
27	Liverpool Bay	20.7	96.0	1.5	0.9	1.1	21.1	100.6	1.5	0.9	1.1	20.3	91.5	1.5		1.1
44	Burbo Bight	21.0	97.3	1.7	1.2	1.0	21.7	108.4	1.7	1.2	1.0	20.3	86.2	1.6		1.0
78	St Bees	21.5	114.5	1.4	1.0	1.1	22.6	138.2	1.4	1.0	1.2	20.3	90.9	1.4		1.1
90	South East Isle of Man	19.9	85.3	1.8	0.9	1.1	20.2	89.4	1.7	0.9	1.1	19.5	82.0	1.9		1.1
91	Morecambe Bay	20.8	102.3	1.5	0.9	1.1	21.5	115.9	1.6	0.9	1.2	20.1	88.7	1.4		1.1
100	N Cardigan Bay	21.2	96.4	1.3	0.8	1.0	23.1	121.6	1.2	0.8	1.0	19.3	71.3	1.4		1.0
101	Outer Cardigan Bay	18.4	66.9	1.5	0.6	1.1	18.0	63.2	1.6	0.6	1.1	18.7	69.6	1.4		1.0
123	Rye Bay	21.6	111.9	1.8	0.5	1.1	22.9	133.1	1.6	0.5	1.1	20.3	90.8	2.1		1.1
153	Outer Gabbard	22.6	124.3	1.9	0.8	1.1	22.9	126.4	1.8	0.8	1.0	22.3	121.3	2.2		1.1
154	Wash	22.9	121.9	2.1	0.8	1.0	22.9	121.9	2.1	0.8	1.0					
157	Inner Humber	21.3	100.1	2.0	0.9	1.0	23.0	124.1	1.8	0.9	1.0	19.5	76.1	2.2		1.0
157	Humber (Flounder)	26.9	195.9	1.2	1.0	1.0	27.5	215.4	1.0	1.3	1.0	26.2	174.4	1.4	0.2	0.9
165	Off Humber	21.2	98.5	1.5	1.2	1.0	22.9	124.6	1.7	1.2	1.0	19.5	72.3	1.4		1.0
168	Off Flamborough	21.6	102.3	1.7	1.0	1.0	23.3	121.3	1.6	1.0	1.0	19.9	83.3	1.8		1.0
171	Tees	20.9	95.2	1.9	1.1	1.0	21.6	103.8	1.8	1.1	1.0	20.2	86.6	2.0		1.0
192	Amble NMMP 244	21.7	103.0	1.6	2.1	1.0	22.0	109.2	1.6	2.1	1.0	21.4	96.8	1.6		1.0
206	West Dogger	21.4	96.8	1.5	1.3	1.0	22.4	111.0	1.5	1.3	1.0	20.3	82.6	1.5		1.0
209	East Dogger Bremerhaven 9	22.8	113.8	1.4	1.7	0.9	23.7	129.3	1.3	1.7	1.0	21.8	98.3	1.5		0.9
211	North Dogger	23.4	122.3	1.4	1.5	0.9	23.8	122.5	1.3	1.5	0.9	23.0	122.0	1.4		1.0
213	Mid Dogger Hospital Ground	22.9	118.1	1.4	1.0	1.0	24.0	130.6	1.3	1.0	0.9	21.9	107.8	1.4		1.0

Sampling sites off the North West and North East coasts of England tended to have the highest EROD values, and this is almost certainly associated with inputs of organic pollutants from the industrialised estuaries that discharge into these areas. The sites on the Dogger Bank (NMP Nos. 283-286) in the North Sea had the next highest EROD levels and these were approximately half those observed in fish caught in the North West. The samples taken from sites in the South East of England and Wales generally had the lowest EROD levels. In both the North East and the North West, the EROD levels tended to increase in the more northerly samples (See Figure 11).

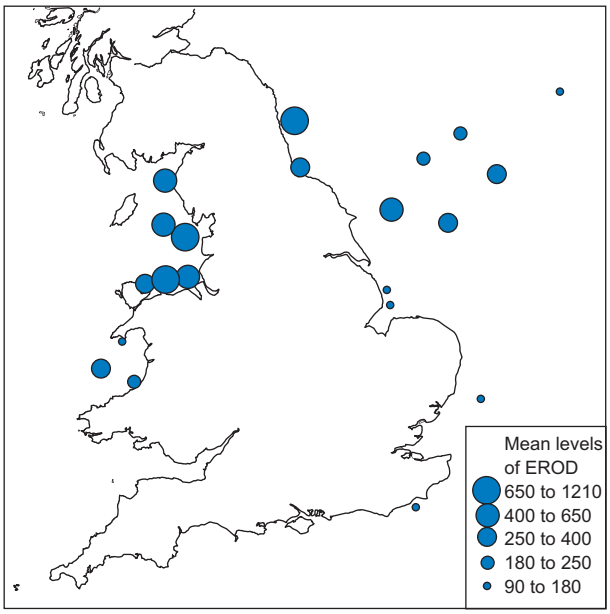

Figure 11. Mean EROD values (pM/min/mg protein) for samples taken during 2002

9.2.3 Conclusions

2002 is the seventh consecutive year of monitoring hepatic EROD activity in dab (*Limanda limanda*) from UK marine sites. The trends observed in previous years have generally been repeated, i.e. high EROD levels at sites in and around Liverpool Bay, and lower EROD activity levels in fish taken from sampling sites along the Welsh coast and Rye Bay. Certain sites have produced samples with consistently high EROD activity levels over the past 7 years of monitoring. This may indicate that fish species resident in these areas are being exposed to higher levels of EROD-inducing chemicals over longer periods of time than those in other areas. This potential long term exposure to biologically active levels of organic contaminants could be having a detrimental affect on these fish populations, because elevated activity of the MFO system suggests the presence of contaminants, such as PAH, which are known to be genotoxic and carcinogenic and, in some cases, to have endocrine disrupting properties. There appeared to be no clear correlation between EROD and any of the standard somatic indices (Table 11) therefore the results can be assumed to represent a response that is mainly due to the exposure to active contaminants via a number of potential routes.

9.3 Bile

9.3.1 Introduction and methods

PAH are ubiquitous contaminants in UK estuarine, coastal and marine waters. They originate mainly from incomplete combustion processes and from the petrochemical industry (National Research Council, 1985). PAH are hydrophobic molecules that have the ability to become associated with sediments once released to the marine environment and so may become biologically available to benthic organisms. PAH levels in fish tissues are often found to be very low even in highly contaminated areas (Varanasi *et al*., 1989) as, once ingested, these compounds are readily metabolised in the liver and excreted into the gall bladder. As a result, the bile of fish exposed to PAH may contain oxygenated PAH metabolites, usually in the form of glucuronide, sulfate or glutathione conjugates. Therefore the bile represents an important body fluid in which to investigate the exposure of fish to these contaminants.

Analysis of all possible PAH metabolites using HPLC or gas chromatography is both costly and time consuming. As an alternative, a fast screening method using synchronous fluorescence scanning (SFS) spectrometry based on the work of Ariese *et al*. (1993) and adapted by D. Barbe (unpublished) has been used. 1-Hydroxypyrene (1-OH pyrene), the main metabolite of pyrene, accounts for a large percentage of the total PAH metabolites in the bile of fish exposed to PAH (Krahn *et al*., 1987). The use of 1-OH pyrene as a standard in this method makes it possible to quantify the 1-OH pyrene glucuronide present in the sample.

Bile samples have been collected from dab taken from National Marine Monitoring Programme (NMMP) stations during research cruises since 1998, and this report presents the data from 2001 and 2002.

Fish were collected during 2001 and 2002 using *RV CIROLANA*'s Granton trawl. Dab of suitable size (>10 cm) were separated into tanks containing flowing seawater. Dissections were performed within one hour of capture and the gall bladder was excised and stored in a cryovial in liquid nitrogen for the duration of the cruise. At the laboratory the samples were transferred to storage at -80°C.

Bile volume in fish is highly variable and can depend upon many factors. The stage at which the fish was caught during it's normal daily behavioural cycle, time of the year and health status are particularly relevant, as these control the feeding cycle and hence the bile volumes available. It is possible to obtain up to 0.5 ml bile from small fish, however, if fish have just fed, there may be no bile in the gall bladder at all. Twenty fish were sampled at all sites but, frequently, only a small proportion of these fish yielded sufficient bile for analysis.

Immediately prior to analysis, the samples are allowed to acclimate to room temperature. Using a Gilson Microman pipette, 20 µl of sample was made up to 10 ml in 50:50 ethanol:nanopure water in a volumetric flask.

The diluted samples were then analysed using a Perkin-Elmer LS50B Fluorescence spectrophotometer. The instrument is used in synchronous fluorescence scanning mode, scanning between 323 and 423 nm with a wavelength offset of 37 nm. 50:50 ethanol: nanopure water was used as a blank. The standard used, 10 ppb1-OH-pyrene, shows a peak at an emission wavelength of 349.75 nm against which the target metabolite 1-OH pyrene glucuronide (peak at 344.30 nm) is quantified.

To calculate the results the following formula is used.

Height sample – blank (peak at 344.3) /
Height standard (peak at 349.75) *
Standard concentration in ppb * dilution factor

The results are expressed as ppb 1-OH pyrene.

9.3.2 Results

In total, 18 sites were sampled in 2001 (see Table 13) from which there were <10 samples at 5 sites i.e. Dundrum Bay (4), Red Wharf Bay (9), Cardigan Bay (7), Outer Humber (5) and Tees (9).

It can be seen from Figure 12 that samples from Morecambe Bay, Liverpool Bay and Red Wharf Bay had the highest levels of the bile metabolite (313, 306 and 238 respectively) and samples from North Dogger, Dundrum Bay and the Tyne the lowest (147, 156 and 157 respectively).

Table 13. Bile Metabolites (ppb 1-OH Pyrene) for RV CIROLANA 5b/01

Site	n	Mean	Standard deviation
Hospital Ground	25	165.0	60.31
Dundrum Bay	4	156.0	46.21
S E Isle of Man	11	157.8	48.08
Red Wharf Bay	9	238.1	51.84
Liverpool Bay	13	173.5	44.97
Liverpool Bay NMP 705	16	306.9	123.57
Morecambe Bay	14	313.9	130.72
Off Cardigan Bay	7	180.4	53.48
Inner Cardigan Bay	11	208.6	56.21
Rye Bay	16	200.6	77.15
Outer Gabbard	15	187.9	63.27
Silver Pit	13	218.1	45.31
NMP 376	5	265.6	91.04
Dogger Bank	17	172.4	74.16
North Dogger	13	147.2	45.05
Tyne	16	157.1	55.71
Tees	9	225.0	71.02
Off Flamborough Head	12	176.6	31.08

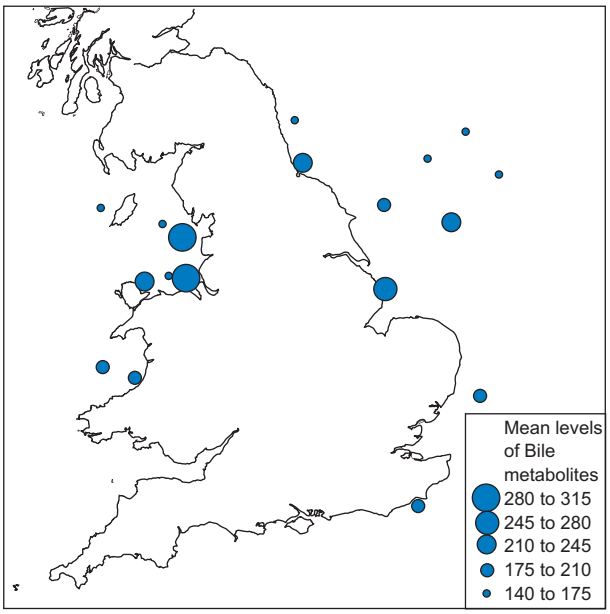

Figure 12. Mean levels of Bile Metabolites (ppb 1-OH pyrene) for samples taken during 2001

Table 14. Bile Metabolites (ppb 1-OH Pyrene) for RV CIROLANA 3b/02

Site	n	Mean	Standard deviation
Inner Cardigan Bay	9	159.7	30.74
Red Wharf Bay	11	243.1	135.10
Liverpool Bay	16	179.7	77.36
Burbo Bight	13	402.5	427.43
St Bees	17	310.8	104.57
S E Isle of Man	11	247.4	71.64
Morecambe Bay	11	391.6	138.78
N Cardigan Bay	12	195.1	45.87
Outer Bardigan Bay	4	158.3	66.58
Rye Bay	5	206.9	17.41
Outer Gabbard	14	279.6	49.47
Wash	7	287.6	50.46
Inner Humber	9	269.3	54.38
Off Humber	9	234.1	81.04
Off Flamborough	12	168.0	23.17
Tees	7	175.1	34.99
Amble NMMP 244	11	206.6	53.98
West Dogger	12	169.0	32.16
East Dogger	14	186.1	58.77
North Dogger	11	148.9	85.01
Mid Dogger	10	118.8	32.22

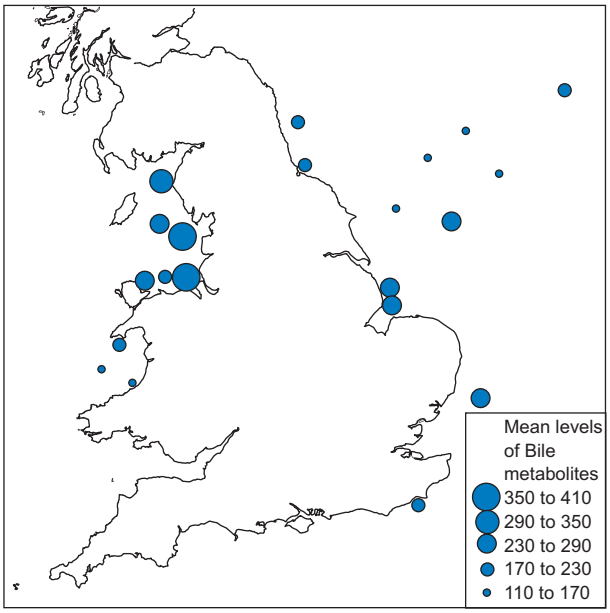

Figure 13. Mean levels of Bile Metabolites (ppb 1-OH pyrene) for samples taken during 2002

Twenty one sites were sampled in 2002 (see Table 14) of which 7 sites yielded <10 samples for analysis. These were Outer Cardigan Bay (4), Cardigan Bay (9), Tees (7) Rye Bay (5), Off Humber (9) and Wash (7).
The mean levels of the bile metabolite found in samples from the Burbo Bight (402) was higher than at all other stations but, due to high inter sample variability (two samples from this site had levels of 1727 and 737), this was not found to be significant ($p<0.05$). Levels found in samples from Morecambe Bay (392) were significantly higher than at all other stations except the Wash, and levels at St Bees were also high (311). The lowest levels of bile metabolite were found in samples from Mid Dogger Bank (119), North Dogger (149) and Cardigan Bay(158) (see Figure 13).

9.3.3 Discussion

The above results suggest that the SFS method provides a useful tool in the monitoring of PAH exposure in fish samples taken during research cruises. A number of practical considerations need to be taken into account when designing monitoring programmes if bile samples are to be taken. As has been stated, sufficient bile for analysis is not always available; therefore, if possible, sampling time/strategy should take this into account. However, in most cases >50% of the fish yielded appropriate volumes, so the data presented here can be considered representative of PAH exposure levels. Concentration of PAH metabolites in the gall bladder can be highly influenced by dietary status (Ruddock *et al.*, 2002). If fish have not recently fed, the metabolites become concentrated in the gall bladder due to resorption of water, therefore it is recommended that the data should be normalised to biliverdin content. However, even though these data have not been normalised to biliverdin, they still provide a good indication of the areas where PAH exposure may give cause for concern.

In both sets of data, higher levels of PAH metabolite were found in fish from the north west coastal area i.e. Liverpool Bay, Red Wharf Bay and Morecambe Bay. In 2001 high levels were found at the Tees site, but in 2002 the level found was lower. In both sets of results, levels from sites on the Dogger Bank are low.
The cost in both time and effort of obtaining bile samples from fish that are already being sampled for other biomarkers, and the subsequent analysis for bile metabolites by SFS spectroscopy, would seem justified as it can provide additional information on the levels of PAH contaminants in UK estuarine waters.

9.3.4 General conclusion

The EROD and bile metabolite data presented above continues to expand the dataset which allows the use of these biomarkers of exposure to be assessed.

Both methods continue to highlight the main areas of concern as the North East and North West coasts of England. The size of the temporal dataset now provides convincing evidence that fish populations in these areas are being exposed to biologically available, if perhaps low, concentrations of organic pollutants such as PAH.

While we continue to build a sufficient temporal dataset for bile metabolite screening to allow a detailed interpretation, the 2002 data presented here represents the seventh consecutive year of EROD monitoring in dab. Figure 14 shows the mean EROD activity levels at four key sites, representative of the Northwest (Red Wharf Bay and the Burbo Bight) and Northeast (off the rivers Tyne and Tees) coasts over a seven year period. While the last monitoring report (Irish (compiler), 2003) suggested a tentative downward mean EROD activity trend in the Liverpool Bay area, this latest dataset does not support this suggestion, with comparatively high EROD activities at those sites. Furthermore, the trend data for the Northeast coast sites continue to fluctuate, with the Off Tyne site returning the highest mean activity in the last seven years and the Off Tees site the lowest.

In general, therefore, the biomarker datasets remain difficult to interpret. Whilst all datasets presented here (and in previous years) show global elevations in some areas, no unambiguous link to known PAH concentrations have yet been made (Kirby *et al*., 2000). It is recognised that a wide range of seasonal and physical factors are influential in determining the ultimate EROD expression or bile metabolite profile in a given individual or population. To some extent, careful survey design and changes to existing protocols (e.g. normalisation of metabolite data to biliverdin content) can minimise weighting attributed to non-contaminant mediated influences in the results. However, while the use of these biomarkers continue to provide a valid and important input to the biological effects monitoring programme, there can be no question that further research is required to increase our understanding and to aid data interpretation.

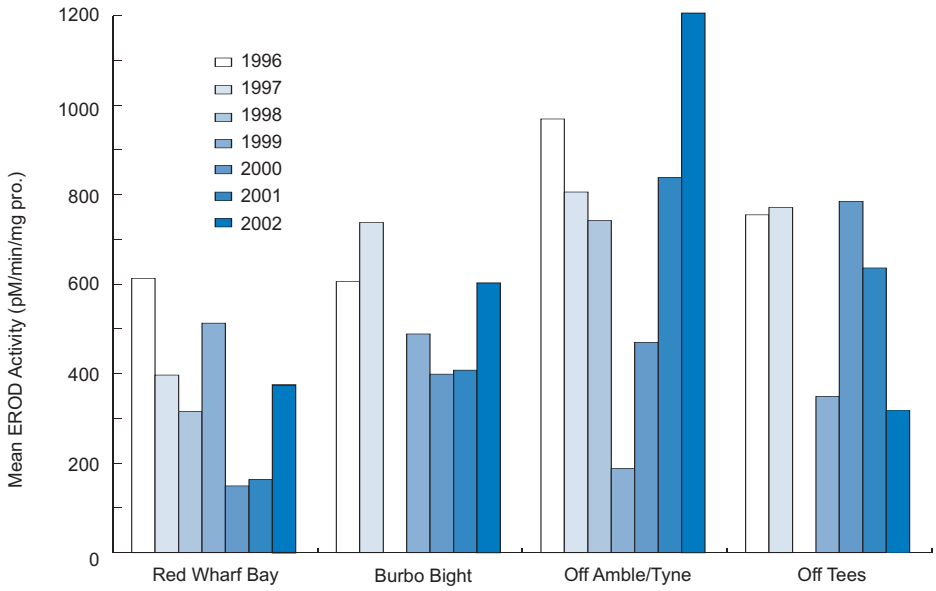

Figure 14. Seven-year trend for Mean EROD at selected sites

10. POLYCYCLIC AROMATIC HYDROCARBONS (PAH) IN SEDIMENTS COLLECTED WITHIN THE UK NATIONAL MARINE MONITORING PROGRAMME: DATA FOR 2000-2003

Authors: Carole Kelly, Robin Law and Paul Roberts

10.1 Introduction

Following the completion of the first phase of the UK NMMP, which was targeted on spatial variability, and the publication of the first NMMP report (MPMMG, 1998), the focus of the programme switched to the investigation of temporal trends. Our PAH temporal trends studies began in 2000, with initial data for offshore stations being presented in an earlier report in this series (Compiled by R. Irish, 2003). The aim was to annually collect sediments from the same NMMP sites until 3 data sets for each station were obtained. We now have 3 or 4 years data available for the 13 offshore and intermediate NMMP stations which are sampled by CEFAS. These were collected following the procedural guidelines given within the NMMP Green Book. This requires the collection of 5 replicate sediment samples randomly within a 50 m radius at each site on each occasion, thus giving an indication of the small-scale variability in PAH concentrations at each station. Table 15 shows the locations of the stations and an indication of sediment type at each site.

During the period 2000-2003, the 10 parent PAH currently defined within the NMMP were determined in sediments at each site. The PAHs were extracted from the sediment samples using alkaline saponification and analysed by coupled gas chromatography-mass spectrometry, with associated quality assurance (Kelly *et al*., 2000). The mean concentrations for individual PAH and ΣPAH, the sum of the PAH determined, are shown in Table 15. This allowed the ranking of the sites spatially (Figure 15), with the highest concentrations occurring at NMMP 245 off the River Tyne, ΣPAH 1,340 µg kg^{-1} dry weight. Intermediate concentrations within the range 500-700 µg kg^{-1} dry weight were observed at NMMP sites 376 off the Wash, 605 in the Celtic Deep, and 655 in Cardigan Bay. Lower concentrations in the range 100-400 µg kg^{-1} dry weight were found at NMMP sites 345 off the Humber/Wash, 466 Thames, 484 Dungeness, and 805 SE Isle of Man. The lowest concentrations were found at NMMP site 475 at the outer Gabbard, where the substrate is clean sand. These concentrations are much lower than those found in some estuarine locations, as determined during earlier spatial studies (Woodhead et *al*., 1999).

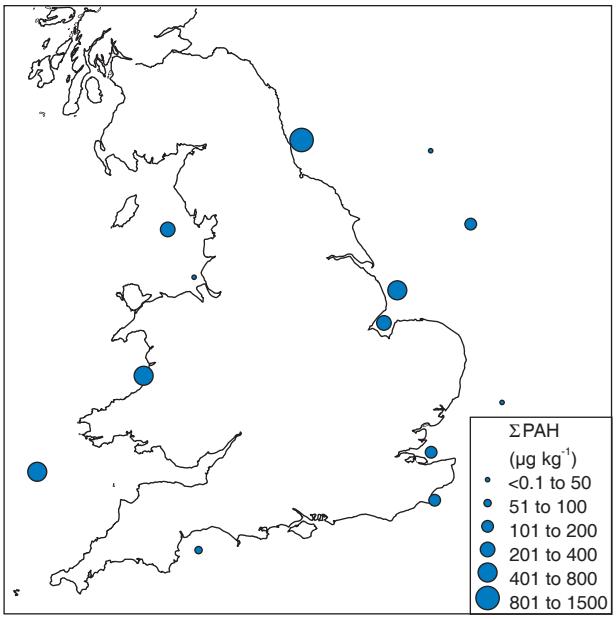

Figure 15. Map showing ΣPAH in sediment at NMMP stations

To assess how well the data meet the current aim of the NMMP to study temporal trends in contaminant concentrations, Table 16 summarises the PAH data by location and year, with an indication of the variability in PAH concentrations across the 5 replicate samples. At most of these stations it appears that the PAH concentrations are too low, and so the associated variability expressed as a percentage coefficient of variation too high, to allow the ready detection of time trends. Of the 13 stations studied, the best candidates are station 245 off the Tyne (CVs in the range 6.7 to 13%) or station 655 in Cardigan Bay (CVs in the range 4.3 to 14%).

In order to better investigate temporal trends in PAH concentrations, one approach could be to identify additional sites at which PAH concentrations are

Table 15. NMMP site locations, sediment type, range of PAH determined and concentrations found (µg kg^{-1} dry weight)

NMMP no.	Location	Sediment type	Naphthalene	Phenanthrene	Anthracene	Fluoranthene	Pyrene	Benz[*a*]anthracene	Chrysene/ Triphenylene	Benzo[*a*]pyrene	Indeno[1,2,3-*cd*]pyrene	Benzo[*ghi*]perylene	ΣPAH
245	Off Tyne	Mud and sand	108	206	41	237	161	114	132	130	99	109	1337
285	Off Tyne/Tees	Sand and shells	1.4	3.2	0.3	2.1	1.6	1.5	1.1	1.1	1.0	1.3	15
345	Off Humber/Wash	Mud and sand	7.9	20	2.1	24	20	12	15	14	21	22	158
376	Off Wash	Mud and shells	88	133	15	96	82	43	53	53	30	54	647
386	Wash	Mud, sand and small stones	32	80	6.7	55	45	20	27	26	21	25	338
466	Thames	Sand and shells	7.8	14	4.2	32	27	18	15	24	20	19	181
475	Outer Gabbard	Sand	0.6	0.4	<0.1	0.1	0.3	<0.1	0.1	<0.1	<0.1	0.2	1.7
484	Dungeness	Sand	2.9	9.0	1.8	19	14	21	10	13	12	11	114
536	Lyme Bay	Mud and sand	1.6	6.5	1.4	12	7.7	7.6	5.4	7.0	6.3	5.3	61
605	Celtic Deep	Mud and sand	26	80	24	152	99	61	67	64	48	36	657
655	Cardigan Bay	Mud and sand	24	68	13	97	64	42	59	50	45	40	502
715	Liverpool Bay	Mud and sand	2.6	4.4	0.7	4.8	3.8	3.5	2.9	3.3	3.8	3.3	33
805	SE Isle of Man	Mud and sand	11	23	3.3	34	28	13	18	31	44	34	239

Table 16. Summary of PAH data by location and year, with an indication of variability at each site

NMMP no.	Location	Year	mean ΣPAH (µg kg^{-1})	sd	CV%
245	Off Tyne	2000	1282	128	10
		2001	1407	178	13
		2002	1651	112	6.7
		2003	1005	67	6.7
285	Off Tyne/Tees	2001	11	3.1	29
		2002	17	3.4	20
		2003	17	6.0	36
345	Off Humber/Wash	2000	171	42	25
		2001	118	18	15
		2002	193	25	13
		2003	149	35	23
376	Off Wash	2000	687	169	25
		2001	526	132	25
		2002	705	107	15
		2003	671	96	14
386	Wash	2001	359	216	60
		2002	278	90	32
		2003	375	51	14
466	Thames	2000	208	85	41
		2001	125	22	18
		2002	286	105	37
		2003	123	83	67
475	Outer Gabbard	2000	5.4	5.6	104
		2001	0.8	0.4	50
		2002	0.2	0.2	100
		2003	0.8	0.2	25
484	Dungeness	2001	104	35	34
		2002	136	34	25
		2003	101	25	25
536	Lyme Bay	2000	55	45	82
		2001	45	19	42
		2002	58	55	95
		2003	84	33	39
605	Celtic Deep	2001	431	80	19
		2002	673	177	26
		2003	864	540	63
655	Cardigan Bay	2000	315	28	8.9
		2001	583	25	4.3
		2002	565	69	12
		2003	541	75	14
715	Liverpool Bay	2000	30	26	87
		2001	17	14	82
		2002	38	31	82
		2003	47	46	98
805	SE Isle of Man	2000	279	64	23
		2002	227	24	11
		2003	213	29	14

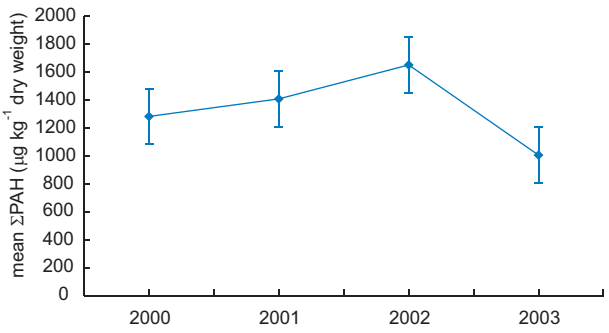

Figure 16. Mean PAH concentration in seiments from the Off Tyne site NMMP 245 (±SD)

reasonably high, and to begin to study those sites in addition to those currently sampled. Data gathered by Woodhead *et al*. (1999) suggest that two stations sampled during the earlier spatial phase of the NMMP have suitably high PAH concentrations – these are the stations off the Tees (NMMP 295) and off the Tamar (NMMP 575, which is no longer sampled). Other possibilities may be to increase the number of samples taken at each station, or to increase the frequency of sampling. These alternatives are currently being considered within the NMMP redesign study. As an initial step, the size of the trends which could be detected in the concentrations of individual PAH has been assessed from the data currently available (Rob Fryer, FRS MLA, personal communication). It is clear that the detection of a 10% trend over a 10 year period is not possible for most PAH compounds at most stations. Also, at around one-fifth of the sites, it would not be possible to detect a 20% trend over the same period. As can be seen from Figure 16, the major problem is the 'between-year' variability, which is too high. Much of this may actually be short scale temporal or spatial variation, that shows up as between-year variation because samples are collected at only one place and at one time each year. In this case, repeated sampling, say a few weeks apart, may help to average out short-scale temporal variation in contaminant levels. Also, redefining sites to have a larger area, and then taking samples at random from that area (and possibly collecting and analysing more samples, or bulking a number of samples for each analysis) could help to average out local spatial variation.

11. ASSESSMENT OF THE REHABILITATION OF THE SEABED FOLLOWING CESSATION OF AGGREGATE EXTRACTION

Authors: Rebecca Smith, Siân Boyd, Keith Cooper, David Limpenny, Hubert Rees, Mike Dearnaley, Jim Stevenson, William Meadows and Claire Morris

11.1 Introduction

Concerns over the effects of sand and gravel extraction on the environment and fisheries have grown with time, and is particularly the case at localities off the eastern and southern English coastlines. Studies of the biological and physical status of licensed areas following cessation of dredging are limited, and so judgements as to the likely timescales for restoration are based on predictions rather than real data. This project was established to address this deficiency with the following scientific objectives:

- to understand the rate at which the seabed recovers
- to identify measures to enhance the potential for rehabilitation at dredged areas
- to investigate whether different historical levels of dredging intensity affect the subsequent rate and nature of recolonisation

11.2 Methods

Four sites (Figure 17) were selected for comprehensive time-series investigations of the sediments and benthic fauna. They are considered representative of current dredging practices and habitats in the UK. Each location varies in the time-interval since cessation of dredging, and also, the degree of dredging intensity (see Table 17 for summary site descriptions).

Information from the ship-board Electronic Monitoring System (EMS) placed on all dredgers since 1993 was used to precisely target locations of high and lower dredging intensity (see Figures 18 and 19 for an example). As no information was available on the pre-dredging status of each extraction site, reference

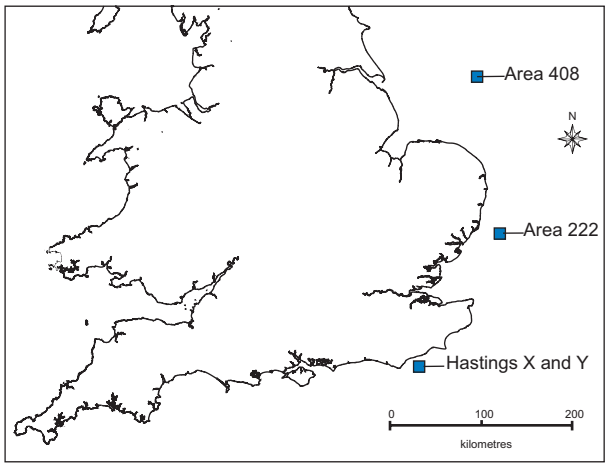

Figure 17. Map showing location of aggregate extraction areas surveyed between 2001-2003

locations which were considered representative of the wider, non-dredged, environment and away from the influence of dredging operations were also sampled

Benthic assemblages and sediment characteristics were determined using a 0.1 m^2 Hamon grab fitted with a video camera and light, and a heavy duty 2 metre beam trawl. Remote survey methods such as sidescan sonar, bathymetric systems, acoustic ground discrimination (AGDS) and photography were also employed to provide an indication of the spatial distribution of sediments, the likely extent of dredging disturbance, and to provide a regional context to investigations. Further detailed information on methodology can be found in Boyd *et al.* (2003, 2004) and Limpenny *et al.* (2002).

11.3 Results

11.3.1 Biology

Relationships between the level of dredging intensity, physical characteristics and assemblages were investigated using a range of univariate and multivariate statistical techniques.

Evidence from this study suggests that the infauna remains in a perturbed state in areas previously exposed to high dredging intensity for a number of years. Complementary surveys of the epifauna similarly show a reduction or absence of a range

Table 17. Main characteristics of the extraction sites studied as part of this research programme

Parameter	Area 222	Hastings Area X	Hastings Area Y	Area 408 (Zone 2)
Geographic location of study site	20 miles east of Felixstowe, southern North Sea	Hastings Shingle Bank, 6 miles south of Hastings, eastern English Channel	Hastings Shingle Bank, 6 miles south of Hastings, eastern English Channel	60 miles east of Humber Estuary, North Sea
Size of licensed area	0.3 km^2	1.35 km^2 (prior to 2001)	3.1 km^2 (prior to 2001)	2.6 km^2
Total quantities extracted over lifetime of dredging activity	10.2 Mt. Unknown proportion of this extracted from outside licensed area	The only dredging campaign prior to 2002 was in 1996, when 1.3 Mt was extracted	Total of 16 Mt extracted during annual campaigns between 1988 and 2000	1.5 Mt in annual campaigns between 1996 and 1999
Lifetime of dredging activity	1971 - 1996	Dredged during 1996, extraction resumed during 2002	1988-2000	1996-1999
Maximum hours of dredging per year in hours, in the high dredging intensity box recorded in 100 m by 100 m area (since 1993)	39.5	28.5	10.25	14.25
Type of dredger employed	Static suction hopper dredger and trailer suction hopper dredgers	Trailer suction hopper dredgers	Trailer suction hopper dredgers	Trailer suction hopper dredgers
Screening	There is limited information from historical records, although it is probable that screening occurred at this site	All-in cargoes	All-in cargoes	Sand returned to seabed as screened material
Water Depth	27-35 m	15-21 m	16-25 m	20-25 m
Geological provenance of the resource	Localised thickened layer of reworked lag deposits ~3 m thick	Infilled palaeovalley >10 m thick	Infilled palaeovalley >10 m thick	Reworked lag sediments in localised lens ~1-2 m thick
Maximum tidal velocity	2.3 kn (1.17 m s^{-1})	2.6 kn (1.32 m s^{-1})	2.6 kn (1.32 m s^{-1})	1.4 kn (0.71 m s^{-1})

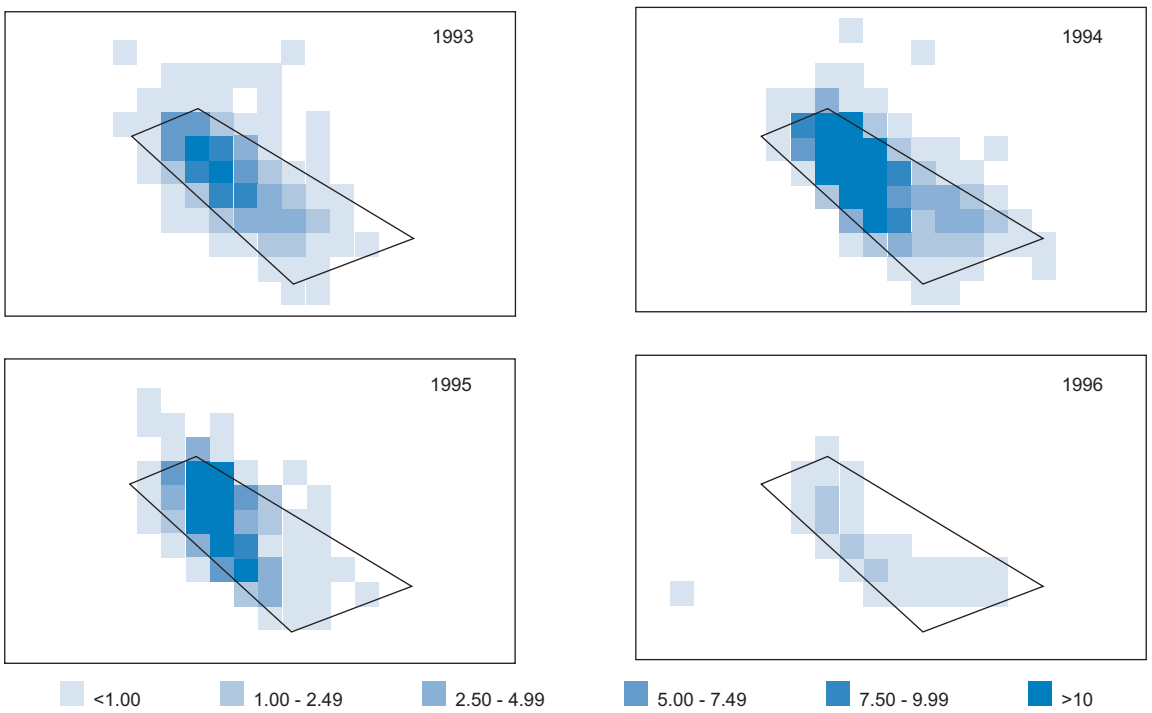

Figure 18. Location and intensity of dredging (in hours) over each 100 m x 100 m block at Area 222 between 1993 and 1996

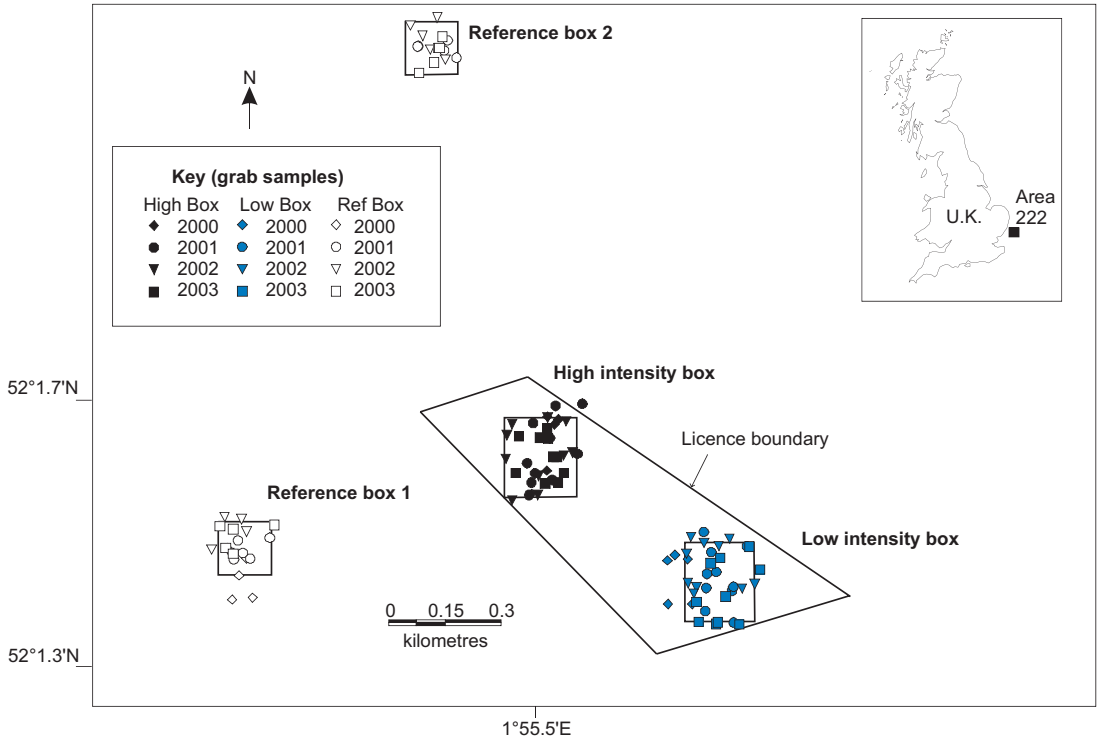

Figure 19. Map showing location of area 222 aggregate extraction licence and sampled stations from the surveys carried out in 2002-2003

of epifaunal species in previously dredged areas, compared to reference locations. There is also evidence of a relationship between the prevailing environmental conditions (tidal current strength and sand mobility) and the composition of epifaunal species in each region.

11.3.2 Physical

Sidescan sonar records have allowed the detection of weathered dredge tracks and pits several years after the cessation of dredging at all four sites (see Figure 20).

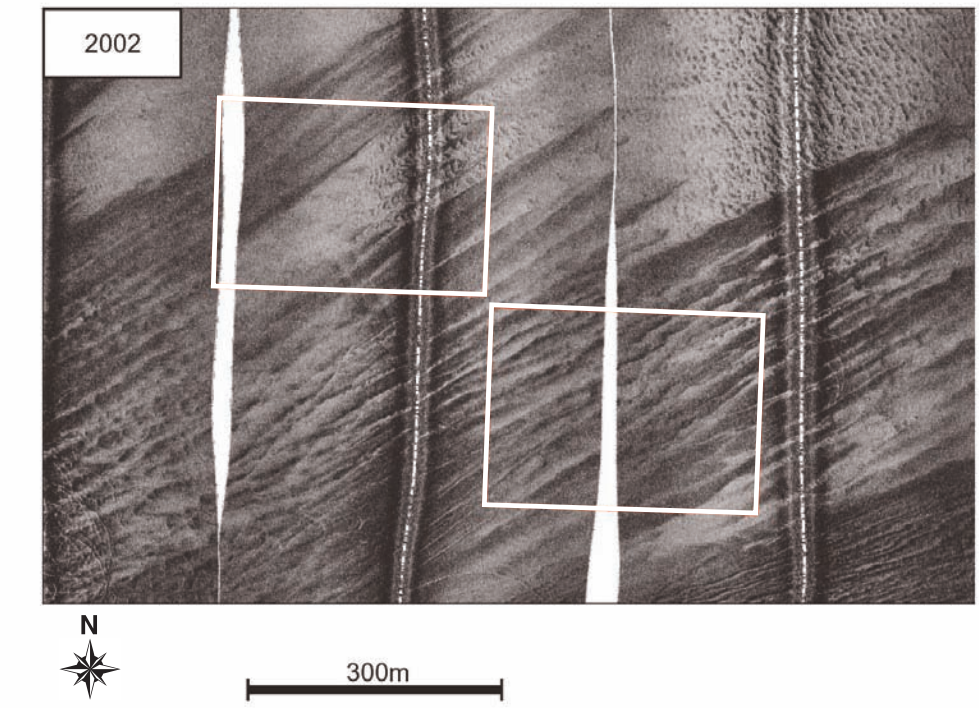

Figure 20. Sidescan sonar image of high and low dredging intensity boxes at Area Y

Sediments collected within the extraction areas tended to be variable in nature and contained more sand in comparison to reference locations. This variability was also evident in the biological samples from dredged locations and may be symptomatic of perturbed conditions.

11.4 Conclusions

Observations from this study indicate that the fauna at areas exposed to high dredging intensities remain in a perturbed state up to 7 years after the cessation of dredging. Therefore, commonly cited recovery rates of 2-3 years should not be assumed to be universally applicable.

This study is unique in providing the beginnings of a consistent time-series of data at contrasting extraction areas around the UK. This information is valuable and is essential for:

- elucidating cause-effect relationships and their amelioration over time, so contributing to better management and monitoring practices
- providing a reliable means to evaluate the significance of aggregate extraction impacts relative to other human (and natural) influences on the seabed
- retrospective testing of the utility and robustness of potential indicators prior to their application in routine environmental management and for understanding the processes involved in recolonisation

Two new measures of hydrodynamic conditions have also been developed and have proved to be an important means of interpreting biological data obtained during this study. Further refinement may improve predictive capabilities regarding environmental effects of dredging activity. A full report of the results arising from this study is given in Boyd *et al.* (2004).

11.5 Future work

This project has advanced our understanding of the ecological effects of marine extraction and the factors likely to be responsible for observed differences in progress towards recovery. Furthermore, the study has shown that information on the nature and rate of physical and biological recovery cannot be uncritically applied to other areas. Therefore, there is a need to establish the long-term biological and physical consequences and scale of the impact of marine aggregate extraction following its cessation in a greater range of habitats and under differing dredging practices.

SEA DISPOSAL

12. LICENSING OF DEPOSITS IN THE SEA

Author: Andrew Dixon and Chris Vivian

12.1 Introduction

This section gives information about the licensing of deposits in the sea around the coasts of England and Wales between 2002 and 2003 under Part II of the Food and Environment Protection Act 1985 (as amended) (FEPA) (Great Britain Parliament, 1985a). In order to provide a complete picture for the UK as a whole, licensing statistics for Scotland and Northern Ireland are also included in this section.

12.2 Legislation and licensing authorities

The deposit of substances and articles in the sea, principally the disposal of dredged material (as opposed to discharge into the sea via pipelines) and the use of material during marine construction and coastal defence works, is controlled by a system of licences issued under Part II of FEPA. Certain operations (e.g. the deposit of scientific equipment or navigation aids) are exempt from licensing under the Deposit in the Sea (Exemptions) Order 1985 (Great Britain Parliament, 1985b).

Following devolution in 1999, Defra (then MAFF) continued to license deposits in the sea around

the Welsh coast on behalf of the Welsh Assembly Government. In Scotland, the licensing function is the responsibility of the Scottish Executive (then SERAD). In Northern Ireland the issuing of licences remained the responsibility of the Environment and Heritage Service, an agency of the Department of the Environment for Northern Ireland.

12.3 Enforcement

Scientists from the CEFAS Burnham Laboratory have the powers to enforce Licence provisions. Visits are made to construction sites and disposal vessels. Samples are taken and records, including logbooks, are checked. Scientific staff carried out 51 inspections between 2002 and 2003.

Officers of the Department's Sea Fisheries Inspectorate (SFI) are charged with enforcing the provisions of FEPA (Part II) and undertake regular inspections from a network of port offices in England and Wales. The SFI carried out 379 inspections between 2002 and 2003 in relation to construction works and the disposal of waste materials (dredged materials and a small amount of shellfish waste) at designated disposal areas. Of these inspections 226 were carried out in 2002 and 153 were carried out in 2003. Further information is set out in Table 18.

In England and Wales 15 written warning letters were issued for apparent breaches of licensing controls between 2002 and 2003 of those 5 were issued in 2002 and 10 in 2003. Details are as follows:

- Investigations into unlicensed construction works in Milford Haven, Pembrokeshire resulted in an official warning being issued in May 2002.
- Investigations into unlicensed construction works at Bowness-on-Solway, North Cumbria resulted in an official warning being issued in May 2002.
- Investigations into unlicensed construction works at a Quay in Southampton, Hampshire resulted in an official warning being issued in June 2002.
- Investigations into unlicensed disposal works on the Helford River in Cornwall resulted in an official warning being issued in September 2002.
- Investigations into unlicensed construction works on the River Colne resulted in an official warning being issued in November 2002.
- Investigations into unlicensed construction works at St Mawes in Cornwall resulted in an official warning being issued in March 2003.
- Investigations into a breach of licence conditions at Mostyn, Flintshire resulted in an official warning being issued in April 2003.
- Investigations into unlicensed disposal works in the Helford River, Cornwall resulted in an official warning being issued in April 2003.
- Investigations into a breach of licence conditions at Padstow Harbour resulted in an official warning being issued in April 2003.
- Investigations into a breach of licence conditions at St Mawes resulted in an official warning being issued in April 2003.
- Investigations into unlicensed disposal works in Bognor Regis, Sussex resulted in an official warning being issued in June 2003.
- Investigations into unlicensed works in Bognor and Worthing, West Sussex, resulted in an official warning being issued in September 2003.
- Investigations into unlicensed construction works at Queenborough, Kent resulted in an official warning being issued in September 2003.
- Investigations into a breach of FEPA between Lancing to Shoreham resulted in an official warning being issued in October 2003.
- Investigations into unlicensed disposal works at Langstone Harbour, Hampshire resulted in an official warning being issued in October 2003.

Table 18. Inspection activity by the SFI during 2002 and 2003

District	2002		2003	
	No. of Inspections	No. of Infringements	No. of Inspections	No. of Infringements
North East	23	0	8	0
Humber	8	0	2	0
East	23	2	32	0
South East	90	8	71	6
South West	23	8	5	1
Wales	45	9	15	0
North West	14	1	13	0
London	0	0	7	1
Total	226	28	153	8

In England and Wales between 2002 and 2003 there were 3 successful prosecutions for illegal marine works. The details are as follows:

- Investigations into unlicensed disposal works at a Quay on the Helford River in Cornwall resulted in a successful prosecution in 2002 where the defendant was fined £1500 and ordered to pay £500 costs.

- Investigations into unlicensed construction works in Hythe which resulted in a successful prosecution in 2002 where the defendant was fined £5000 and ordered to pay £580 costs.

- Investigations into unlicensed construction works in Caernarfon North Wales, resulted in a successful prosecution in 2003 where the defendant was conditionally discharged and ordered to pay £2000 costs.

In Scotland, certain authorised staff of the Fisheries Research Services (FRS) Marine Laboratory, Aberdeen and the Scottish Fisheries Protection Agency (SFPA) hold similar enforcement powers. The FRS made 27 enforcement visits between 2002 and 2003 with 12 being carried out in 2002 and 15 in 2003. The FRS also carried out 9 investigation visits with 3 carried out in 2002 and 6 carried out in 2003. The SFPA made 25 enforcement visits between 2002 and 2003 of which 10 were in 2002 and 15 were in 2003. The SFPA also assisted in 2 investigation visits in 2002 and 4 in 2003.

In Northern Ireland the Environment and Heritage Service (EHS) made 13 enforcement visits of which 7 were carried out in 2002 and 6 were carried out in 2003. EHS also carried out 9 investigation visits of which 6 were carried out in 2002 and 3 were carried out in 2003. This resulted in a warning letter being issued to a local council who promptly removed the unlicensed deposit.

12.4 Licensing of dredged material

Table 19 gives details for the period 1999 to 2003 of the number of sea disposal licences issued, the quantity of waste licensed and the quantity actually deposited, together with information on those contaminants in the wastes, which the UK is required to report internationally to meet obligations under the OSPAR and London Conventions. A proportion of the trace metals in this dredged material is natural, but the mineral structure is such that it will not be available to marine organisms.

Table 19. Summary of dredged material licensed and disposed of at sea, 1999-2003

Country	Year	Licences Issued	Licensed quantity (Tonnes)	Wet tonnage deposited	Dry Tonnage deposited	Quantities of metal contaminants in wastes deposited (tonnes)						
						Cd	Cr	Cu	Hg	Ni	Pb	Zn
England and Wales	1999	131	47,028,123	52,409,430	31,114,127	13.05	1,907	1,064	6.07	898	1,370	4,001
	2000	119	55,902,025	28,257,192	14,077,169	8.76	1,043	663	4.78	485	1,099	2,948
	2001	124	39,297,549	29,660,448	14,881,254	7.61	1,040	731	5.84	478	1,099	3,310
	2002	124	72,851,190	27,884,495	14,725,603	5.53	912	457	4.66	409	1,166	2,664
	2003	97	31,836,123	29,526,580	15,800,897	5.41	950	498	4.29	443	1,183	2,694
Scotland	1999	30	4,044,300	2,352,954	945,563	0.25	57	55	0.78	36	66	130
	2000	30	6,135,400	4,155,018	2,034,213	0.51	87	80	1.79	73	139	298
	2001	29	3,307,800	2,217,981	1,162,856	0.36	79	48	0.74	36	77	165
	2002	21	2,959,045	2,203,016	1,188,129	0.33	59	46	0.85	29	69	134
	2003	29	3,573,981	2,764,020	1,647,881	0.61	70	57	1.40	41	101	175
Northern Ireland	1999	5	1,923,000	2,058,506	768,609	0.54	32	21	0.56	18	23	92
	2000	3	3,950,000	640,815	455,222	0.13	45	7	0.05	13	14	42
	2001	3	183,000	3,420,411	2,495,714	0.72	246	37	0.42	66	76	226
	2002	8	1,161,500	976,102	458,108	0.46	31	19	0.19	19	26	86
	2003	2	189,900	115,404	73,382	1.47	8	4	0.06	3	2	12
UK Total	1999	166	52,995,423	56,820,890	32,828,299	13.85	1,997	1,141	7.41	953	1,459	4,223
	2000	152	65,987,425	33,053,025	16,566,605	9.39	1,176	750	6.63	571	1,252	3,289
	2001	156	42,788,349	35,298,840	18,539,824	8.69	1,365	816	7.01	579	1,251	3,701
	2002	153	76,971,735	31,063,613	16,371,841	6.31	1,003	522	5.70	457	1,261	2,884
	2003	128	35,600,004	32,406,004	17,522,159	7.50	1,027	540	5.75	475	1,270	2,881

Notes: Tonnages deposited relate to quantities in the calendar year 2003, which may be coved by 2 or more licences, including one or more issued in previous years

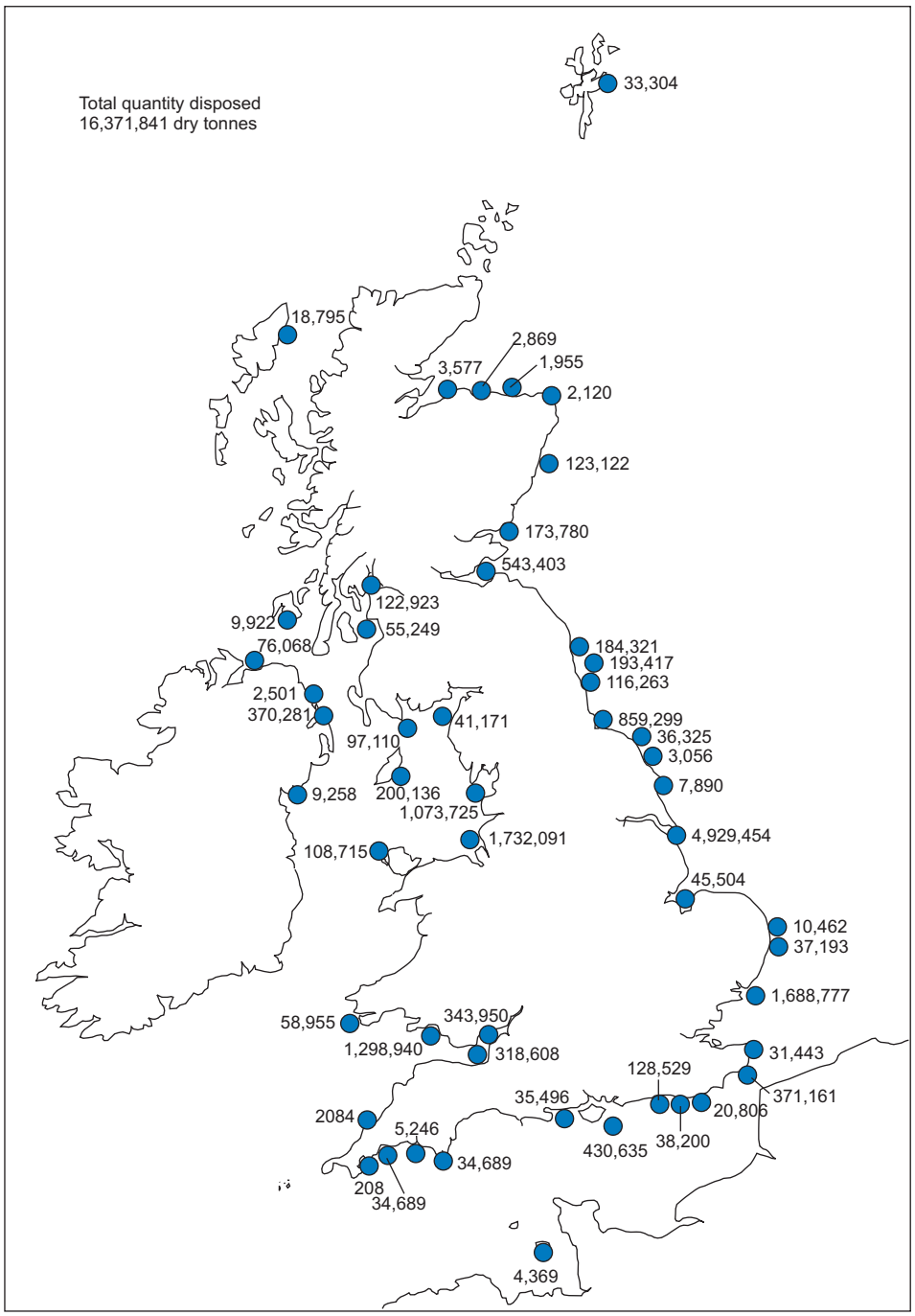

Figure 21. Quantities of dredged material deposited at licensed sites at sea in 2002 (dry tonnes)

Figures 21 and 22 show the main disposal sites used in 2002 and 2003 and the quantities used at each site. Although applications for licences are required to show evidence that they have considered alternative disposal options including beneficial use, the problems of having silty materials, and matching the timing of dredging campaigns and the demand for sediments, have meant that most of the finer materials, in particular, are deposited at sea.

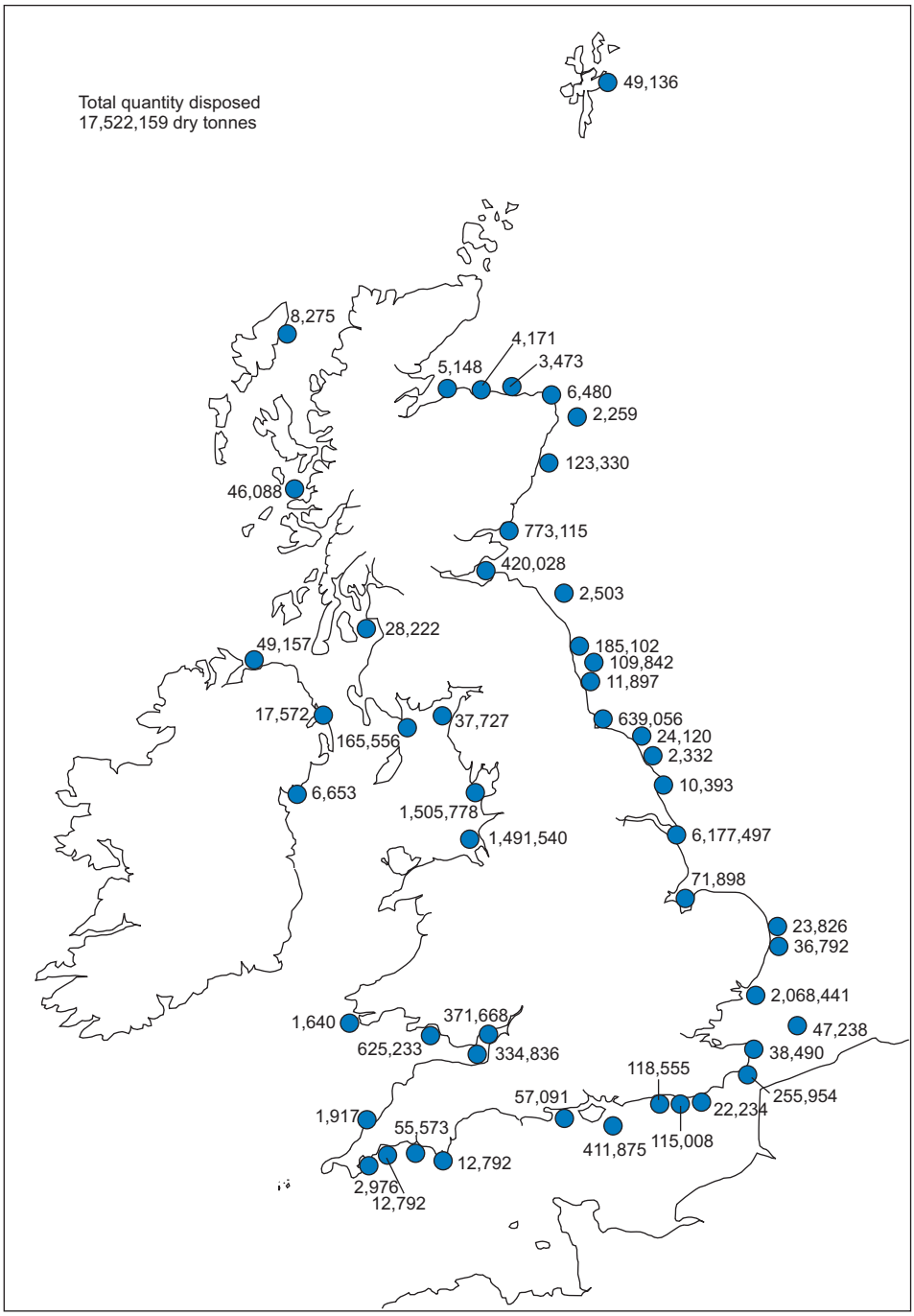

Figure 22. Quantities of dredged material deposited at licensed sites at sea in 2003 (dry tonnes)

12.5 Other licensed activity

Under Part II of FEPA, licences are also required for certain other activities or deposits made below the mean high water springs mark for construction purposes. Each licence application is carefully considered, in particular, to assess the impact on the tidal and intertidal habitat, hydrological effects, potential interference to other users of the sea and risk to human health. Details of these licences issued between 2002 and 2003 are shown in Tables 20 and 21.

Further activities involve the use of tracers, the application of biocides, and burial at sea. Generally the anticipated environmental impact from these deposits is minimal and little or no monitoring is required. Details of these licences issued between 2002 and 2003 are also shown in Tables 20 and 21.

Such licences have also authorised the disposal of a small amount of fish waste. Table 22 provides details of the quantities disposed of between 1999 and 2003.

Table 20. Other categories of licence issued in 2002

Licence category	England and Wales	Scotland	Northern Ireland
Construction – New and Renewals	239	126	2
Tracers, Biocides etc	13	1	0
Burial at Sea	21	0	0

Table 21. Other categories of licence issued in 2003

Licence category	England and Wales	Scotland	Northern Ireland
Construction – New and Renewals	248	99	8
Tracers, Biocides etc	10	1	0
Burial at Sea	14	0	0

Table 22. Summary of fish waste licensed and disposed of at sea, 1999-2003

Country	Year	Licences Issued	Licensed quantity (tonnes)	Wet tonnage deposited	Dry tonnage deposited
England and Wales	1999	1	1,000	956	956
	2000	1	1,000	1,559	1,559
	2001	3	938	687	687
	2002	2	2,200	808	808
	2003	1	6,000	953	953
Scotland	1999	1	200	137	110
	2000	1	200	45	36
	2001	0	0	66	53
	2002	0	0	0	0
	2003	0	0	0	0
Northern Ireland	1999	0	0	0	0
	2000	0	0	0	0
	2001	0	0	0	0
	2002	0	0	0	0
	2003	0	0	0	0
UK Total	1999	2	1,200	1,093	1,066
	2000	2	1,200	1,604	1,595
	2001	3	938	753	740
	2002	2	2,200	808	808
	2003	1	6,000	953	953

13. POLYCYCLIC AROMATIC HYDROCARBONS IN DREDGED MATERIAL FROM PORTS AND NAVIGATION CHANNELS IN ENGLAND AND WALES: 1998-2003

Authors: Carole Kelly, Kerry Baker, Robin Law,

13.1 Introduction

Maintenance dredging is conducted at ports in order to maintain harbour depths and navigation channels. Around England and Wales, during the period 1998-2003, 100-200 licences for disposal of dredged material were issued annually. The quantity of material disposed to sea varied between 28 and 52 million tonnes per annum over the same period. These dredged materials are deposited at defined disposal grounds which are usually close to shore. Before authorisation is granted (under the Food and Environment Protection Act, 1985) the concentrations of selected contaminants are determined in representative samples. Historically, this has involved the determination of trace elements, organochlorine pesticides, chlorobiphenyls, and butyltin compounds. Since 1998, concentrations of polycyclic aromatic hydrocarbons (PAH) have also been determined, with a view to establishing the current levels of contamination and if possible, to develop guideline limit values for use in the approval process.

Approximately 380 samples of dredged material have been analysed within this study to date, from 30 main locations around England and Wales. The results are shown in Figure 23.

From the suite of PAH analysed, which included both parent and alkylated PAH compounds, it is possible to tentatively assign the sources of the PAH observed in each sample. Individual PAH have been assigned as predominantly oil derived or predominantly combustion-derived on the basis of earlier studies using principal components analysis for source discrimination (Law *et al*., 1999). The sums of the predominantly oil-derived and combustion-derived PAH were then calculated and expressed as percentages of the total PAH concentration. The differing source inputs and the total PAH concentrations are illustrated in Figure 23. It can clearly be seen that many areas are affected by mixed PAH source inputs, but that others show mainly single sources (e.g. combustion, Devonport; oil, River Tyne).

The data gathered to date have shown that some sediments which are dredged in order to maintain navigational access to ports show high PAH concentrations which, based on the results of previous studies (Woodhead *et al*., 1999), are in many cases high enough to cause toxicity to sediment-dwelling organisms. This is obviously of concern to those regulating sea disposal of dredged material. Disposal site monitoring for PAH is now being undertaken in order to assess the levels of contamination at these locations. Currently, all this data is being assessed alongside ecotoxicological studies of dredged material with the aim of developing a framework and guidelines for assessment of dredged sediments for use within the licensing process.

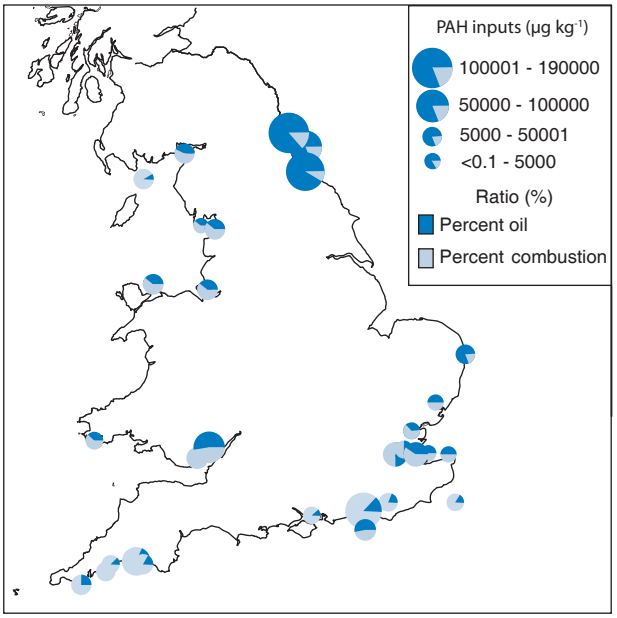

Figure 23. Map showing sites, input sources and sum of PAH in dredged material

14. PRELIMINARY RESULTS FROM POLYCYCLIC AROMATIC HYDROCARBONS ANALYSIS OF SEDIMENT FROM SELECTED DISPOSAL SITES

Authors: Carole Kelly, Robin Law and Lisa Johnsey

14.1 Introduction

Sediment dredged from ports and licensed for disposal at sea can be disposed of to designated disposal grounds. There are over 150 designated disposal sites around the UK not all of which are used in any given year. In 2003, sediments which had been collected from 11 of these disposal sites during 1999-2002, were analysed for a range of polycyclic aromatic hydrocarbons (PAH) and the preliminary findings are given below.

As can be seen from section 13 above, many of the areas which are dredged have elevated PAH concentrations. In areas where samples have been collected from both disposal sites and for a dredging licence application, the data show that the concentrations are generally similar within the estuary and at the disposal site. The highest concentrations were found at disposal sites off the Rivers Tyne, Tees and off Cardiff and Swansea. This is due to these historic disposal grounds still being the most frequently used and taking the highest tonnages of licensed material. One exception was at the disposal site off Shoreham, where PAH could not be detected. It is known from earlier studies that sediments within and around Shoreham Harbour are contaminated with PAHs as a consequence of leaching from a former local gasworks site (Law *et al*., 2002). However the disposal site used for dredged material from Shoreham is very close to shore and in an area of strong longshore drift, which carries disposed material to the east and prevents any build-up of contamination. Assignment of sources as oil or combustion generally also show agreement between the two sample sets, exceptions being where there are small patches of contamination by bulk oil. The source inputs and total PAH concentrations are illustrated in Figure 24.

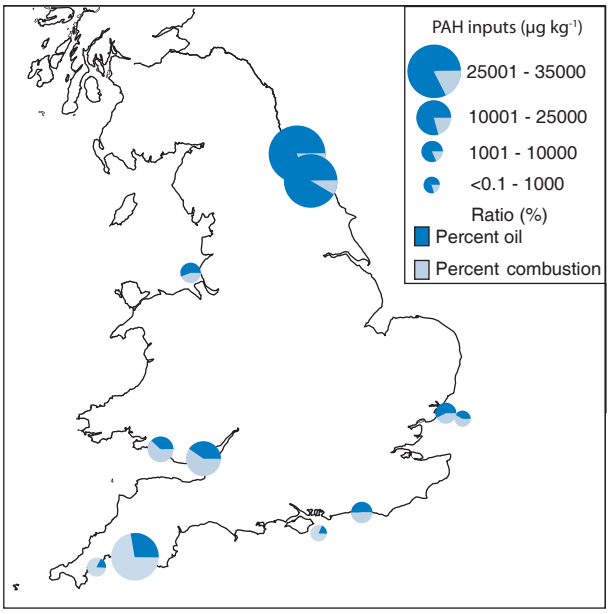

Figure 24. Location of disposal grounds inputs and PAH concentrations

Following on from this preliminary study, PAH analysis of sediments from disposal sites will now be extended to cover other grounds and we also intend to continue monitoring at some sites so as to investigate trends over time.

15. RADIONUCLIDE CONCENTRATIONS IN DREDGED SEDIMENT

Author: David McCubbin

15.1 Introduction

In England and Wales, Defra issues licences to operators for the disposal of dredged material under the Food and Environment Protection Act, 1985 (Great Britain Parliament, 1985a). The protection of the marine environment is considered before a licence is issued. Since dredged material may contain radioactivity, assessments are undertaken, where appropriate, for assurance that there is no significant foodchain or other risk from the disposal. In 2002, a specific assessment was carried out for the disposal of spoil from Heysham Approaches in Lancashire. The Approaches contain artificial radionuclides due to discharges from BNFL Sellafield and the nearby Heysham nuclear power station and from other widespread sources such as weapon test fallout.

15.2 Materials and methods

Samples of surface sediments were collected from a variety of locations to ensure the data provided representative information. Radionuclide assay was achieved using gamma-ray spectrometry by which it is possible to simultaneously measure a wide range of radionuclides commonly found in radioactive wastes.

15.3 Results and discussion

Results from the sediment analyses are provided in Table 23.

The assessment showed that the impact of the radioactivity associated with the disposal operation was very low, below '*de minimis*' levels of exposure. '*De minimis*' relates to doses of the order of 0.010 mSv or less, and guidance on exemption criteria for radioactivity in relation to sea disposal is available from the International Atomic Energy Agency (IAEA, 1999 and IAEA, 2003).

Table 23. Radioactivity in sediment dredged from Heysham Approaches, Lancashire, 2002

Location		Radioactivity concentration (dry), Bq kg^{-1}									
Latitude	Longitude	^{40}K	^{60}Co	^{106}Ru	^{137}Cs	^{212}Pb	^{214}Bi	^{226}Ra	^{228}Ac	^{234}Th	^{241}Am
54° 00.93'N	2° 56.70'W	350	0.86	<2.5	34	9.3	6.0	7.9	8.1	18	31
54° 00.98'N	2° 56.86'W	330	0.82	5.2	28	8.1	5.9	7.4	7.2	*	27
54° 01.08'N	2° 56.78'W	330	0.78	<2.6	30	8.8	6.0	7.7	8.1	20	27
54° 01.03'N	2° 56.57'W	340	<0.27	<2.6	20	7.7	5.4	6.9	6.6	10	18
54° 00.96'N	2° 56.68'W	340	0.59	<2.6	29	8.3	5.6	7.3	7.9	*	29
54° 01.04'N	2° 56.61'W	340	0.62	<2.5	27	8.3	5.7	7.3	7.4	10	25

** Not detected by the method used*

16. TRIBUTYLTIN AT DISPOSAL SITES OFF THE COASTS OF ENGLAND AND WALES

Authors: Bryn Jones and Michaela Schratzberger

16.1 Introduction

There are a number of sites around the coast of England and Wales, which are designated for the disposal of licensed dredged material derived from marine construction sites, harbours and navigation channels (see section 12). Prior to the issue of licences under the Food and Environment Protection Act, (1985), dredged material is evaluated for its suitability for sea disposal, principally in relation to its likely impact on the marine environment. One important compound, which is routinely assessed, is the marine antifouling pesticide tributyltin, (TBT).

Historically, TBT has been used in marine paints in order to control fouling organisms on ships' hulls, which cause increased drag and hence raise fuel costs. TBT is leached from the surface of the paint and thus prevents attachment, particularly of algal slimes. TBT is highly toxic to marine invertebrates generally, with observed effects including imposex in gastropod molluscs, increased mortality of a variety of non-fouling organisms and resultant changes to natural communities in areas subject to contamination (e.g. Waldock *et al*., 1999). In aerobic conditions, TBT breaks down to less toxic dibutyl- and monobutyltin (DBT and MBT). Under anaerobic conditions, TBT can remain unchanged for up to ~30 years or more (Sayer *et al*., 2004). TBT has a strong affinity to bind with sediments, but this is a reversible process, and TBT can be remobilised following sediment disturbance (Watanabe *et al*., 1995).

The IMO International Convention on the Control of Harmful Anti-Fouling Systems on Ships (2001) required the global prohibition, by January 1, 2003, of the application of organotin compounds employed as biocides in antifouling systems on ships. This has now been implemented in the EU by Council Directive 2002/62/EC and, in the UK, approval for their use as antifoulants on ships, granted under the Food and Environment Protection Act or the Control of Pesticides Regulations, has now been revoked. The IMO Convention also imposes a complete prohibition, by January 1, 2008, on the presence of organotin compounds on ships' hulls. The difference of 5 years between the two IMO prohibitions reflects the fact that large ocean-going cargo vessels may be repainted only once every 5 years, and allows for coatings still present following applications to ships' hulls in the period preceding the 2003 ban. These represent the final acts in a regulatory process which, in the UK, commenced in 1987 with the ban on TBT use on small (<25 m in overall length) vessels.

Decisions on whether to allow marine disposal of dredged material are influenced by two action limits for TBT: 1) values above 0.1 mg kg^{-1} dry weight where concerns over the quantity and nature of the material, and characteristics of the receiving area are taken into account and 2) values above 1 mg kg^{-1} which will generally preclude disposal at sea.

The characteristics of receiving areas around England and Wales are very variable, but many are located in areas of high tidal and wave energy, resulting in the dispersion of fine particulates following disposal. A variety of disposal sites are regularly monitored by CEFAS in order that: 1) environmental conditions at newly designated sites are suitable for the commencement of disposal activities, 2) disposal operations conform with license conditions, 3) predictions for established sites concerning limitations of effects are met.

16.2 Collection of samples

Samples were collected during cruises on *RV CIROLANA, RV CEFAS ENDEAVOUR* and *RV PRINCE MADOG* between years 1998 to 2004. The samples

were collected using a Day grab with stainless steel jaws and stored in hexane-rinsed glass jars at -20°C until analysed.

16.3 Method

The analysis of organotin in the sediment was carried out using an extraction procedure utilising a mixture of methanol and 0.1% sodium hydroxide. An internal standard of tripropyltin was added to correct for recovery. The organotin compounds were converted to the respective hydride by the addition of sodium borohydride and extracted into hexane. The butyltin compounds were then determined by gas chromatography with flame photometric detection optimised for tin (GC-FPD) using a Hewlett Packard 6890GC. Quality control was achieved by the inclusion within each batch analysed of at least one blank and certified reference material (BCR 646) and the monitoring of control charts derived from these samples. Results are reported as mg kg^{-1} of butyltins on a dry weight basis.

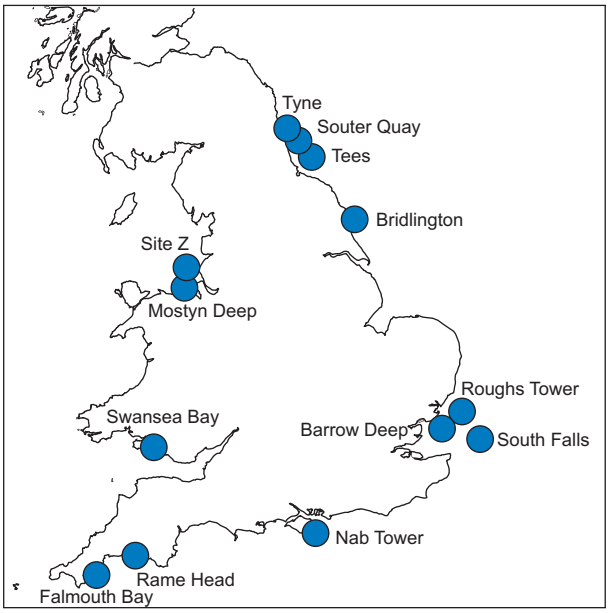

Figure 25. Map of England and Wales with disposal sites monitored

16.4 Results

Samples from Mostyn Deep, Site Z, Swansea Bay, Barrow Deep, South Falls and Bridlington (Figure 24) all contained sediment concentrations of TBT close to or below the detection limit of 0.001 mg kg^{-1} (Table 26). For Rame Head, 1 sample out of 47 contained a concentration of TBT over the action limit 1 (at 0.153 mg kg^{-1}) with all other concentrations close to or below the detection limit. Other sites also contained single stations with high concentrations of TBT but these were below action limit 1, (Falmouth, 0.029 mg kg^{-1}; Nab Tower 0.035 mg kg^{-1}; Inner Gabbard 0.045 mg kg^{-1}; Roughs Tower, 0.073 mg kg^{-1}).

For sites off the North East coast of England, a number of samples showed higher concentrations of TBT, namely off the Tees and the Tyne (North Tyne and Souter Point). At the Tees disposal site, none of the concentrations found was above 0.1mg kg^{-1}, whereas at the North Tyne disposal site, 9 of the 85 stations were above 0.1 mg kg^{-1} and, at the Souter Point disposal site, 7 out of 45 stations were above 0.1 mg kg^{-1}.

Table 24. Mean and maximum concentrations of butyl tins measured

Site	DBT mg kg^{-1} mean	DBT mg kg^{-1} maximum	Number of samples	TBT mg kg^{-1} mean	TBT mg/kg maximum	Number of samples
North Tyne	0.013	0.446	85	0.073	2.815	85
Souter Point	0.008	0.063	45	0.064	0.600	45
Tees	0.003	0.014	36	0.011	0.099	36
Bridlington	0.012	0.023	2	<0.001	<0.001	2
Roughs tower	0.002	0.006	14	0.007	0.073	14
Inner Gabbard	0.001	0.003	17	0.010	0.045	17
South Falls	0.001	0.003	5	<0.001	<0.001	5
Barrow Deep	0.001	0.002	8	0.001	0.003	8
Nab Tower	0.002	0.012	15	0.004	0.035	15
Rame Head	0.002	0.013	47	0.005	0.153	47
Falmouth	0.003	0.008	17	0.004	0.029	17
Swansea Bay	0.005	0.012	6	0.003	0.006	6
Mostyn Deep	<0.001	<0.001	3	<0.001	<0.001	3
Site Z	0.002	0.004	10	0.001	0.004	10

16.5 Discussion

All of the sites except for the Tyne were found to have low to background concentrations of TBT. At Rame Head and Falmouth, relatively high values at one station were encountered at each location. Of the 310 stations sampled, only one sample exceeded the action limit 2 of 1 mg kg^{-1}. This sample was from the North Tyne site. Both this and the Souter Point site generally contain relatively higher concentrations of TBT than elsewhere. The uneven nature of butyltin concentrations reflects an uneven distributions of TBT within the sediment, as it may be present both adsorbed to sediment particles and as TBT- coated paint particles, in which the TBT concentrations are much higher.

Historically, The Tyne and Souter sites have received dredged material associated with the maintenance or development of the port and ship-building facilities (including dry docks) located on the River Tyne, which may account for elevated levels of TBT compared to other disposal sites. However, the overall mean TBT concentrations at these two sites remain below the action 1 limit (Figure 26).

In May 2000, a meiobenthic field survey conducted along a transect from the outer Tyne estuary to the Souter Point dredged material disposal ground revealed that the structure of assemblages of meiobenthos collected at two stations within the disposal ground differed significantly from that recorded at two reference sites to the South. A statistically significant relationship between nematode assemblage structure and TBT concentrations in the sediment indicated that TBT might be responsible for the observed differences. Although the field assessment provided information on general species distribution patterns and helped to identify a potentially adverse effect of high TBT concentrations in the sediment on benthic biota, conclusive cause-effect relationships could not be demonstrated by field data alone because of the broad spectrum of environmental variation. This problem was overcome by the design of an appropriate small-scale community-level experiment in the laboratory which provided a means of assessing the specific response of meiofaunal assemblages to a variety of experimental treatments under replicated, controlled and repeatable conditions (Schratzberger *et al*., 2002). This study unambiguously showed that the response of nematode species depends not only on the level of TBT contamination, but also on the duration and mode of exposure to contaminated sediment. This must be taken into account when assessing the effects of TBT on aquatic communities. The combined results of research conducted in the field and the laboratory has improved our understanding of the response of benthic communities to ecologically relevant and realistic TBT concentrations in the sediment by showing that structural (e.g. diversity, species composition) and functional (feeding types, length distributions) attributes of meiobenthic communities can serve as good indicators for the ecological effects of TBT contamination.

16.6 Conclusion

TBT concentrations at the majority of disposal sites provide strong evidence that the present screening process prior to the issue of disposal licenses has been effective in minimising contamination of sediments at sea.

The mean concentrations at most sites are an order of magnitude below action limit 1. While the mean TBT concentration at the North Tyne and Souter Point disposal sites (0.073 mg kg^{-1} and 0.064 mg kg^{-1} respectively) are below the action limit 1 of 0.1 mg kg^{-1}, the levels are proportionally higher than elsewhere. Actions presently being taken are designed to promote a reduction of concentrations at these sites to ensure that no unacceptable environmental consequences occur. Enhanced monitoring effort will continue at these two sites.

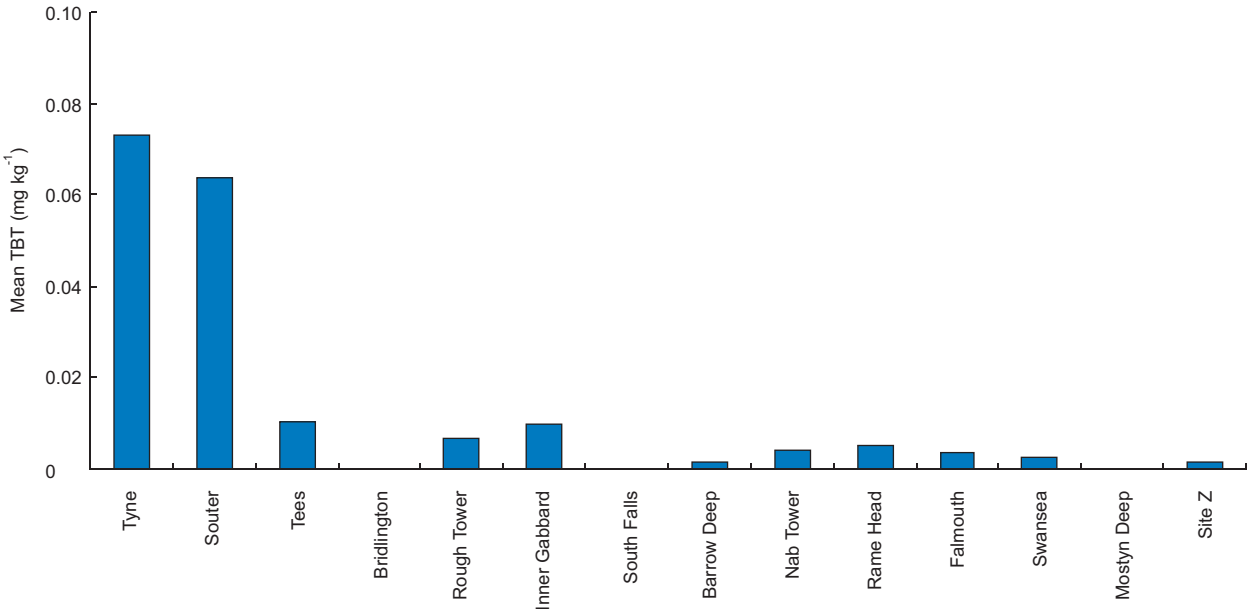

Figure 26. Mean TBT concentration at disposal sites

17. MONITORING THE EFFECTS OF DREDGED MATERIAL DISPOSAL OFF PLYMOUTH, UK

Authors: Rebecca Smith, Hubert Rees and David Limpenny

17.1 Introduction

CEFAS have been sampling annually at the Rame Head disposal site since 2001 in support of Defra's regulatory role under FEPA (Great Britain Parliament, 1985b). This report deals mainly with the outcome of benthic sampling effort in 2002 and 2003, and also includes the results of acoustic and camera survey work that was carried out in 2001 and 2002.

The recent increase in CEFAS survey effort was prompted, in part, by local concerns about the impact of the dredging operations on the area, including litter being washed ashore and higher than usual siltation levels on a popular dive spot, namely the wreck of the *JAMES EGAN LANE*.

17.2 Methods

The survey was designed in alignment with the tidal flow and also to take account of the possible inshore transport of disposal material from the site. In 2001, single samples were taken at 23 stations for later analysis of the benthic macrofauna, sediment particle size distribution and the concentration of a range of trace contaminants with a 0.1 m^2 mini Hamon grab and a Shipek grab. Also, a sidescan survey was conducted to assess the spatial distribution of substratum type. For the 2002 survey, the number of stations was reduced to eleven, at which repetitive samples for the benthic fauna and sediments were collected along with single samples for contaminants (Figure 27). Underwater video footage was also obtained both inside and outside of the disposal site.

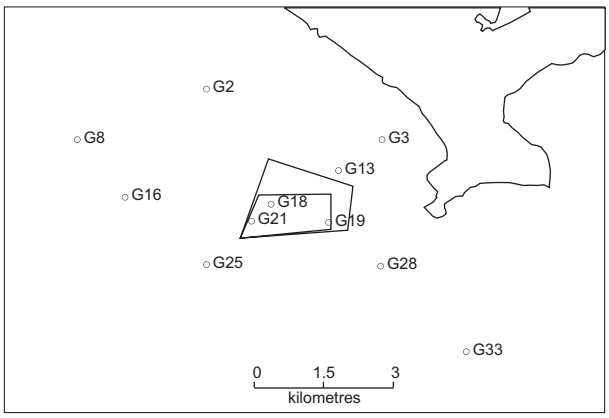

Figure 27. 2002 sampling effort

The 2003 survey built on the 2002 sampling effort, with 9 stations being sampled for benthos and sediments (filled circles), with the addition of 13 inshore sampling locations (open circles) in response to concerns that disposed material was moving southwards into the bay and then deposited along the shore line (Figure 28). These were sampled using a hand held van Veen grab and sediment was collected for analysis of the concentration of a range of trace contaminants.

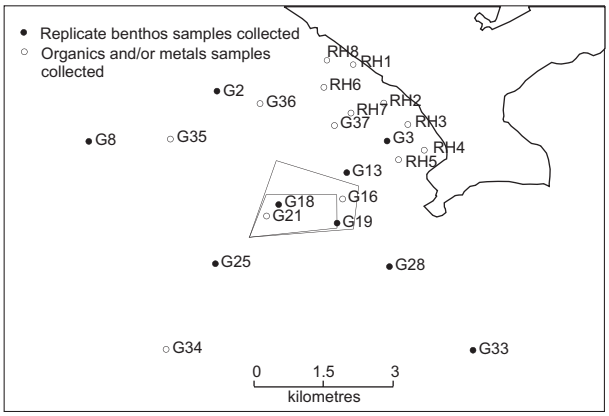

Figure 28. 2003 sampling effort

17.3 Preliminary results

17.3.1 Sidescan sonar

The sidescan survey carried out in 2001 indicated a high variety in substratum type and the presence of dredged material was also detected within the disposal site Figure 29).

17.3.2 Underwater video

The video footage from 2002 confirmed the variability in substratum type shown in the sidescan mosaic but also highlighted the presence of sections of cable and other discarded items in the southeast corner of the disposal site.

17.3.3 Benthic macrofauna

A sub-set of samples from 7 stations (see Figure 30) have been analysed from the 2002 and 2003 surveys to allow an analysis of the status of the benthos within and outside of the disposal sites in each year and also to show a comparison of any change between years.

A total of 191 species were recorded in 2002 of which 68 occurred only once. In 2003, 233 species were recorded and 92 of these were only counted once. The most abundant species in both years were all polychaete species, namely *Scalibregma inflatum*, *Magelona filiformis* and *M. johnstoni*, *Mediomastus fragilis* and *Melinna palmata*.

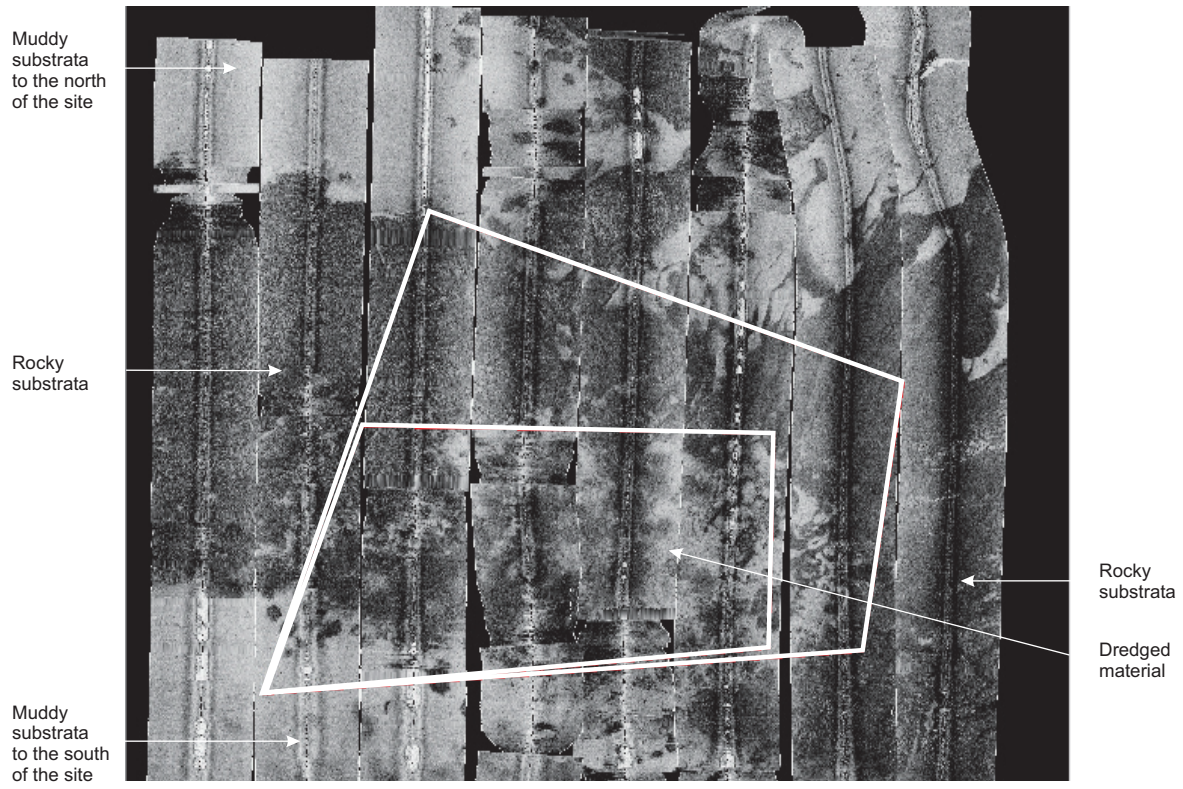

Figure 29. Mosaiced sidescan image collected in 2001 at Rame Head

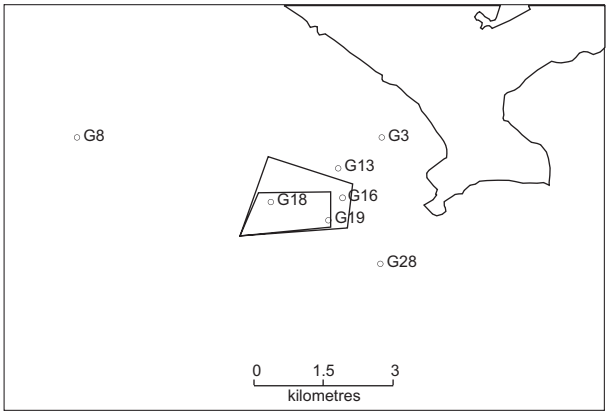

Figure 30. Locations of a sub-set of stations from 2002/2003 for analysis of the benthic macrofauna

Univariate analyses of the samples show that those within and in the vicinity of the disposal site (G16, G18, G19, G3 and G13) had reduced numbers of species (Figure 30a) and individuals (Figure 30b) compared to the two stations located at a greater distance from the disposal site (G8 and G28) in both 2002 and 2003. Numbers of species and individuals were also generally higher in 2003 compared to 2002 with the exception of numbers of species at G3.

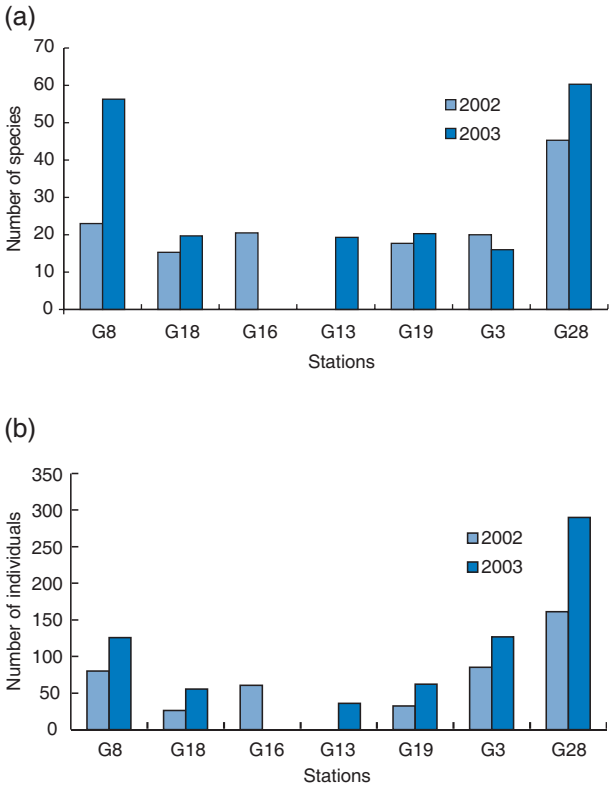

Figure 31. Numbers of (a) species and (b) individuals

17.3.4 Sediments

A sub-set of sediment samples have been analysed from 2001, 2002 and 2003 for particle size and contaminants (metals/hydrocarbons/TBT) and results are currently undergoing full validation and interpretation before reporting at a later date. Preliminary observations, both at sea and as a result of laboratory analyses on sediment quality include the following:

Sediments collected from within the disposal site at G18 in 2001 were seen to have an oily sheen. Chemical analysis of the samples found elevated levels of hydrocarbons at this station (similar field observations were also made at G19 and G33 in 2004). Samples collected at G18 in 2003 contained fragments of sanitary towel.

Field observations of samples collected in 2004 from G18, G19 and G28 were noted to contain items of rubbish such as wire, brass screws, plastic, glass and sanitary towel fragments.

TBT concentrations were either very low or below detection limits at all stations.

17.4 Discussion

The difference in numbers of species and individuals between years may be explained by the disposal history of the site (See Figure 32). When sampling was carried out in 2002, the last disposal operation was approximately 6 months previously and was relatively large (~200,000 tonnes) which could have had a detrimental and longer lasting effect. Sampling in 2003 was preceded by approximately four months of relatively minor disposal (max. 35,000 tonnes per month) in comparison to previous dredging campaigns. Overall, there appears to be evidence of a predictable impact on the benthic fauna from disposal operations, but these are largely confined to the disposal site. A full account of the outcome of survey work to date, including inter-relationships between the benthic fauna and trace contaminants in sediments is in preparation for reporting in 2005.

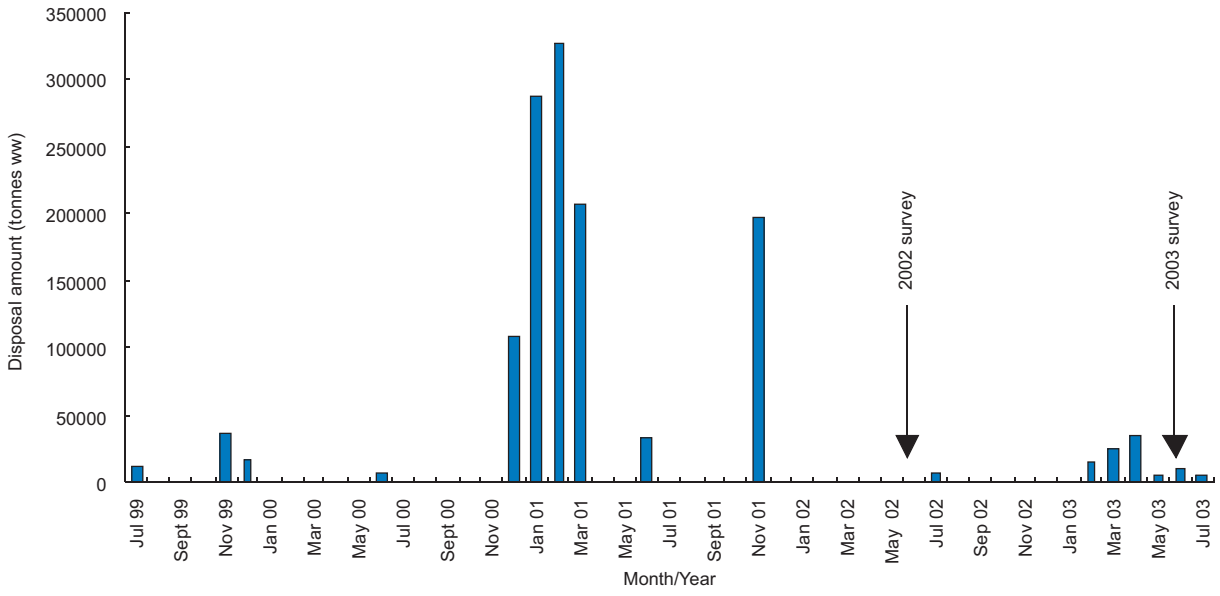

Figure 32. Disposal operations at Rame Head

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