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# Scope for growth and contaminant levels in North Sea mussels *Mytilus edulis*

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ABSTRACT: Mussels *Mytilus edulis*, collected from 26 coastal sites from the Shetland Islands to the Thames estuary and 8 offshore light vessels, were used to monitor changes in environmental quality along the North Sea coastline of the UK (July 1990 and August 1991). The combined measurements of the stress response, scope for growth (SFG), and chemical contaminants in the tissues of mussels were able to detect, quantify and identify some of the major toxicants causing the observed pollution effects. SFG declined from north to south, reflecting both the major inflow of clean water from the North Atlantic via the north of Scotland, and the overall increase in environmental contamination with increasing urbanisation and industrialisation towards the south. There were coastal regions (e.g. Humber-Wash area and the Thames estuary) as well as specific sites (e.g. Ythan, Montrose, Blyth, Teesmouth, Whitby) which showed markedly reduced SFG. Using experimentally derived tissue concentration-response relationships it was shown that at over half the sites the reduced SFG could be entirely explained by the recorded concentrations of contaminants in the tissues. At the majority of sites, a large contribution towards the observed decline in SFG was caused by toxic (mainly polyaromatic) hydrocarbons, largely reflecting urbanisation and shipping activity. In addition, reductions in SFG appear to be partially explained by the accumulation of significant amounts of 'polar organic compounds' and tributyltin. At no sites were metals accumulated to concentrations that could cause a significant reduction in SFG. At those sites with a large 'unexplained component' to the very low SFG values, there was a significant correlation between this 'residual unexplained toxicity' and concentrations of organochlorines in the mussels. More research on the toxicity of these organochlorine compounds to mussels is needed.

KEY WORDS: North Sea · Pollution · Mussels · Monitoring

## INTRODUCTION

A summary report on the 'Quality Status of the North Sea' prepared in 1987 for the Second International Conference on the Protection of the North Sea (DoE 1987) concluded that: 'In general, deleterious effects have only been shown in the immediate vicinity of identifiable pollution sources, and more work needs to be carried out to establish how extensive these effects are. Improved monitoring and scientific programmes need to be developed to provide more consistent data and to permit links between inputs, concentrations and effects to be established with greater confidence'. This conclusion represents a major objective of the present study.

To date, North Sea monitoring programmes have focussed primarily on the concentrations of chemical contaminants accumulated in biota (Franklin 1987, 1992) and on the composition of benthic communities (reviewed by Rees & Eleftheriou 1989). While these are important components of any monitoring programme, they have major limitations and need to be complemented by more sensitive sublethal biological effects measurements and ones that have the potential to identify the causes of any observed deleterious effects. Establishing environmental quality must ultimately be a combination of physical-chemical (cause) and biological (effect) measurements, but this cannot be achieved using the current approach for the following reasons:

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(1) Chemical monitoring programmes are usually selective, primarily focussing on those classes of contaminants that can be easily measured at relatively low cost (e.g. metals) or those of 'known concern' in a particular area. This selectivity is inevitable due to: (a) the large number of potentially toxic contaminants (i.e. >100000 compounds released into the environment) and their degradation products, (b) their chemical diversity, and (c) the time-consuming and costly nature of the chemical analyses. Therefore all potentially toxic contaminants cannot be monitored solely by chemical measurements, hence the need for an integrated biological measure of environmental quality to complement the chemical analyses.

(2) While the species diversity and the well-being of communities are the ultimate concern, indices of community change are: (a) relatively insensitive, (b) slow to respond (based on lethal effects and recruitment), (c) largely descriptive and retrospective, and (d) labour intensive/time consuming.

Consequently, chemical and community analyses need to be complemented by more sensitive, integrative and sublethal effects measurements that are both predictive (anticipatory) of likely population effects and allow toxicological interpretation of complex mixtures of contaminants and identification of causality through mechanistic understanding of readily established concentration-response relationships and QSARs (Quantitative Structure-Activity Relationships).

Since the mid-1970s the common mussel *Mytilus edulis* has been widely used as a 'sentinel' organism in 'Mussel Watch' monitoring programmes to assess the spatial and temporal trends in chemical contamination in estuarine and coastal environments (reviewed by Widdows & Donkin 1992). However, none of these programmes have included accompanying biological measurements to establish whether the contaminant levels were inducing adverse biological effects or whether all potential toxicants were being analysed. During the 1980s research was concerned with the development and evaluation of biological stress responses that will serve to complement these chemical monitoring programmes and provide a toxicological interpretation of tissue residue chemistry data. To date, many field studies including international workshops organised by the Group of Experts on the Effects of Pollutants (GEEP) of the Intergovernmental Oceanographic Commission (IOC) have consistently shown that the physiological response termed 'scope for growth' (SFG) is one of the most sensitive measures of pollution induced stress (reviewed by Widdows & Donkin 1992, Widdows 1993).

SFG reflects the balance between processes of energy acquisition (feeding and digestion) and energy expenditure (metabolism and excretion), and thus provides an instantaneous measure of the energy status of an animal. This can range from maximum positive values under optimal conditions, declining to negative values when the animal is severely stressed and utilising body reserves. While direct measurements of total production and growth rate are often difficult to quantify and interpret in relation to pollution (see Widdows & Donkin 1992), SFG is rapidly determined, providing a sensitive, quantitative and integrated response which can be related to the tissue residue chemistry. It is also provides insight into the underlying mechanisms of toxicity and the components which effect changes in growth rate (Widdows & Donkin 1992).

Previous field studies (reviewed by Widdows & Donkin 1992) have successfully used the combined measurement of biological effects (SFG) and chemical contaminants in mussels to detect, quantify and identify the causes of pollution gradients in estuaries and bays (typically over a relatively small spatial scale of ca 10 km). The aim of this North Sea study was to use mussels to quantify environmental contamination and pollution effects over a larger spatial scale (>1000 km). To achieve this overall objective it was necessary to conduct some preliminary studies in order to establish the most appropriate and effective procedures. Firstly, an important consideration in any environmental pollution study is the selection of appropriate 'clean/uncontaminated' reference sites. The dominant water movement into the North Sea is from the North Atlantic via the north of Scotland and southwards along the coastline of eastern Scotland and northeast England (Reid et al. 1988). Previous studies (Widdows et al. 1987, 1995) confirmed that mussels from specific sites in the Shetland Islands, UK, consistently have the lowest recorded levels of anthropogenic contaminants (e.g. metals, alkyltins, and hydrocarbons). These northern sites therefore represented important 'clean/reference' sites for the North Sea study. Secondly, it was necessary to modify procedures enabling mussel samples to be transported to a single laboratory for measurement, because the use of a mobile laboratory was considered costly and impractical over such a large spatial scale. Thirdly, it was necessary to establish that any recorded differences in the physiological performance of mussels collected from the north and south were due to environmental factors rather than inherent (genetic) differences. After establishing appropriate procedures, the primary objectives of this study were:

(1) To quantify the degree of pollution by measuring the physiological stress responses (scope for growth) and the concentration of major metal, organometal and organic contaminants in the tissues of mussels collected from sites around the North Sea.

(2) To provide a toxicological interpretation of the tissue residue data based on the available experimentally derived relationships between the tissue concentrations of toxicants and sublethal physiological responses.

## MATERIALS AND METHODS

**Preliminary studies.** Initial studies were concerned with evaluating modifications to the established procedures for sampling mussels and measuring their physiological responses (Widdows 1985, 1993, Widdows & Johnson 1988).

***Transportation of samples:*** Mussels *Mytilus edulis* from all potential sampling sites in the UK can be transported to Plymouth by commercial delivery services within 24 h. To avoid shell valve gape and to minimize stress, bivalves were held in air and at low temperatures (i.e. between ca 5 and 8°C in an insulated container with ice packs). Preliminary experiments therefore examined the time-course of recovery after a 24 h period of air exposure at a temperature of 7°C. Mussels of 4 cm shell length were collected from 3 sites in southwest England (Whitsand, Exmouth and Tamar), packed in thermally insulated containers (using disposable nappies to provide an absorbent, thermal buffer between the mussels and the ice packs) and transported to the Plymouth Marine Laboratory (PML). After 24 h, 16 mussels from each site were placed in seawater (clean offshore Eddystone seawater, EFSW) at 15°C, salinity 32 psu, and fed an algal diet of *Phaeodactylum tricornutum* (cell concentration of $9 \times 10^3$ cells $ml^{-1}$). Clearance rate, defined as the volume of water cleared of particles per hour, is generally the most sensitive component in response to stress and was therefore measured at frequent intervals over a period of 72 h. Other physiological components (respiration rate and absorption efficiency) were measured after 24 h recovery. In addition, 2 groups of 5 mussels from each population were sampled after 24, 48 and 72 h in EFSW and analysed for 2- and 3-ring aromatic hydrocarbons (see below for details).

***Comparison between field-laboratory measurements (testing of water quality at PML):*** The high quality of EFSW used in the aquaria (3 $m^3$ total volume) at PML was tested by comparing the physiological responses of mussels measured first in the field at a known clean site in the Shetlands and then in EFSW at PML. Mussels were collected from an established clean reference site at Gluss Voe in the Shetlands (Widdows et al. 1987, 1995) and measured in the field using a mobile laboratory. Mussels were then packed in insulated boxes and held in air at 7°C and transported back to PML. After 24 h of air exposure, they were allowed to recover for 24 h in EFSW (3 $m^3$ aquarium) before measuring their physiological responses (clearance rate, food absorption efficiency and respiration rate).

***Population differences in physiological responses (environmental vs genetic):*** The hypothesis that 'observed differences in physiological responses of mussels collected from populations ranging over a distance of 1000 km may be the result of inherent (genetic) rather than environmental differences' was tested. Previous studies have examined this aspect over smaller spatial scales (e.g. between 2 Scottish lochs, Okumus & Stirling 1994; Tamar and Swansea, Widdows et al. 1984; Baltic and North Seas, Kautsky et al. 1990) and demonstrated that recorded differences were environmentally induced. In this study, mussels were transplanted from the Shetlands to the Tamar estuary (i.e. the 2 extremes of the spatial/latitudinal gradient) where they were caged and exposed to their new environment for 12 mo. In June 1989 their physiological responses were then measured at the PML under standard conditions (EFSW, 15°C, salinity 32 psu, *Phaeodactylum* algal ration).

**North Sea study Phase I — coastal sites.** In Phase I mussels were collected from 26 coastal sites bordering the North Sea. The sites were selected on the basis of:

(1) Sites previously sampled by MAFF (Ministry of Agriculture, Fisheries and Food, UK) and SOAFD (Scottish Office Agriculture & Fisheries Department, UK) as part of their 'Monitoring Programme of Contaminants in Fish and Shellfish'

(2) Sites with an established intertidal mussel population containing a range of size classes including those of ca 40 mm shell length.

(3) Sites reflecting general environmental quality in the coastal zone and at the mouth of estuaries, rather than at specific 'hot-spots' or sites near industrial inputs.

(4) Sites ranging from the Shetland Islands at the northern entrance to the North Sea (representing established 'clean' reference sites) to the north Kent coast at the southern entrance of the North Sea (i.e. English Channel).

**Outline of experimental procedures.** Mussels were sampled from 7 sites in Scotland and 19 coastal sites in England between 27 June 1990 and 20 July 1990 (Fig. 1). A total of 200 mussels (mean shell length 41 mm ± 0.2 SE) were collected from 1 or 2 sites per day. Salinities at all sites were within the range 30.2 to 35 psu, with the exception of Whitby Harbour (17.4 psu). The prevailing water temperatures at sites from Gluss Voe to Humber Bull Fort ranged from 11.4 to 15.1°C, whereas sampling between Cleethorpes and Whitstable coincided with elevated summer temperatures and the prevailing midday seawater tempera-

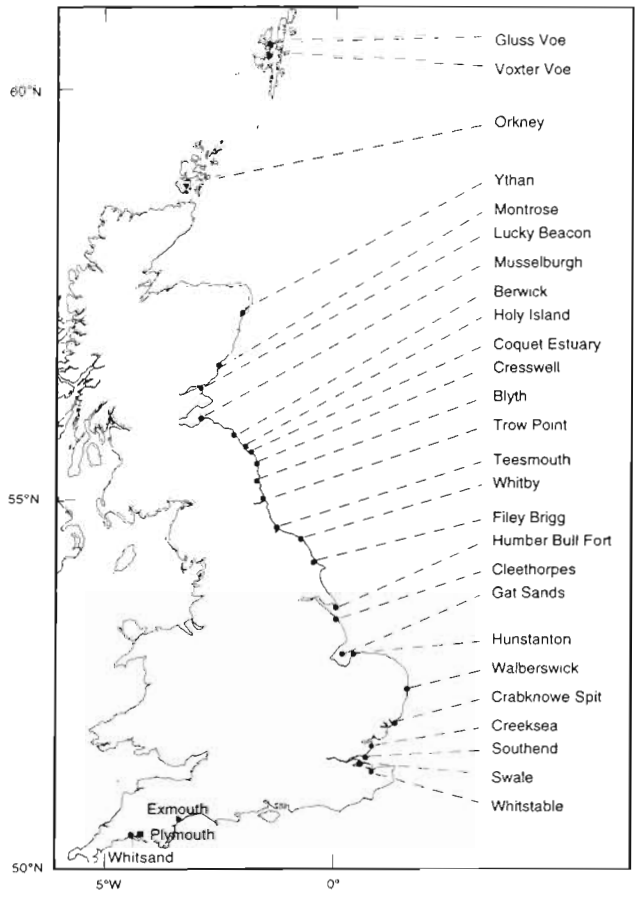


Fig. 1 Locations of coastal sampling sites

tures ranged from 17.2 to 23°C. Mussels were immediately packed in polystyrene insulated containers with 4 frozen ice-packs (2 at the bottom and 2 at the top) with thick absorbent material providing insulation between the mussels and the icepacks. The containers were sealed and transported to the PML within 24 h via express delivery services. Mussels from all sites remained boxed for a standard period of 24 h (from the time of sampling on the shore), before unpacking, recording the temperature and reimmersing in EFSW.

The 200 mussels were divided into 3 groups:

(1) 'Physiology' mussels (n = 25) of a standard size [mean shell length = 41.7 ± 0.04 mm (SE); mean dry tissue weight = 0.50 ± 0.03 g] were cleaned of epibionts and sediment, placed in the aquarium, and allowed 24 h to recover from aerial exposure/transportation before measurement of the physiological responses, clearance rate, respiration rate, food absorption efficiency and SFG. Physiological responses were determined under 'standard' conditions (15°C, salinity 33 psu and an algal cell concentration of 0.4 mg $l^{-1}$).

(2) 'Inorganic/metal chemistry' mussels (n = 50) were cleaned and held in EFSW for 8 h to allow the discharge of faecal/pseudofaecal material with a significant metal content.

(3) 'Organic chemistry' mussels (n = 50 for MAFF, n = 25 for PML and n = 50 as a reserve stock) were cleaned and held in EFSW for a shorter period (1 h), to minimise depuration of more polar contaminants.

All mussels were frozen and stored at −25°C until analysed for chemical contaminants.

The following contaminants were analysed:

- Metals (Cd, Cu, Hg, Pb, Zn);
- Alkyltins (tributyltin, TBT; dibutyltin, DBT);
- Organochlorines (dieldrin; dichlorodiphenyl-trichloroethane, DDT; hexachlorobenzene, HCB; α- and γ-hexachlorocyclohexane, HCH; polychlorinated biphenyls, PCBs);
- Total hydrocarbons;
- Aromatic hydrocarbons (2- and 3-ring compounds);
- Polar organic compounds.

Additional measurements were carried out during the course of the sampling period:

(1) Mussels were collected from Whitsand (S Cornwall) at the beginning and the end of the sampling period in order to test whether there were any significant changes in the physiological responses over this period of time.

(2) Measurements of mussels from the clean reference sites in the Shetlands were performed after 24 h in EFSW at PML and at intervals over the next 21 d to test for any significant changes in water quality during the overall period of measurement.

**North Sea Phase II — offshore light vessels.** In Phase II, 2000 mussels (ca 40 mm shell length) were collected from Exmouth (S Devon) on 15 July 1991. A total of 50 mussels were placed in each plastic-coated wire mesh cage (18 × 18 × 3.5 cm) and then held for 2 wk in EFSW, recirculated in a 3 $m^3$ aquarium at the PML, to allow byssal attachments to form before transplanting to sites in the North Sea. Permission was obtained from the relevant authorities, Trinity House Lighthouse Service (London) and MAFF, to suspend cages of mussels from 10 offshore Trinity House light vessels anchored in the North Sea between the Humber and the Dover Straits. The light vessels from north to south (see Fig. 2) were 'Dowsing', 'Dudgeon', 'Newarp', 'Smiths Knoll', 'Shipwash', 'Sunk', 'Falls', 'East Goodwin', 'Sandettie', and 'South Goodwin'.

Four cages containing a total of 200 mussels were transplanted to each site using an 18 m vessel ('Greyhound Tracker' of Bure Marine Ltd), between 5 and 7 August 1991. At each site the cages were mounted horizontally in a stainless steel frame suspended 4 m below the surface on 6 mm stainless steel wire and weighted to prevent streaming in the water currents. After 6 wk exposure to the environmental conditions in the North Sea, mussels were collected from 2 sites per

day between 16 and 20 September 1991 and transported to the PML using express delivery services. The procedures for sampling, transportation and measurement were identical to those used in our 1990 coastal survey.

Mussels were also sampled from Exmouth on 23 September 1991, and held in identical conditions to the transplanted mussels prior to physiological measurement and chemical analysis of body tissues.

**Chemical analyses.** Metals, organochlorines and total hydrocarbons were analysed according to the standard protocols described in MAFF reports (Franklin 1987, Law et al. 1988, Harper et al. 1989). Total hydrocarbon fluorescence was measured with an excitation wavelength of 310 nm and emission wavelength of 360 nm, and calibrated with Ekofisk crude oil.

***Aromatic hydrocarbon analysis:*** Two groups of 12 mussels from each site were dissected immediately following the 1 h depuration. The resulting tissues were bulked into 2 solvent-cleaned glass jars with foil-lined screw caps and stored frozen (–25°C). In the first stage of the analytical procedure the tissues were thawed, homogenised at 0°C, and 6 g subsamples extracted by saponification/steam-distillation into n-hexane. The distillates were analysed for 2- and 3-ringed aromatic hydrocarbons by means of high performance liquid chromatography (HPLC) on an amino-cyano column (Whatman Partisil-5 PAC, 250 × 2 mm) eluted with n-hexane at a flow rate of 0.6 ml $min^{-1}$ (Donkin & Evans 1984). The 2- and 3-ring aromatics were quantified by reference to 2,3-dimethylnaphthalene and 1-methylphenanthrene respectively. Each distillate was analysed twice by HPLC and analysis repeated if the results fell outside the range of ±5%. A mean value for the 2 groups of 12 mussels was then calculated.

***Polar organics:*** The distillates prepared as above were also analysed using a modified HPLC procedure designed to measure polar hydrocarbons of the type detected in Bermudan mussels by Burns et al. (1990). The column was eluted, beginning with a 1:1 mixture of n-hexane and dichloromethane, followed by a linear gradient to 100% dichloromethane at 15 min. Eluting compounds were detected by means of a diode array UV detector set at 300 nm with a bandwidth of 50 nm. This procedure produced a distinctive peak doublet from most samples, with retention times similar to those of phthalate plasticisers. Evidence of identity was sought by scanning the UV spectrum of the peaks and by transferring material from the HPLC peaks to capillary gas chromatography [GC; 15 m × 0.32 mm SE-54 (methyl phenyl) polysiloxane column]. GC retention times of the unknown polar compounds and phthalates were compared. The peaks were quantified on HPLC relative to dioctyl phthalate, then corrected using a phthalate/unknown intercalibration factor determined by GC. Since the GC with flame ionisation detection largely responds to carbon, estimation of the quantity of the unknown polar compounds by this means is more accurate than HPLC/UV quantification using an arbitrarily selected standard. HPLC fractions containing the peaks of interest were also analysed by capillary GC-mass spectrometry (Hewlett Packard 5970 mass selective detector) to provide further information on the identity of the constituents present. The GC [12 m × 0.02 mm HP (methyl polysiloxane) capillary column] using helium as a carrier gas was programmed from 40 to 300°C at 5°C $min^{-1}$ and then held at 300°C for 20 min.

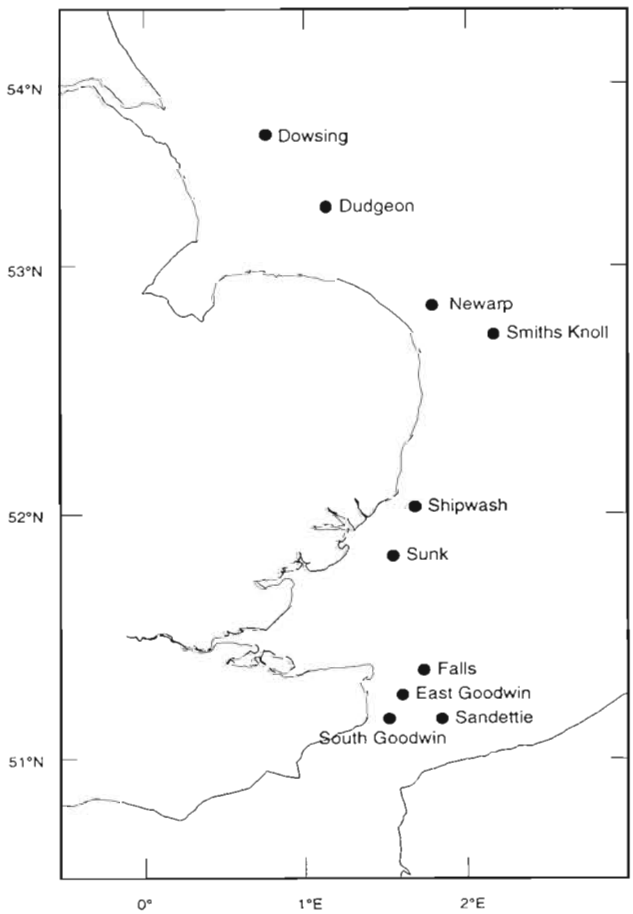


Fig. 2. Locations of offshore sampling sites (i.e. North Sea light vessels)

## RESULTS

### Preliminary studies to establish procedures and experimental design

In order to conduct a combined chemical and biological effects programme based on mussels sampled over

a large spatial scale of approximately 1000 km, it was first necessary to establish the most appropriate and effective procedures.

### Recovery from transportation and comparison between laboratory and field responses of mussels

Fig. 3 shows the time-course of recovery for clearance rate after a period of 24 h air exposure/transportation. The clearance rate (mean of 16 mussels) rapidly increased over a period of 12 h, and this was followed by the maintenance of a relatively steady rate from 12 to 48 h. The clearance rates of mussels from the Exmouth and Tamar populations were significantly lower ($p < 0.001$) than those from Gluss Voe and Whitsand. All components of the energy budget (clearance rate, food absorption efficiency, respiration rate and SFG) were measured after 24 h of recovery and are presented in Fig. 4 (note that the total bar length represents the energy *consumed* as food and the SFG is the difference between the energy *absorbed* from the food and the energy *respired*). In addition, the laboratory measurement of SFG (after 24 h air exposure and 24 h recovery at PML) is compared with the field measurement of SFG using a mobile laboratory at the Gluss Voe site in Shetland.

This comparison between field and laboratory measurements and among populations demonstrates:

(1) There were no significant differences between the field and laboratory measurement of Gluss Voe mussels. These results therefore showed that mussels are able to fully recover within 24 h, following a 24 h period of air exposure at low temperature, and the physiological responses of mussels measured at Shetland (in North Atlantic water) were not significantly different from the responses measured at PML (in Eddystone water). Such findings confirmed previous comparisons of field-laboratory measurements following 3 h of air exposure (Widdows 1985).

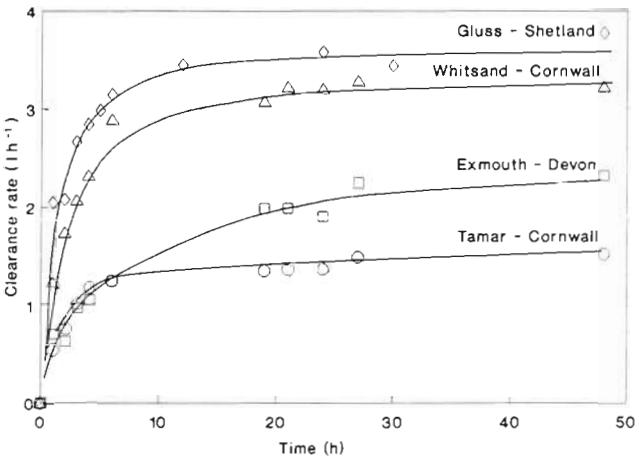


Fig. 3. *Mytilus edulis*. Time-course of recovery of clearance rate of mussels following 24 h of air exposure at 7°C (mean of 16 mussels). Sites: Gluss Voe, Shetland (◇); Whitsand, Cornwall (△); Exmouth, Devon (□); Tamar, Cornwall (○)

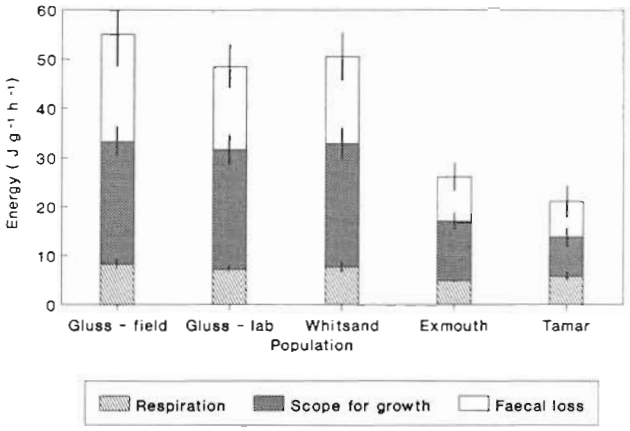


Fig. 4. *Mytilus edulis*. Components of the energy budget of mussels after 24 h of recovery from air exposure. Total bar height = energy ingested, which is partitioned into respiratory energy loss, the net energy gain termed scope for growth and faecal energy loss; mean ± 95% CI, n = 16

(2) Population/site differences were maintained throughout the recovery (0 to 12 h) and the steady state (12 to 48 h) phases. The clearance rates of mussels from the Exmouth and Tamar sites were significantly reduced relative to mussels from Gluss Voe and Whitsand (Fig. 3). Fig. 4 illustrates the various components of the energy budget and demonstrates that the SFG of mussels from Exmouth and Tamar were significantly reduced as a result of the lower clearance rates.

The degree of hydrocarbon contamination in mussels was significantly different at the 4 sites and this difference was maintained during the 48 h in the laboratory (Table 1). There was a significant negative correlation between SFG and log aromatic hydrocarbons ($r = -0.91$). The recorded concentration of hydrocarbons in the Tamar mussels (159 μg $g^{-1}$ dry wt) was considerably higher than any previous measurements at this site and probably reflects a significant oil input/spillage in the Tamar and the low flushing of the estuary during the exceptionally dry summer of 1989. As a result, the SFG (Fig 4) was considerably lower than previous measurements (e.g Fig. 5).

Comparison between mussel populations therefore confirmed that after 24 h of recovery mussels still retained the pollution induced stress effects, when measured under 'standard laboratory conditions'.

Table 1 *Mytilus edulis*. Hydrocarbon concentration (µg $g^{-1}$ dry wt) in the tissues of mussels during reimmersion and recovery from 24 h of air exposure (July 1989). Mean ± semi-range (based on 2 pools of 5 individuals)

| Site | Time (hours following reimmersion) 0 | 24 | 48 |
|---|---|---|---|
| Gluss Voe (Shetland) | 4.10 ± 0.14 | – | – |
| Whitsand (Cornwall) | 9.42 ± 0.28 | – | 11.42 ± 0.86 |
| Exmouth (Devon) | 20.71 ± 0.11 | 21.24 ± 1.32 | 18.24 ± 1.36 |
| Tamar (Cornwall) | 159.58 ± 14.28 | 140.30 ± 14.28 | 114.81 ± 11.07 |

### Environmental versus genetic differences

Mussels transplanted from Gluss Voe (Shetlands) to the Tamar estuary (Cornwall) were held in cages for 12 mo prior to measurement of the Gluss Voe transplants and the Tamar natives. The individual physiological responses and the resultant SFG were not significantly different (Fig. 5) thus confirming that the performance of mussels originating from the 2 extremes of the spatial/latitudinal gradient were not significantly different when the mussels were living in the same environment. Consequently, any recorded differences among mussel populations reflect environmental differences rather than inherited differences. These findings confirmed previous transplantation experiments which have shown that physiological responses and growth of mussels reflect environmental rather than genetic differences (e.g. Tamar-Swansea, Widdows et al. 1984; Baltic-North Sea, Kautsky et al. 1990; Scottish lochs, Okumus & Stirling 1994). This does not imply that there are no genetic differences between these populations, simply that genotypic differences in physiological responses are usually only apparent towards the upper and lower limits of environmental factors, such as temperature and salinity (Bayne et al. 1977, Kautsky et al. 1990).

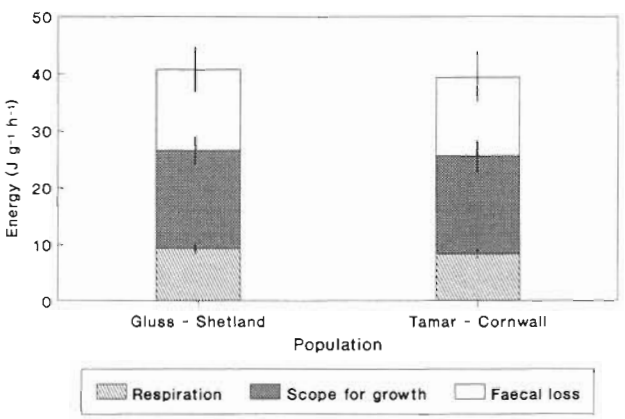


Fig. 5. *Mytilus edulis*. Components of the energy budget of mussels from Gluss Voe (Shetland) and Tamar (Cornwall) following transplantation and 12 mo acclimation to the Tamar. Total bar height = energy ingested, which is partitioned into respiratory energy loss, scope for growth and faecal energy loss; mean ± 95% CI, n = 16

### Scope for growth of mussels at coastal sites (Phase I)

The major physiological responses which form components of the energy budget, including the integrated response SFG of mussels from sites along the UK North Sea coastline, are presented in Table 2 and illustrated in Fig. 6. There were significant differences among sites recorded in the clearance rates, food absorption efficiency, respiration rate and SFG of mussels. Clearance rate was significantly reduced to <4 l $g^{-1}$ $h^{-1}$ at the following sites: Ythan, Cresswell, Teesmouth, Filey Brigg, and at sites in the Humber-Wash region (Cleethorpes, Gat Sand, Hunstanton) and in the Thames estuary (Creeksea, Southend, Swale). Food absorption efficiency was significantly lower than the overall mean of 0.42 at Ythan, Montrose, Blyth, Humber Bull Fort, and sites between Crabknowe Spit and Whitstable. Mussels from Gat Sand and Hunstanton sites in the Wash had higher absorption efficiencies compared with the other southern sites and this partially compensated for the reduced feeding rates. Respiration rates of mussels from Orkney and Creeksea sites were significantly lower than the overall mean (19.7 µmol $O_2$ $g^{-1}$ $h^{-1}$) and Humber Bull Fort was significantly higher.

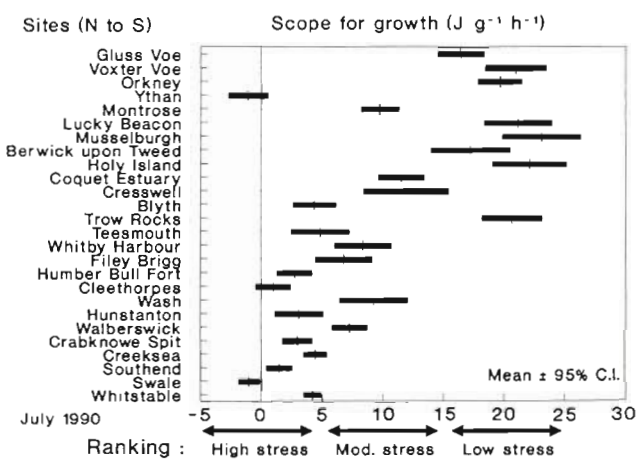


Fig. 6. *Mytilus edulis*. Scope for growth of mussels collected from sites along the UK North Sea coastline (mean ± 95% CI, n = 16)

Table 2. *Mytilus edulis*. Physiological responses of North Sea mussels (mean ± 95% CI). Energy conversion factors used in the calculation of scope for growth: SFG = [(clearance rate × energy equivalent of algal food concentration, 9.2 J $l^{-1}$) × absorption efficiency] – (respiration rate × 0.456 J $\mu mol^{-1}$ $O_2$)

| Sites (north → south) | Clearance rate ($l\ g^{-1}\ h^{-1}$) | Absorption efficiency | Respiration rate ($\mu mol\ O_2\ g^{-1}\ h^{-1}$) | SFG ($J\ g^{-1}\ h^{-1}$) | SFG (temperature corrected) |
|---|---|---|---|---|---|
| **Mussels from coastal sites** | | | | | |
| Gluss Voe | 6.17 ± 0.54 | 0.46 | 20.6 ± 2.3 | 16.41 ± 1.90 | |
| Voxter Voe | 6.74 ± 0.54 | 0.51 | 23.3 ± 2.2 | 20.94 ± 2.52 | |
| Orkney | 7.65 ± 0.56 | 0.38 | 15.9 ± 1.1 | 19.63 ± 1.82 | |
| Ythan Estuary | 3.00 ± 0.47 | 0.33 | 18.8 ± 2.0 | −1.08 ± 1.64 | |
| Montrose | 5.04 ± 0.44 | 0.33 | 18.9 ± 1.0 | 9.75 ± 1.57 | |
| Lucky Beacon | 8.19 ± 0.82 | 0.43 | 18.1 ± 1.4 | 21.11 ± 2.78 | |
| Musselburgh | 7.79 ± 0.72 | 0.46 | 21.3 ± 2.0 | 23.07 ± 3.22 | |
| Berwick upon Tweed | 6.75 ± 0.90 | 0.44 | 21.6 ± 1.8 | 17.19 ± 3.26 | |
| Holy Island | 6.95 ± 0.74 | 0.51 | 22.9 ± 1.8 | 22.08 ± 3.06 | |
| Coquet Estuary | 5.17 ± 0.51 | 0.43 | 19.5 ± 1.3 | 11.49 ± 1.89 | |
| Cresswell | 4.07 ± 0.74 | 0.57 | 21.2 ± 1.8 | 11.89 ± 3.48 | |
| Blyth | 5.03 ± 0.69 | 0.30 | 20.4 ± 1.7 | 4.35 ± 1.75 | |
| Trow Rocks | 5.68 ± 0.49 | 0.60 | 23.3 ± 2.5 | 20.62 ± 2.47 | |
| Teesmouth | 3.68 ± 0.62 | 0.45 | 22.8 ± 2.0 | 4.82 ± 2.41 | |
| Whitby Harbour | 4.60 ± 0.57 | 0.42 | 20.3 ± 2.0 | 8.33 ± 2.34 | |
| Filey Brigg | 3.42 ± 0.56 | 0.47 | 17.4 ± 1.2 | 6.77 ± 2.32 | |
| Humber Bull Fort | 5.41 ± 0.55 | 0.30 | 27.2 ± 2.0 | 2.73 ± 1.46 | |
| Cleethorpes | 2.60 ± 0.46 | 0.43 | 20.3 ± 1.4 | 0.98 ± 1.45 | 1.44 ± 1.74 |
| Gat Sand – Wash | 3.36 ± 0.46 | 0.59 | 20.3 ± 2.6 | 9.23 ± 2.81 | 11.63 ± 3.33 |
| Hunstanton | 2.15 ± 0.44 | 0.56 | 17.8 ± 1.6 | 3.05 ± 1.99 | 4.10 ± 2.42 |
| Walberswick | 4.59 ± 0.48 | 0.35 | 16.6 ± 1.5 | 7.22 ± 1.44 | 8.96 ± 1.72 |
| Harwich | 4.49 ± 0.68 | 0.25 | 16.5 ± 1.8 | 2.95 ± 1.21 | 3.95 ± 1.46 |
| Creeksea | 3.84 ± 0.36 | 0.32 | 15.1 ± 1.2 | 4.41 ± 0.95 | 5.68 ± 1.14 |
| Southend | 3.07 ± 0.42 | 0.31 | 15.8 ± 1.5 | 1.49 ± 1.07 | 2.16 ± 1.28 |
| Swale | 3.09 ± 0.45 | 0.25 | 17.9 ± 2.1 | −1.02 ± 0.88 | −0.82 ± 1.06 |
| Whitstable | 4.38 ± 0.41 | 0.32 | 19.0 ± 1.6 | 4.20 ± 0.73 | 5.51 ± 0.89 |
| **Mussel transplants to light vessels** | | | | | |
| Dowsing | 3.30 ± 0.31 | 0.60 | 8.71 ± 1.2 | 14.08 ± 1.84 | |
| Dudgeon | 3.36 ± 0.40 | 0.62 | 10.0 ± 1.2 | 14.91 ± 2.28 | |
| Newarp | 3.28 ± 0.38 | 0.54 | 8.9 ± 0.6 | 12.22 ± 1.87 | |
| Smiths Knoll | 3.08 ± 0.34 | 0.53 | 10.2 ± 1.2 | 10.29 ± 1.65 | |
| Shipwash | nd | nd | nd | nd | |
| Sunk | 3.89 ± 0.46 | 0.50 | 10.6 ± 1.2 | 13.15 ± 1.30 | |
| Falls | 3.14 ± 0.34 | 0.46 | 9.8 ± 0.6 | 8.87 ± 1.28 | |
| East Goodwin | 3.65 ± 0.52 | 0.54 | 10.6 ± 1.2 | 13.11 ± 2.46 | |
| Sandettie | 2.82 ± 0.42 | 0.63 | 9.3 ± 0.6 | 12.22 ± 2.36 | |
| South Goodwin | 3.50 ± 0.28 | 0.57 | 10.7 ± 0.9 | 13.61 ± 1.37 | |
| Exmouth – source | 3.00 ± 0.32 | 0.51 | 11.7 ± 0.98 | 8.55 ± 1.66 | |

The SFG provides an integration of these physiological energetic responses and an overall assessment of the performance of the mussels from north to south (Fig. 6). There is a general trend of high SFG values in the north (>15 J $g^{-1}$ $h^{-1}$) and lower SFG in the south, with particularly low SFG values (i.e. <5 J $g^{-1}$ $h^{-1}$) at specific sites (e.g. Ythan, Blyth, Teesmouth, Humber Bull Fort, Cleethorpes, Hunstanton, Crabknowe Spit, Creeksea, Southend, Swale and Whitstable). With the exception of the Ythan site, mussels in the vicinity of estuaries with major urban and industrial developments had the lowest SFG values. Furthermore, there were significant trends in SFG at coastal sites in (1) the Humber-Wash region (declining south of Filey Brigg and increasing again in the outer Wash and at Walberswick on the East Anglian coast), and (2) the Thames estuary (declining from Creeksea to Swale, the innermost site, and then increasing at Whitstable).

## Additional measurements

The quality of the EFSW at the PML during the 3 wk period of measurements was checked at weekly intervals by determining the clearance rate of mussels collected from Gluss Voe and Voxter Voe. These mea-

Table 3. *Mytilus edulis*. Weekly measurement of clearance rates of Shetland (Gluss Voe and Voxter Voe) mussels to monitor the consistent high water quality (EFSW) at the PML (mean ± SE, n = 16)

| Date (1990) | Clearance rate ($l\ g^{-1}\ h^{-1}$) |
|---|---|
| 30 June | 6.56 ± 0.27 |
| 7 July | 6.71 ± 0.36 |
| 15 July | 6.67 ± 0.43 |
| 20 July | 6.19 ± 0.36 |

surements showed that the rates did not alter significantly during this period (Table 3), thus confirming that the high water quality was maintained throughout the field sampling programme.

Mussels from Whitsand (Cornwall) measured at the beginning and end of the field sampling period showed a small, but significant ($p < 0.05$), decline in SFG of 4.3 J $g^{-1}$ $h^{-1}$ (Table 4). This decline resulted from a reduction in both clearance rate (22%) and respiration rate (18%), but there was no change in the absorption efficiency. Generally, components of the energy budget and SFG of mussels *Mytilus edulis* are maintained temperature independent as a result of thermal acclimation over a wide range of temperatures (6 to 20°C; Widdows 1973, 1978), including fluctuating temperatures (Widdows 1976). However, above 20°C there is gradual decline in the SFG of *M. edulis* with increasing temperature (Widdows 1973, 1978). The observed reduction in SFG of the Whitsand mussels coincided with a sudden increase in air and shallow inshore seawater temperatures (from 14.5 to 21.5°C) in mid-July, caused by the onset of a 'high pressure' weather system which then dominated the unusually warm summer of 1990.

The sudden increase in environmental temperatures during the North Sea sampling programme influenced sites south of Cleethorpes, which could account for part of the observed reductions in clearance rate, respiration rate and SFG of these mussels. This was confirmed by the slight but significant negative correlation ($r = -0.5$, $p < 0.05$) between clearance rate, respiration, SFG and temperature for all sites ($n = 26$), but no significant correlation with temperature was apparent when sites were divided into 2 groups (northern: Gluss to Humber;

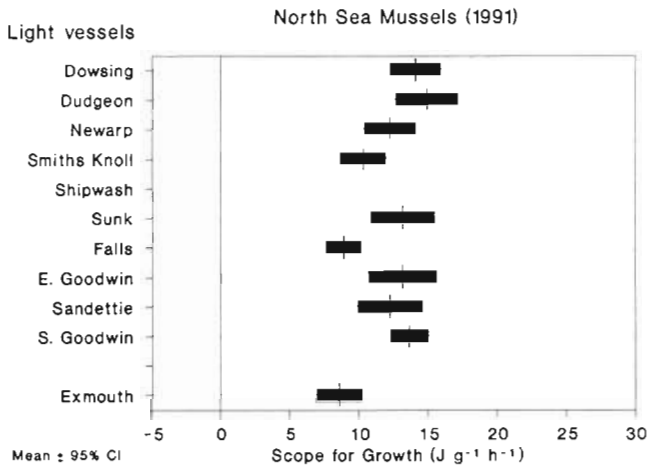


Fig. 7. *Mytilus edulis*. Scope for growth of mussels after 6 wk transplantation to North Sea light vessels (mean ± 95% CI, n = 16)

southern: Cleethorpes to Whitstable). Consequently, after applying a correction for the temperature-induced stress effects recorded in the Whitsand mussels (i.e. clearance rate = 22%, respiration = 18%) to mussels from sites south of Cleethorpes (Table 2), there was no significant correlation with temperature from north to south. Subsequent measurements of mussels from the Wash sites (Gat Sand and Hunstanton) and Holy Island in July 1994 (Widdows unpubl. data) not only confirmed the low SFG values in mussels from the Wash, but also demonstrated that the temperature corrected SFG values for 1990 were not significantly different from those recorded values for 1994.

## Scope for growth of mussels at offshore light vessels (Phase II)

Caged mussels were successfully recovered from all light vessels after 6 wk deployment and exposure to conditions in the North Sea. Mortalities at all sites were <5%. The major physiological responses which form components of the energy budget are given in Table 2 and the integrated response of SFG summarized in Fig. 7. The physiological responses of mussels from 'Shipwash', the last site to be measured, were lost

Table 4. *Mytilus edulis*. Comparison between physiological responses of Whitsand mussels at the beginning and end of the North Sea field study (mean ± SE, n = 16)

| Date (1990) | Temperature (°C) | Clearance rate ($l\ g^{-1}\ h^{-1}$) | Absorption efficiency | Respiration rate ($\mu mol\ O_2\ g^{-1}\ h^{-1}$) | SFG ($J\ g^{-1}\ h^{-1}$) |
|---|---|---|---|---|---|
| 26 Jun | 14 | 6.07 ± 0.40 | 0.48 ± 0.08 | 22.6 ± 1.4 | 16.4 ± 1.6 |
| 27 Jul | 21 | 4.70 ± 0.42 | 0.47 ± 0.08 | 18.4 ± 1.3 | 12.1 ± 0.8 |

due to the failure of the seawater temperature control system at PML.

The clearance rates, absorption efficiencies and respiration rates of offshore mussels showed no marked changes or trends north to south, although there were a few sites that were significantly different ($p < 0.05$) from the majority of sites. For example, mussels from Sunk had higher clearance rates than those from Sandettie and the Exmouth (source), while mussels from Dowsing, Falls and Sandettie had respiration rates that were significantly lower than those from the remaining sites. Scope for growth, however, provides a more integrated assessment of the mussels' performance and shows a slight but significant decline from north to south in the northern group of light vessels (Dowsing to Smiths Knoll along the Humber plume), and a significant decline at the most northern light vessel in the southern group (Falls at the mouth of the Thames estuary).

### Chemical contaminants in mussels

The concentration of chemical contaminants in the tissues of mussels (µg $g^{-1}$ dry wt) from the 26 North Sea coastal sites and the 9 light vessel sites are presented in Table 5. Generally there was little variation in the metal concentrations along the UK coast. Those metals and sites which were significantly elevated (i.e. 5 to 10× above the lowest 'background' concentration) include: Cd at Musselburgh (10×) and Pb at Trow rocks (13×). Tributyltin (TBT) concentrations at most sites were significantly higher than at Gluss Voe (i.e. >10×), with the highest concentrations in the Coquet estuary (73×), Whitby Harbour (34×), and Blyth, Trow rocks, Teesmouth, Humber and Thames estuaries (13 to 28×). Even the 5 southern offshore sites (i.e. light vessels in the English Channel and mouth of the Thames estuary) had TBT levels that were 7× higher than at Gluss Voe. Organochlorines were not detectable in a large proportion of the sites, but dieldrin, ppTDE and HCH were all 10× the detection limit at Cleethorpes. The highest PCB concentrations were at Ythan (136× detection limit) and Swale (93×), with Lucky Beacon, Musselburgh, Blyth, Humber, Creeksea and Southend between 20 and 35× the detection limit. Organochlorine concentrations in offshore mussels were below the detection limit at all sites except Sunk where mussels accumulated significant levels of ppDDE and PCBs.

The measurement of total hydrocarbons in mussels by UV fluoresecence detects aromatic hydrocarbons but it is calibrated against weighed solutions of complete crude oil. Thus, all components of the oil, including those which are nontoxic, are effectively used to determine the value given in Table 5. These values are therefore higher than the 2- and 3-ring polyaromatic hydrocarbon (PAH) concentrations, which more closely reflect the toxic component of bioaccumulated petroleum hydrocarbons. PAH concentrations ranged over 2 orders of magnitude with the highest concentrations at Montrose, Blyth, Whitby and Swale (>100× background). Coastal sites not in the immediate vicinity of urban and industrial developments and also the 5 most southerly light vessels in the English Channel and southern North Sea had 2- and 3-ring PAH concentrations of between 1 and 2.5 µg $g^{-1}$ (4 to 10× background levels recorded in the Shetlands).

HPLC analysis of polar compounds failed to provide evidence for the occurrence of aromatic hydrocarbon oxidation products of the type described by Burns (1993). This may have been partly due to the relatively low sensitivity of the analytical procedures available to us. However, HPLC chromatograms of mussel distillates from all sites contained several notable peaks. All samples gave rise to a distinctive doublet of peaks, which varied substantially in size between sites; these peaks were quantified and the data given in Table 5. GC-MS analysis of the appropriate HPLC fraction indicated the presence of a series of oxygenated compounds including ketones, alcohols, aldehydes and phenols with between 7 and 18 carbon atoms. Identifications were based on a mass-spectral library search and are therefore preliminary, but the quality of the spectral match was high for several compounds. The presence of bis(2-ethylhexyl) phthalate was also clearly demonstrated, but since no special precautions had been taken to avoid possible phthalate contamination during handling of the mussels, this result must be treated with caution.

## DISCUSSION

Analysis and interpretation of the combined measurement of SFG and chemical contaminants in the tissues of mussels can be considered as a 2 stage process: (1) the detection and quantification of environmental quality, and (2) the toxicological interpretation and identification of the causes of the observed deleterious effects.

Clearly, it is necessary to extend the analysis of the results beyond statistical correlations, which will inevitably be poor without a high correlation between any single contaminant and SFG over such a large spatial scale with complex and varying contaminant mixtures. This is in contrast to previous studies, involving small-scale pollution gradients in bays and estuaries, where there have been high correlations between any single contaminant and SFG (typical correlation coefficients are $r = -0.9$; Widdows et al. 1990) due to the

Table 5. *Mytilus edulis*. Concentration (µg $g^{-1}$ dry wt) of chemical contaminants in body tissues of North Sea mussels. Mean values of duplicate samples. nd: not determined; wet to dry weight conversion factors: *PML data × 5, MAFF data × 5.86; conversion of 2- and 3-ring PAH to total toxic hydrocarbons: ×7.1; ppDDT [or 1,1,1-trichloro-2,2'-bis(*p*-chlorophenyl)-ethane] and αHCH (or hexachlorocyclohexane): all samples were below the detection limit of 0.006 µg $g^{-1}$ dry wt. DBT: dibutyltin; TBT: tributyltin; ppDDE: 1,1-dichloro-2,2'-bis(*p*-chlorophenyl)ethylene; ppTDE: 1,1-dichloro-2,2'-bis(*p*-chlorophenyl)ethane; PCB: polychlorinated biphenyls; THC by UVF: total hydrocarbons by UV fluorescence (Ekofisk crude oil); PAH: polyaromatic hydrocarbons

| Mussel sites (north → south) | Metals | | | | | Organometals | | Organics | | | | | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | Cd | Cu | Hg | Pb | Zn | DBT | TBT | Dieldrin | ppDDE | ppTDE | HCB | γHCH | PCB | THC by UVF | 2- + 3-ring *PAH | *Total toxic HC | *Polar |
| **Mussels from coastal sites** | | | | | | | | | | | | | | | | | |
| Gluss Voe | 1.17 | 7.02 | 0.12 | 3.63 | 173 | 0.012 | 0.018 | <0.006 | 0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 28 | 0.23 | 1.6 | 49.5 |
| Voxter Voe | 0.85 | 6.73 | 0.12 | <3.00 | 208 | 0.041 | 0.117 | <0.006 | 0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 36 | 0.46 | 3.3 | 12.5 |
| Orkney | 1.00 | 5.27 | 0.21 | 9.36 | 94 | 0.035 | 0.059 | <0.006 | 0.018 | <0.006 | <0.006 | <0.006 | <0.006 | 45 | 1.73 | 12.4 | 2.5 |
| Ythan | 0.49 | 7.61 | 0.15 | 3.80 | 72 | <0.018 | 0.059 | <0.006 | 0.006 | <0.006 | <0.006 | <0.006 | 0.820 | 70 | 2.96 | 21.1 | 20 |
| Montrose | 0.50 | 7.02 | 0.18 | 4.36 | 87 | 0.041 | 0.293 | <0.006 | 0.006 | 0.006 | <0.006 | <0.006 | 0.064 | 193 | 22.70 | 162.1 | 125 |
| Lucky Beacon | 0.82 | 5.27 | 0.18 | nd | 58 | nd | nd | <0.006 | 0.018 | 0.012 | <0.006 | <0.006 | 0.141 | 117 | 3.55 | 25.4 | 2.5 |
| Musselburgh | 5.18 | 8.48 | 0.21 | 18.72 | 130 | nd | nd | <0.006 | 0.006 | 0.006 | <0.006 | <0.006 | 0.176 | 199 | 7.84 | 56.0 | 21.5 |
| Berwick upon Tweed | 0.93 | 7.42 | 0.29 | <3.00 | 100 | 0.053 | 0.105 | 0.012 | 0.012 | <0.006 | <0.006 | <0.006 | <0.006 | 44 | 2.17 | 15.5 | 14 |
| Holy Island | 0.89 | 8.88 | 0.23 | <3.00 | 92 | 0.064 | 0.108 | 0.006 | nd | <0.006 | <0.006 | <0.006 | <0.006 | 46 | 1.21 | 8.6 | 13 |
| Coquet Estuary | 0.62 | 7.06 | 0.18 | 4.70 | 118 | 0.257 | 1.316 | 0.006 | 0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 258 | 18.60 | 132.8 | 11.5 |
| Cresswell | 1.50 | 5.63 | 0.38 | <3.00 | 125 | 0.023 | 0.029 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 0.029 | 158 | 1.13 | 8.1 | 100 |
| Blyth | 0.88 | 7.81 | 0.25 | 11.88 | 150 | 0.140 | 0.462 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 0.188 | 170 | 52.18 | 372.6 | 16 |
| Trow Rocks | 0.66 | 8.75 | 0.34 | 28.80 | 303 | 0.158 | 0.509 | 0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 0.023 | 158 | 4.45 | 31.8 | 2.5 |
| Teesmouth | 1.30 | 9.71 | 0.06 | 11.51 | 173 | 0.088 | 0.421 | 0.006 | 0.018 | <0.006 | <0.006 | <0.006 | 0.035 | 334 | 10.88 | 77.7 | 3 |
| Whitby Harbour | 1.13 | 10.31 | 0.25 | 13.75 | 169 | 0.120 | 0.615 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 434 | 54.76 | 390.0 | 37 |
| Filey Brigg | 0.75 | 7.50 | 0.30 | 10.00 | 95 | 0.029 | 0.047 | 0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 129 | 3.30 | 23.6 | 37 |
| Humber Bull Fort | 3.28 | 10.00 | 0.17 | <3.00 | 184 | 0.123 | 0.360 | 0.023 | 0.012 | 0.023 | <0.006 | <0.006 | 0.123 | 123 | 6.55 | 46.8 | 43 |
| Cleethorpes | 2.53 | 7.96 | 0.16 | <3.00 | 116 | 0.099 | 0.263 | 0.053 | 0.012 | 0.064 | <0.006 | 0.070 | 0.053 | 193 | 17.49 | 124.9 | 43 |
| Gat Sand – Wash | 1.06 | 6.87 | 0.11 | 4.01 | 98 | 0.047 | 0.029 | 0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 49 | 1.25 | 8.9 | 75.5 |
| Hunstanton | <0.32 | 7.03 | 0.11 | <3.00 | 73 | 0.041 | 0.035 | 0.018 | 0.012 | 0.018 | <0.006 | <0.006 | <0.006 | 47 | 1.18 | 8.4 | 20.5 |
| Walberswick | 0.86 | 9.28 | 0.36 | 7.50 | 128 | 0.053 | 0.187 | 0.012 | 0.012 | 0.012 | <0.006 | <0.006 | <0.006 | 82 | 2.46 | 17.6 | 46 |
| Harwich | 1.14 | 6.39 | 0.25 | <3.00 | 111 | 0.111 | 0.176 | 0.012 | 0.012 | 0.012 | <0.006 | <0.006 | 0.053 | 82 | 2.36 | 16.9 | 20 |
| Creeksea | 0.95 | 7.51 | 0.22 | <3.00 | 102 | 0.234 | 0.427 | 0.023 | 0.023 | 0.018 | <0.006 | 0.006 | 0.217 | 123 | 10.74 | 76.7 | 22 |
| Southend | 1.50 | 8.42 | 0.11 | 3.42 | 118 | 0.205 | 0.357 | 0.018 | 0.009 | <0.006 | <0.006 | <0.006 | 0.217 | 105 | 2.83 | 20.2 | 7.5 |
| Swale | 1.25 | 8.06 | 0.11 | 3.34 | 125 | 0.234 | 0.486 | 0.035 | 0.020 | 0.023 | <0.006 | 0.006 | 0.557 | 422 | 26.73 | 190.9 | 11 |
| Whitstable | 2.29 | 8.23 | 0.24 | <3.00 | 109 | 0.088 | 0.082 | 0.012 | 0.012 | <0.006 | <0.006 | <0.006 | 0.182 | 82 | 2.85 | 20.4 | 18.5 |
| **Mussel transplants to light vessels** | | | | | | | | | | | | | | | | | |
| Dowsing | 0.95 | 7.14 | <0.06 | 3.00 | 64 | 0.053 | 0.059 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 53 | 1.00 | 7.1 | nd |
| Dudgeon | 1.26 | 7.14 | <0.06 | 3.08 | 69 | 0.053 | 0.059 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 40 | 0.68 | 4.9 | nd |
| Newarp | 1.05 | 7.25 | <0.06 | 3.00 | 63 | 0.047 | 0.053 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 43 | 0.99 | 7.1 | nd |
| Smiths Knoll | 0.92 | 6.12 | 0.08 | <3.00 | 78 | 0.041 | 0.047 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 53 | 0 84 | 6.0 | nd |
| Shipwash | 1.25 | 7.25 | <0.06 | 3.00 | 73 | 0.059 | 0.059 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 36 | 0.98 | 7.0 | nd |
| Sunk | 0.88 | 6.67 | 0.06 | <3.00 | 76 | 0.117 | 0.117 | <0.006 | 0.038 | <0.006 | <0.006 | <0.006 | 0.041 | 59 | 1.18 | 8.4 | nd |
| Falls | 0.95 | 7.02 | 0.05 | <3.00 | 92 | 0.059 | 0.117 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 59 | 1.61 | 11.5 | nd |
| East Goodwin | 0.72 | 6.67 | 0.11 | 3.61 | 72 | 0.059 | 0.117 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 0.059 | 56 | 1.28 | 9.1 | nd |
| Sandettie | 0.35 | 5.75 | 0.05 | <3.00 | 50 | 0.047 | 0.117 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 43 | 1.15 | 8.2 | nd |
| South Goodwin | 0.67 | 5.64 | 0.13 | <3.00 | 62 | 0.117 | 0.117 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 0.129 | 54 | 1.02 | 7.3 | nd |
| Exmouth (source) | 0.32 | 4.68 | 0.04 | <3.00 | 40 | 0.117 | 0.117 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | <0.006 | 53 | 2.20 | 15.7 | nd |

processes of dilution, dispersion and degradation along the axis length. Over a large spatial scale (e.g. 1000 km), contaminants will not necessarily covary or correlate with a single 'marker' of overall contamination. Hence the need to extend interpretation of the data beyond statistical correlations and provide a quantitative toxicological interpretation of the biological and chemical data.

A quantitative toxicological interpretation of the combined tissue residue chemistry and SFG measurements was possible using 'cause-effect' relationships, including QSARs (Quantitative Structure-Activity Relationships), as a basis for assessing the toxic effects. Experimentally derived 'cause-effect' relationships between the concentration of contaminants in mussel tissues and the SFG response thus provide a means of identifying and partitioning the cause(s) of observed effects. Consequently, the additive toxicological contribution of different groups of contaminants towards the decline in SFG can be assessed. Research has confirmed that both structurally related compounds, forming a single QSAR line, are simply additive, and that toxic effects of the majority of structurally unrelated toxicants also tend to be additive (i.e. on the basis of proportional additivity; Widdows & Donkin 1991).

While it is feasible to establish a database of concentration-response relationships for individual contaminants such as metals and organometals, clearly it is unrealistic to examine the sublethal effects of >100000 individual organic contaminants. A QSAR approach provides a means of overcoming this problem and facilitates the prediction of toxicological properties of organic compounds from their structure and physicochemical properties (Donkin & Widdows 1990).

## Quantitative toxicological interpretation of North Sea mussel data

### Hydrocarbons

A large contribution towards the observed decline in SFG of mussels along the North Sea coast (see Fig. 8) is caused by toxic hydrocarbons acting through the mechanism of 'nonspecific narcosis' resulting in the inhibition of clearance rate. The concentration-response relationship describing the effect of log concentration of toxic hydrocarbons in the tissues on the response SFG is illustrated in Fig. 9. Identification of the range of aromatic and aliphatic hydrocarbons exerting toxic effects has been established in QSAR studies (Donkin et al. 1989, 1991, Widdows & Donkin 1992). This total toxic hydrocarbon concentration is primarily aromatic hydrocarbons with log octanol-water partition coefficient (log $K_{ow}$) values < 5.5 (e.g. molecular weight cut-off at fluoranthene), but aliphatics of $<C_{11}$ can also contribute to the 'toxic load'. This hydrocarbon-SFG relationship (Fig 9) has been found to be very consistent in laboratory, mesocosm and field studies (Widdows & Donkin 1992).

Consequently, we have used the measurement of 2- and 3-ring PAHs (by HPLC) converted to a total toxic hydrocarbon value (by GC) using a factor of ×7.1 (Widdows et al. 1982, 1995). The measurement of total hydrocarbon concentration (by UVF) in North Sea mussels includes both toxic and nontoxic hydrocarbons and is therefore approximately 5-fold higher than the toxic hydrocarbon concentration (Table 5). However, there is a significant correlation (r = 0.87) between the 2 measurements of hydrocarbons.

The extent to which SFG is reduced by toxic hydrocarbons can be estimated from the relationship in Fig. 9. Their contribution to the observed reduction in SFG can then be plotted as in Fig. 8. It is apparent that at the majority of North Sea coastal sites toxic hydrocarbons are at concen-

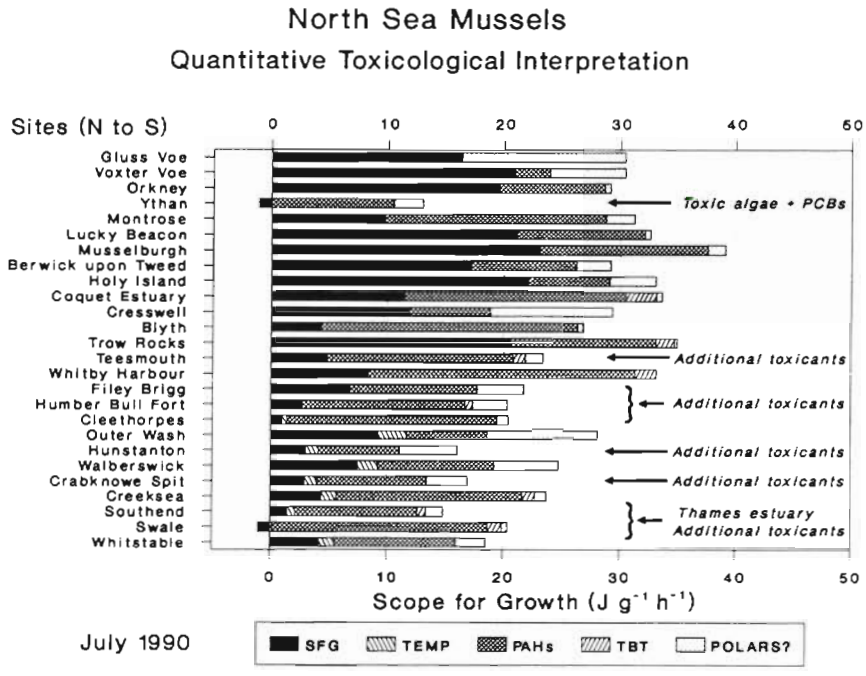


Fig. 8. *Mytilus edulis*. Quantitative toxicological interpretation and partitioning of the reduced scope for growth of mussels from the North Sea coastal sites

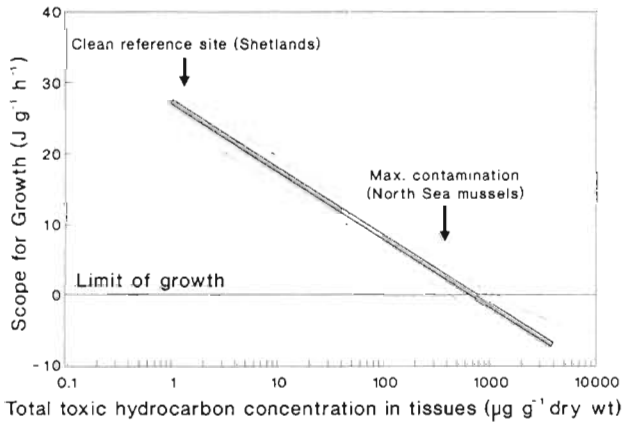


Fig. 9. *Mytilus edulis*. Relationship between scope for growth and the log concentration of total toxic hydrocarbons in the tissue of mussels (mean ± 95% CI) (from Widdows et al. 1992, 1994)

trations capable of inducing a significant inhibition of SFG and forming a major component of the overall reduction in SFG. Hydrocarbon concentrations were lowest at Gluss Voe and were particularly elevated (i.e. >100 µg $g^{-1}$ dry wt) at Montrose, Coquet estuary, Blyth, Whitby Harbour, Cleethorpes and Swale. Overall, the North Sea sites showed a significant negative correlation ($r = -0.41$, $p < 0.05$, $n = 26$) between SFG and log hydrocarbon concentration. At Musselburgh, the toxic effects predicted from the hydrocarbons data may be slightly overestimated, resulting in a higher than average 'contaminant corrected SFG' (i.e. >30 to 35 J $g^{-1}$ $h^{-1}$). This is probably the result of a significant contribution to the hydrocarbon body burden from coal particles. Mussels at this site are in close proximity to the Monkton coalfield and power station. Coal particles will be extracted and analysed by the chemical methodology, but they will be less bioavailable compared to petroleum hydrocarbons.

## Polar organics

The 'polar organics' represent a group of compounds that are the least well-defined, hence the most speculative toxic component in Fig. 8. Their identity, spatial distribution and abundance in the North Sea study suggest that they are largely of natural origin. Compounds likely to be of biogenic origin are often detected in extracts of mussels prepared for hydrocarbon contaminant analysis (Tibbetts et al. 1982, Burns et al. 1990, Hellou et al. 1993). They are generally considered to be derived from the algal diet. Algae can produce long chain ketones (Sicre et al. 1990), but the smaller molecules which we detected have not been reported in previous environmental studies. The justification for including these compounds in our toxicological assessment is, firstly, that their physicochemical properties indicate that they must act at least as non-polar or polar narcotics (Donkin & Widdows 1990), and, secondly, that they are accumulated in significant amounts and are able to account for relatively small but otherwise unexplained reductions in SFG, particularly at relatively 'clean' sites which do not receive significant industrial inputs (e.g. Gluss Voe, Gat Sand and Walberswick; Fig. 8). This particularly applies to Gluss Voe, which has been studied annually (1982 to 1989; Widdows et al. 1995) and is regarded as a very 'clean' reference site with consistently high SFG values (>20 J $g^{-1}$ $h^{-1}$). During the 1990 North Sea study, however, the SFG at Gluss Voe (16.4 J $g^{-1}$ $h^{-1}$) was lower than the SFG of mussels from another 'clean' Shetland site at Voxter Voe (20.9 J $g^{-1}$ $h^{-1}$), and this reduction in SFG was accompanied by a higher concentration of polar organics. Gluss Voe is a relatively enclosed site experiencing relatively low water exchange, and mussels appear to accumulate natural products from macroalgae (ketones) and adjacent peat beds (lignin-like methoxy phenols; Reeves & Preston 1991) to concentrations that are potentially toxic to mussels. Recent experimental studies have also shown that some microalgae can produce exudates which can influence the feeding rate of mussels (Ward & Targett 1989). These results are clearly of a preliminary nature and further research is required on the analysis, source, occurrence and toxicity of these compounds. However, the results indicate that naturally occurring compounds, which are readily bioaccumulated, should be considered in any precise interpretation of field-derived toxicity measurements.

## Organotins

Tributyltin (TBT) and its breakdown product, dibutyltin (DBT), were accumulated by mussels at all sites. Highest concentrations of both TBT and DBT occurred in mussels at the mouth of the Coquet estuary (situated near a harbour/marina) and the lowest were at Gluss Voe, Shetlands. TBT's primary mechanism of toxicity in the mussel is the uncoupling of oxidative phosphorylation at concentrations above 0.2 µg TBT $g^{-1}$ dry wt. TBT's secondary mechanism of toxicity is via a neurotoxic effect on the gill's ciliary activity which causes a rapid decline in feeding rate and thus SFG at concentrations above 4 µg TBT $g^{-1}$ dry wt. (Widdows & Page 1993). None of the North Sea sites had TBT concentrations capable of inducing adverse effects on feeding rate. However, at several sites mussels had TBT concentrations above the threshold of effect on respiratory uncoupling (0.2 µg TBT $g^{-1}$ dry wt). The increase in energy expenditure associated

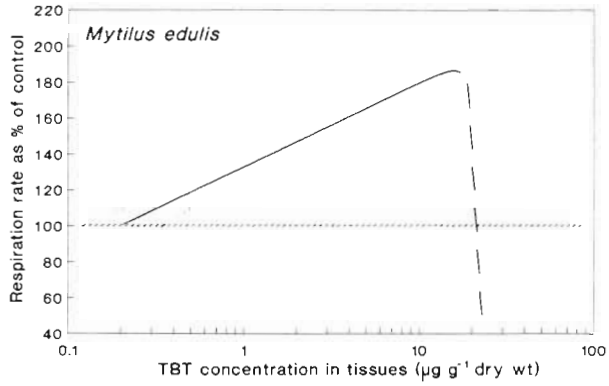


Fig. 10. *Mytilus edulis*. Relationship between increased respiration rate and the log concentration of tributyltin (TBT) in the tissues of mussels (mean ± 95% CI) (from Widdows & Page 1993)

with respiratory uncoupling between 0.2 and 10 µg TBT $g^{-1}$ dry wt is calculated from the relationship in Fig. 10 (from Widdows & Page 1993) and its contribution to the reduced SFG is illustrated in Fig. 8. In the present study the respiratory uncoupling effect of TBT did not manifest itself as a significant increase in respiration rate because the metabolic rate was already elevated (and near maximum scope for activity; Widdows 1973) as a result of the relatively high ration level and the associated costs of feeding and growth. Consequently, the uncoupling effect was masked at high ration levels (Widdows unpubl. obs.) and was only detectable in terms of a significant increase in respiration rate in field and laboratory studies when mussels are fed at low ration levels (Widdows et al. 1990, Widdows & Page 1993).

Clearly, even at the highest concentrations TBT only induces a minor inhibitory effect on growth. DBT is approximately an order of magnitude less toxic than TBT when expressed on a tissue concentration basis (Widdows & Page 1993). Consequently, the DBT concentrations recorded in North Sea mussels did not have any deleterious effects.

## Metals

At no sites were metals accumulated to concentrations that could cause a significant reduction in SFG. Recorded 'no observed effect thresholds' on feeding, growth or SFG of mussels are:

Cd: >150 µg Cd $g^{-1}$ dry wt (Poulson et al. 1982)

Cu: >25 µg Cu $g^{-1}$ dry wt (Calabrese et al. 1983, Widdows & Johnson 1988); lethal concentration of 60 µg Cu $g^{-1}$ dry wt (Martin 1979, Widdows & Johnson 1988)

Hg: >12 µg total Hg $g^{-1}$ dry wt (Riisgaard et al. 1985, Riisgaard & Hansen 1990)

Pb: >10 000 µg Pb $g^{-1}$ dry wt (Schulz-Baldes 1974)

Zn: >300 µg Zn $g^{-1}$ dry wt (Manley et al. 1984, Fischer 1986).

## 'Unexplained component'

Following the initial process of quantitative toxicological interpretation of the combined biological and chemical data, it was then possible to identify those sites and regions with a large 'unexplained component' to the very low measured SFG values (i.e. those sites with 'explained' SFG values of <25 J $g^{-1}$ $h^{-1}$; and highlighted by the arrows in Fig. 8). This 'unexplained component' can be due to the effects of unknown stressors, particularly unidentified industrial and agrochemical contaminants, and/or due to presence of identified contaminants for which there is no toxicological data available at present.

The Hunstanton site in the Wash is an example of the former, where mussels have a markedly reduced SFG but none of the routinely analysed contaminants appear to be sufficiently elevated to account for the observed effects. The mussel population at Hunstanton, although situated near a sewage outfall, may also be influenced by the ouflow from the Great Ouse (Widdows & Donkin unpubl. data resulting from subsequent studies) which has a major sewage input and this may have been the source of additional unidentified contaminants. The Ythan provides an example of the latter, where mussels had very low SFG values and had high levels of PCBs and were influenced by toxic algal blooms (see below), but at present we do not have the appropriate tissue concentration-response relationships to provide a quantitative toxicological interpretation of these toxicants.

Other sites with markedly reduced SFG values which cannot be explained in terms of metals, tributyltin, hydrocarbons and polar contaminants include Teesmouth, sites in the Humber-Wash area and in the Thames estuary (Fig. 8). These are all areas likely to be influenced by additional classes of toxicants from industrial and agrochemical sources. Chemical analyses indicate that mussels had accumulated significant levels of organochlorine compounds (e.g. dieldrin, DDTs, HCB, HCHs and PCBs) at all these sites with a large 'unexplained component' to the very low SFG values. While the effects of organochlorines on SFG are not quantifiable at present (i.e. no concentration-response data available), the data reveal a statistically significant correlation ($r = -0.64$) between the 'unexplained component' of the partitioned SFG and the total concentration of organochlorines in the North Sea mussels.

Another important feature of environmental monitoring is the identification of unexpected 'hot spots'. In this study mussels from the Ythan, a rural and non-industrialised area in Scotland, were found to have very low SFG values. The subsequent and independent chemical analysis by MAFF Burnham established that the Ythan had the highest PCB levels. At present the source of the PCBs is unknown, but subsequent sampling in January and October 1993 confirmed the high concentrations of PCBs in mussels at the mouth of the Ythan, as well as at 2 sites 1 and 2 km upstream and at a coastal site north of the Ythan (NE River Purification Board, Aberdeen, and MAFF Burnham; Franklin unpubl. data).

Mussels from the Ythan also showed a behavioural response (sustained and consistent closure of the exhalent siphon in open mussels) previously only observed in response to algal toxins (Widdows et al. 1979). As a result of this observation and 'press reports' of toxic algal blooms along the northeast UK coast in late spring 1990, we obtained results of the algal toxin monitoring programme (PSP, paralytic shellfish poisoning) carried out by MAFF Weymouth. Mussels were collected from sites (Dornoch Firth to Whitstable) at frequent intervals throughout spring and summer 1990, and the results confirmed the presence of high concentrations in mussels from coastal sites between Aberdeen and Findochty (i.e. Ythan) at the time of the North Sea mussels sampling programme. This was the only area to be affected by toxic algal blooms at the time of collection (data from D. J. Alderman, MAFF Weymouth). It appears that these 2 classes of toxicant (PCBs and algal toxins) are the likely causes of the observed reduction in SFG in mussels from the Ythan. However, at present we do not have the appropriate concentration-response data to provide a quantitative toxicological interpretation and establish their relative contributions.

Further research is required to establish an appropriate toxicological database for those contaminants that are routinely included in monitoring programmes (e.g. organochlorines) as well as those which are generally not analysed in such programmes (e.g. pesticides including organophosphates, detergents and phthalates). Environmental monitoring programmes based on combined chemical and biological effects measurements should also attempt to quantify algal and other natural toxins, as well as anthropogenic contaminants.

## Offshore environmental quality

Mussels sampled from the offshore light vessels had moderately high SFG values between 12 and 14 J $g^{-1}$ $h^{-1}$, with the exception of those from Smiths Knoll and Falls which had values of 10.29 and 8.87 J $g^{-1}$ $h^{-1}$. These higher offshore SFG values are comparable to SFG values recorded at coastal sites in the north of England and largely reflect the generally lower contaminant concentrations at the offshore sites compared to southern coastal sites (Table 5).

The slight but gradual decline in SFG from Dudgeon to Smiths Knoll probably reflected a reduction in water quality resulting from the coastal currents flowing north to south and the dispersion of contaminants from the Humber plume into the coastal water. The Smiths Knoll light vessel is also in the vicinity of the gas production platforms in the North Sea. The decline in SFG at the Falls site probably reflected the light vessel's position at the southern entrance to the Thames estuary where the residual surface currents (Reid et al. 1988) may transport contaminants out of the Thames along the southern shoreline. The Falls site had the lowest SFG and the highest levels of some contaminants, such as hydrocarbons, Zn and TBT. The degree of hydrocarbon contamination found at the offshore sites generally accounted for a reduction in SFG of approximately 7 J $g^{-1}$ $h^{-1}$.

## Comparison between North Sea study and previous mussel SFG studies

In summer 1987 the Water Research Centre (WRc) applied SFG in mussels to assess environmental quality in the Humber and Thames estuaries, at sites similar to those examined in the present North Sea study (Roddie & Johnson 1988). In order to directly compare the results of the 3 studies it was first necessary to standardise to a common food concentration (i.e. 0.4 mg particulate organic matter $l^{-1}$ = 9.2 J $l^{-1}$ and a food absorption efficiency of 0.43). The recalculated SFG values for mussels transplanted by WRc to sites in the Humber and Thames estuaries indicated that the WRc SFG values are consistent with those measured in the North Sea study in 1990. For example, Bull Fort in the Humber had comparable SFG values (WRc data + 3.5 J $g^{-1}$ $h^{-1}$; PML data + 2.73 J $g^{-1}$ $h^{-1}$) and sites in the Thames estuary were also similar (WRc values ranging from +5.03 to −1.76 J $g^{-1}$ $h^{-1}$ at sites in the outer estuary in the vicinity of the sewage disposal ground; PML ranging from 8.87 at the Falls light vessel to −0.82 J $g^{-1}$ $h^{-1}$ at Swale).

An important aspect of any environmental quality or pollution study is to select an appropriate 'clean' reference site, otherwise the degree of pollution impact will be underestimated. In the WRc studies the reference sites were near the mouth of the Humber and Thames estuaries, and the spatial scale was relatively small

(<30 km). As a result of the hydrodynamics, with strong tidal currents and a tidal range of >5 m (springs), the degree of tidal mixing in these estuaries severely limits the powers of discrimination (both chemically and biologically) between sites in terms of environmental quality. Hence the need to establish 'clean' reference sites far removed from contaminant inputs (see also Widdows & Donkin 1992). In the case of the North Sea the most appropriate sites are in the Shetland Islands and the north of Scotland. Therefore many of the problems and criticisms of environmental monitoring techniques, and the results generated, relate to experimental design rather than the inability to detect effects.

## CONCLUSIONS

(1) This study demonstrated that the SFG technique can be successfully adapted and applied as a method of assessing environmental quality and pollution impact over a large spatial scale, such as the UK North Sea coastline. The geographical scale of this study is far greater than that previously used in any other pollution monitoring programme involving biological effects measurements. The technique therefore has considerable potential for application in international cooperative biological effects/chemical contaminant monitoring programmes, such as those operated in the North Sea as a whole and the Northeast Atlantic. However, before it can be used in this way, comparability of the data obtained by participating laboratories must be verified.

(2) The combined measurements of the stress response, SFG, and chemical contaminants in the body tissues of mussels were able to detect, quantify and identify some of the major toxicants causing the observed pollution effects. Experimentally derived 'cause-effect' relationships were used to provide a quantitative toxicological interpetation of the combined tissue residue chemistry and SFG measurements.

(3) The results showed a general trend of declining SFG, or increasing stress, from north to south. This reflected both the major inflow of clean water from the North Atlantic via the north of Scotland, and the overall increase in environmental contamination with increasing population density and industrial/agricultural activity towards the south. In addition, there were individual sites, as well as large estuaries, that showed mussels with relatively high degrees of stress (e.g. Ythan, Blyth, Teesmouth, Humber, parts of the Wash and the Thames estuary).

(4) At the majority of the 26 coastal and the 9 offshore sites, a large contribution towards the observed decline in scope for growth was caused by polyaromatic hydrocarbons (up to 90%). In addition, 'polar organic compounds' (probably of natural origin) could account for some of the reduction in scope for growth. At 8 sites, tributyltin concentrations were sufficient to induce a slight effect (<10% of the reduction in scope for growth). At no sites were metals accumulated to concentrations that could cause a significant effect.

(5) In this North Sea study the data reveal a significant correlation between the total concentration of organochlorines in the tissues and the size of the unexplained component of the partitioned scope for growth of mussels at sites which include the Ythan, Teesmouth, Humber and Thames estuary.

(6) An important feature of environmental monitoring programmes is the identification of 'unexpected hot-spots'. In this study mussels from the Ythan, a non-industrialised area in Scotland, were found to be severely stressed. Subsequent and independent chemical analyses by MAFF established that the Ythan had the highest PCB levels. In addition, it was the only area to be affected by toxic algal blooms at the time of sampling. These 2 classes of toxicant probably account for the reduced scope for growth but at present the appropriate concentration-response data is not available to provide a quantitative toxicological interpretation.

(7) Results from such environmental monitoring studies are important for basic, strategic and applied research in that they identify environmentally relevant toxicological questions and highlight specific areas requiring further research.

(8) The present study has demonstrated that the SFG measurement is both sensitive and rapid (i.e. results obtained within days of sampling). Such biological effects measurements can therefore provide a rapid and cost-effective initial screening of environmental quality. The subsequent analyses of chemical contaminants in tissues, which are more costly and time-consuming (i.e. months to years), could then be focussed on those mussels from sites identified as being severely stressed.

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