

**[Wherever this highlighted box appears within the document, press the F11 key and the cursor will run you through these fields as a prompt to either insert or delete information. Any text, which appears bold within these highlighted boxes, is for information purposes only. Delete this paragraph of text before printing document.]**



MONITORING SURVEY, BENTHIC – SITE SPECIFIC

**MPS/CAR/X/**

FOR

LICENCE REFERENCE NUMBER: CAR/X/

ADDRESS OF PREMISES: Premises address  
Premises address  
Premises address  
Premises address

The responsible person, as named in the licence, shall carry out monitoring at the premises as specified in the protocol below.

The Monitoring Survey Protocol may be modified annually only with the written agreement of SEPA. The modified Monitoring Survey Protocol must be dated and shall clearly state that it replaces and supersedes the previous version.

Version: X [This version supercedes Version X-1, dated YYYYYY]

Dated: XX Month 20YY

## **Introduction**

As stated in SEPA's policy and in the Fish Farm Manual (1998), scientific data are required by SEPA in order to assess the existing condition on the site or leased area.

**SEPA reserves the right to request more detailed information and/or further work if required. This may be particularly relevant, but not exclusively, to sites in areas of recognised conservation or environmental sensitivity, e.g. SAC's. The information asked for by SEPA may be subject to change and any requirements should be checked prior to any fieldwork and laboratory analysis.**

The protocols below shall be followed. The completed survey report using the Data templates obtainable from the SEPA website shall be returned in duplicate to the local SEPA office.

### **SCOPE OF SURVEY**

**Self-monitoring to be carried out by or on behalf of the responsible person in accordance with relevant Schedules of the licence.**<sup>#</sup>

<sup>#</sup> If in-feed treatment chemicals (Slice™ (emamectin benzoate) or Calicide™ (teflubenzuron)) are licensed and have been used, sampling for residues shall be completed **in the last 12 months of production** according to Part B (below). If these treatments have been licensed but **not** administered within 24 months (or current growing cycle), ignore Part B.

<sup>\*</sup> If the application is for a site which is to be located in an area containing natural heritage or other environmental concerns (see SEPA Natural Heritage Handbook Section 3.3 and also Regulation and Monitoring of Marine Cage Fish Farming in Scotland - a procedures manual, Annex C) then additional monitoring may be required, e.g. a visual survey (See Annex F)

**Applicants are strongly advised to discuss these or any alternative proposals in detail with their local SEPA Environmental Protection and Improvement Team or Marine Science staff, before undertaking any survey or analytical work.**

### **Sampling Strategies and Protocols**

Two sampling strategies are applied as part of this licence:

#### **A. Benthic Survey**

#### **B. In-feed Treatment Residues**

Where sites are stocked on a rotational basis with other sites and/or have extended stocking or fallowing regimes, additional monitoring may be required, as stated in the licence.

Any changes to this proposed sampling programme shall be agreed with SEPA prior to fieldwork commencing.

**It is recommended that due cognisance be taken of appropriate health and safety procedures during all sampling and analytical processes. Appropriate safety measures must be taken when handling chemicals, biological preservatives and vital stains.**

## A Benthic Survey

### A.1 Benthic Survey Strategy

The responsible person shall carry out the following monitoring programme during the period defined as: from the date that maximum biomass for the production cycle is first reached until one month after maximum biomass. Ideally, surveys should be conducted between May 1<sup>st</sup> and October 31<sup>st</sup>. The results should be reported in accordance with SEPA's requirements.

[At highly resuspensive sites the 30ITI AZE predicted by AutoDEPOMOD lies close to or under the cage group; in such cases, the Marine Summary sheet does not specify site-specific sampling stations and instead a fixed monitoring protocol is used. Select text from the following two alternatives, as corresponds to the Marine Summary sheet information – use blue highlighted text for modelled sample stations (AZE +/-10m), or green highlighted text for fixed sample stations (25m, 50m, & 100m). **Remove highlighting in the final document and delete this note**]

The AutoDEPOMOD output identifies two transect directions, the primary transect (number 1 in Table A-1) should be used where possible; in the event that sampling is not possible along this transect due to sea bed conditions, the secondary transect (number 2 in Table A-1) should be used. Samples should be obtained from 4 stations lying along one of the survey transects specified in Table A-1 and at 2 reference stations remote from the cage location.

Sampling stations shall be:

- at the edge (within 5m) of the cage group;
- at the edge of the Allowable Zone of Effects (AZE);
- at a station that is a distance 10m beyond the edge of AZE;
- at a station that is a distance 10m short of the edge of the AZE; and
- at 2 reference stations remote from the cage location.

The highly resuspensive conditions, indicated by the current speeds at this site, prevent AutoDEPOMOD from identifying suitable sampling stations; consequently, samples should be obtained from 4 stations lying along one of the survey transects specified in Table A-1, and at 2 reference stations remote from the cage location. The primary transect (number 1 in Table A-1) should be used where possible; in the event that sampling is not possible along this transect due to sea bed conditions, the secondary transect (number 2 in Table A-1) should be used.

Sampling stations shall be:

- at the edge (within 5m) of the cage group;
- at a station 25m from cage group;
- at a station 50m from cage group;
- at a station 100m from cage group;
- at 2 reference stations remote from the cage location.

Where the transect crosses an area of seabed over which cages have previously been positioned, this matter should be discussed with SEPA staff prior to the survey to determine whether an alternative is more suitable.

[Note to EPI staff. Transect details should be copied and pasted directly from the Marine Summary Sheet, which is supplied by the Marine Science team. **Delete this note in the final document.**]

## Table A-1 Benthic monitoring transects

Insert positions table here from the Marine Summary sheet – 'Paste as nested table'
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The reference sample stations should be located, where possible, between 500m and 1km distant from the leased area, in locations of similar exposure, depth, and sediment type and not influenced by discharges or other aquaculture operations. Where SEPA recommends a suitable location, this should be used.

Samples shall be analysed for benthic infauna and the following physico-chemical parameters:

- redox (Eh),
- organic carbon, and
- particle size analysis (PSA).

A visual description of the sediments should also be made.

Field notes must accompany any sampling and shall include any observations on weather conditions, position fixing problems, presence of fungus within the samples, anoxic sediments, etc.

**It is recommended that due cognisance be taken of appropriate health and safety procedures during all sampling and analytical processes. Appropriate safety measures must be taken when handling chemicals, biological preservatives and vital stains.**

### A.2 Benthic Protocols:

#### A.2.1. Position Fixing

All sampling locations shall have positions recorded. Suitable methods of position fixing include DGPS (Differential Global Position Fixing) and Range Position Fixing Systems. Any other method must be agreed with SEPA prior to fieldwork commencing. For more details of this process see Appendix 1

The position of any sample shall be fixed as near to the exact time the sample is taken (e.g. when the grab hits the bottom). For monitoring purposes, positions must be able to be regained irrespective of cage position and orientation in successive years.

#### A.2.2. Sample collection

Samples should be taken using a Van Veen or similar grab with top opening flaps for access and visual examination. The minimum grab size should be 0.02m<sup>2</sup>. At each sampling location 5 replicate samples shall be collected to provide sediment for biological analysis and a minimum of 2 replicates should be collected for chemical analysis. Where grabs of 0.1m<sup>2</sup> are used, 2 replicate samples for biology will suffice, and 2 for chemistry.

**Samples to be analysed for chemical and physico-chemical determinants should be obtained from separate grabs and should not be from sub-sampling of sediment obtained for other uses, e.g. faunal analysis.**

The grab, and other sampling equipment, should be washed out between the collection of each sample. Sampling personnel must follow any on-site disinfection policy.

#### A.2.3. Sediment Characterisation: Visual Assessment

A report on the condition of the sediment describing: colour (black, brown, etc.), physical consistency (sand, mud, shell gravel, etc.) and texture (soft, firm, etc.), shall be made. The presence of feed pellets and/or the white streaks of *Beggiatoa* on the sediment surface shall also be noted. The depth of organic waste overlying 'real' sediment should be noted.

#### A.2.4. Sediment Characterisation: Redox Potential (Eh)

This method is suitable for the determination of Reduction/Oxidation (Redox) potential of marine sediments. The measure of redox potential (Eh) enables comparisons of the intensity of the reducing conditions to be made on a site-to-site basis and hence act as an indication to the degree of organic loading. Eh is a qualitative measurement not quantitative (such as a concentration). In general, the more negative the Eh, the lower the ability of the sediment to exchange electrons, thus impairing chemical reactions vital for the sediment to sustain life.

Redox potential can also be used as an indicator of sulphide in sediments but not a concentration, "Eh (redox potential) is always negative in the presence of sulphide and positive in its absence", Nissenbaum *et al.* 1972. This method is suitable for sediments collected by grabs and cores.

To determine Eh values, a portable redox meter should be used with a suitable electrode and reference probe. For practicality, and as they are more robust, a combined electrode may also be used.

Before use, the probe should be tested using a reference solution of known mV value (e.g. commercially prepared Zobell's solution). All meter readings should be corrected by the difference between the expected reference solution value and actual meter reading. This accounts for inconsistencies between probes. This reference-solution correction value must be added to a second correction value that is specific to each probe type in order to convert the mV values obtained from the samples to Eh. This second step is essential as Annex A of the SEPA fish farm manual gives Eh quality standards that have been corrected to the Standard Hydrogen Electrode (SHE) and so this same correction must apply to the mV readings obtained from the samples. Typically this has been done by the addition of 198mV to the raw redox measurements but it is advisable to check with the manufacturer of the probe to ensure the correct value for the probe. Note that this value varies subtly with temperature so it is recommended that the temperature of the sediment is measured before readings begin. A range of correction values for some commonly used probes is given below in Table A-2. Care must be taken as expected mV readings may already include the SHE correction (refer to manufacturers' specification). Details of all corrections applied must be included in the final report.

**Table A-2 Correction values for some commonly used probes**

Temp. °C	Silver:Silver chloride reference probe			Calomel reference probe			Orion 96-78 combi. probe
	3M KCl*	3.5M KCl*	4M KCl*	3M KCl*	3.5M KCl*	4M KCl*	
0	226	221	224	265	262	260	260
1	225	220	223	265	261	260	259
2	225	220	222	264	261	259	259
3	224	219	221	264	260	258	258
4	224	219	220	263	260	258	258
5	223	218	219	263	259	257	257
6	222	217	218	262	258	257	257
7	222	217	217	262	258	256	256
8	221	216	216	261	257	255	256
9	221	216	215	261	257	255	255
10	220	215	214	260	256	254	255
11	219	214	213	260	256	253	254
12	218	214	212	259	255	253	253
13	218	213	211	259	255	252	252
14	217	212	210	258	254	251	251
15	216	212	209	258	254	251	250

20	213	208	204	257	252	248	249
25	209	205	199	255	250	244	246
30	205	201	194	253	248	241	242

Samples for redox should not be sub sampled for any other chemo-physical analyses.

Samples should be collected by small grab or corer. As a minimum, at each sample location, two profiles of redox measurements should be made. Each profile should be measured in a separate grab/core sample. Where possible, measurements should be made at 1cm intervals from the sediment surface to the bottom of the sampler (i.e. at 0, 1, 2, 3, 4 and 5cm, depending on the depth of sediment in the sampling device) taking care not to allow the probe tip to touch the metal sides of the grab.

Measurements (mV) are taken immediately on collection of the sample whilst still in the sampler. The meter readings should be allowed to stabilise before recording.

#### A.2.5. Sediment Characterisation: Organic Carbon or Organic material (Loss on Ignition)

As a minimum, at each sampling location, one sub-sample (~50ml) of sediment shall be collected from the sample surface (0-2cm). The sample is stored in an airtight container and deep frozen, or frozen then freeze dried, for later analysis.

It is recommended that, as a minimum, the Loss on Ignition (% LOI) method be used following the procedure of Allen *et al.* (1974). Total organic carbon can also be measured by combustion (the method used must be specified).

#### A.2.6. Sediment Characterisation: Particle Size Analysis

As a minimum, at each sampling location, one sub-sample (100ml) of sediment should be collected from the surface (0-2cm) and stored in suitable containers

Assessment of the particle size distribution of sediments is generally determined by dry sieving or laser granulometry, though other methods are possible. The method used must be quoted in any final report.

For laser analyses, fresh samples may be used or samples can be stored overnight in a fridge at 4°C or longer term in a deep freeze at -18°C. Laser granulometry results are reported graphically and in table format.

For dry sieving, storage at -18° C, prior to drying, is recommended. Samples may be freeze or oven dried. Sieved sediments pass through each sieve in a stack. The size fractions are weighed and the results are expressed as a percentage of the total.

#### A.2.7. Benthic Biology: Sample preparation

Each replicate sample must be processed separately. Samples are usually sieved on site with each sample being washed with seawater, through a 1mm metal mesh sieve. The material retained on the sieve is then transferred to a clearly labelled, airtight container, taking care not to leave animals on the sieve. Alternatively in the field, samples may be potted whole and separately into labelled sealed pots or double plastic bags.

Samples are subsequently preserved using buffered formalin (50g sodium tetraborate in 2.5litres of 40% formaldehyde solution, diluted with seawater to 4% solution), as soon after collection as practicable.

#### A.2.8. Benthic Biology: Sample Analysis

Prior to sorting and identification, formaldehyde is removed from the sample by thorough rinsing with fresh water through a 1mm sieve. To further aid sorting, light organic matter and fauna may be elutriated off and sorted separately.

The use of a vital stain, e.g. Rose Bengal, is not essential but is recommended to aid sorting. If the sample is to be stained, the sieve and its contents are placed in a white sorting tray, containing 5ml of Rose Bengal solution and enough water to cover the contents of the sieve; this is left to stain for at least 20mins. The sample is then thoroughly rinsed again.

The sample retained on the sieve (sieve residue) is then washed into a white sorting tray, with enough water to cover the sample. The sieve is checked to ensure no animals are left in the mesh, and then cleaned to ensure no cross-contamination. The sample identification label must remain with the sample at all times.

All the fauna is picked by hand, using forceps and/or pipette, under suitable illumination and magnification, and transferred into suitable labelled containers. Each tray should be sorted in a methodical manner to minimise the risk of missing any fauna. The specimens should be stored in 70% IMS/water preservative which can be made viscous to prevent evaporation by the addition of glycerol.

The analysis must be carried out by a competent benthic taxonomist. Before submission of survey data SEPA requires evidence of training and continuing competency of analysts.

SEPA also requires evidence of participation in external AQC schemes, such as National Marine Biological AQC Scheme (see also Annex B regarding Quality assurance and control).

The fauna should be identified and enumerated to the lowest taxonomic level, with the use of stereo/compound microscopes, and standard identification keys and references (a comprehensive list of these works is available from SEPA on request). Nomenclature should follow the MCS species directory as far as is possible or the most recent taxonomic publication on that group. The level of identification expected for major taxonomic groups is species level.

Analysts should note the following:

- The anterior ends of specimens are counted to determine the abundances of each taxon. Where a species does not have an obvious head, then it is counted if more than half of the specimen is present,
- Nematodes <1cm in length are considered to be meiofauna – their presence may be noted, but only nematodes >1cm are to be counted,
- Estimated counts will only be acceptable for common taxa, and where their abundance in a sample is >200 individuals,
- Certain taxa (e.g. colonial, encrusting) are not included as quantitative taxa but are simply recorded as present (qualitative taxa), and
- A note will also be made of any fauna that are listed on the UK Biodiversity Action Plan list, which may be found on JNCC's website ([www.ukbap.org.uk](http://www.ukbap.org.uk)).

**Sub-sampling of taxa is not recommended. If considered necessary, it must be agreed with SEPA in advance.**

Following analysis, the containers of preserved identified specimens must be retained for a period of at least 12 months and must be made available on request by SEPA for auditing purposes.

The raw data shall be reported as a species/abundance matrix both on a by-replicate and by-station basis. Faunal indices including diversity (Shannon-Weiner  $H' \log_2$ ), evenness indices (Pielou, 1975) and Infaunal Trophic Index (Codling and Ashley, 1992) should be derived.

Further notes to aid statistical analysis is given in Appendix 2. Additionally, extensive descriptions of the methodologies can be found in Rees, Moore, Pearson *et al.* (1990), and Peilou (1975).

Quality assurance and Analytical Quality Control procedures are detailed in Annex B of the Fish Farm procedures manual.

## B In-feed Treatment Residues

### B.1 In-feed Treatment Residues Survey Strategy

If the in-feed sea lice treatments Slice™ and/or Calicide™ are included in the licence, and the products have been used, then sediment samples shall be collected and analysed for these compounds.

Sediment samples shall be collected **in the last 12 months of production at the same time as the benthic survey , if possible** according to the following strategy:

- Slice™. Samples shall be taken between 80 and 169 days after the cessation of the treatment,
- Calicide™. Samples shall be taken between 10 and 30 days after the cessation of treatment.

Note that if it the timings of the peak biomass and in-feed sampling window do not coincide then SEPA Marine Science should be contacted for advice.

**If Slice™ and/or Calicide™ have not been used within 24 months (or current growing cycle), this survey need not be carried out.**

Samples should be obtained from 2 stations along one survey transect lying along the same direction as the benthic monitoring, as specified in Table A-1 above. Sampling stations shall be:

- at the cage edge; and
- 100m distant from the cage edge.

**[Note to EPI staff. Additional samples for residues analysis may be requested at some sites by the modelling team. If requested then the text given below should be completed and inserted here using the information given in the modelers email. Delete this note and the highlighted text below if additional sampling is not required.]**

**In addition, a single sampling station at a distance of XXXm along the predominant current direction, that is XXXdegrees from the centre of the cage group (as near to XXXd XXXm N XXXd XXXm W as is practicable), shall be included in the sampling strategy.**

For clarity, this information is summarized in Table 2 below:

**Table 2 In-feed sampling station positions**

Station name	position
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Cage edge	0m along direction as given in Section A1.
100m	100m along direction as given in Section A1.
Supplementary station	[EPI STAFF ALSO FILL IN INFO FROM MODELERS HERE IF REQUIRED]

The samples taken at these locations shall be analysed for in-feed treatment residues only.

**B.2 In-feed Residues Protocols:**

**B.2.1. Position fixing**

Refer to the Position Fixing section (A.2.1) in the Benthic Survey protocol above.

**B.2.2. Sample collection**

At each sampling stations are collected by grab (as per benthic biology, A.2.2 above) although care should be taken when sub-sampling from within a grab to ensure that cores are representative of undisturbed sediment and that the surface layer is intact. 3 sediment cores shall be taken to 5cm depth and kept separate as individual replicates. Samples shall be placed in suitable glass containers with a metal-foil-sealed lid.

**B.2.3. Sample analysis**

Sediment samples should be frozen immediately after sampling then submitted to a reputable laboratory for analysis (evidence of laboratory quality and/or appropriate analytical capability will be sought by SEPA).

For emamectin benzoate, a validated technique must have a limit of detection (LOD) that will be suitable to match the requirements of the near field trigger value and far field sediment quality standard; this should be one third of the standard in question or lower. Ideally LODs that meet one tenth of the relevant standard will improve overall confidence in the data. Detailed methods are available from Schering-Plough Animal Health Limited.

For teflubenzuron, a validated technique must have a limit of detection (LOD) that will be suitable to match the requirements of the near field trigger value and far field sediment quality standard; this should be one third of the standard in question or lower. Ideally LODs that meet one tenth of the relevant standard will improve overall confidence in the data. Detailed methods are available from Nutreco Limited.

Results should be reported in a table indicating both replicate number and sample location. The unit of measurement must be clearly stated.

## **Reporting**

The data and accompanying text shall be reported in the Site Specific Monitoring template. The details of this system can be found in Attachment XII. These are spreadsheet-based templates and the preferred format for submission to SEPA shall be CD or DVD. If the operator has difficulties in completing this survey template then they should contact SEPA to seek advice. It is not recommended that operators submit paper records as an alternative without prior discussion.

Completed benthic survey reports shall be submitted to SEPA within 16 weeks. Should additional surveys be required e.g. seabed video, the submission period is 12 weeks. If difficulty in achieving this is experienced, then the local SEPA team must be notified as soon as possible.

## **Evaluation of survey report**

The survey report shall be evaluated by SEPA with reference to the environmental standards set out in Annex A of the Fish Farm Manual. Normally, reports will be classified either as: Satisfactory, Borderline, or Unsatisfactory. Borderline evaluations are nominally a satisfactory classification but indicate stress on the sea bed.

## **Auditing of Results**

SEPA may require (at any time) evidence of quality assurance and control on any procedures or processes being undertaken by the responsible person, or their agents, or require independent audit of any resulting data.

## **References**

Allen, S.F., Grimshaw, H.M., Parkinson, J.A. & Quimby, C. (1974) Chemical Analysis of Ecological Material, 1st edn. Blackwell Scientific Publications, Oxford, UK

Codling, I. D. and Ashley, S. J. (1992). Development of a biotic index for the assessment of pollution status of marine benthic communities. Final report to SNIFFER and NRA. NR 3102/1

Pielou, E.C (1975) Ecological diversity. Pub. Wiley, New York 165pp

Rees, H. L., Moore, D.C., Pearson, T.H., Elliot, M., Service, M., Pomfret, J. and Johnson, D. 1990 Procedures for the Monitoring of Marine Benthic Communities at UK Sewage Sludge Disposal Sites. Dept. of Agriculture and Fisheries for Scotland. Scottish Fisheries Information Pamphlet No. 18. 79p.

## Appendix 1 Notes to Aid Position Fixing

### Position Fixing

The position of any point on a surface can be fixed using a two-dimensional co-ordinate system (X and Y). Two principle systems of X and Y co-ordinates are used within the UK:

- **National Grid References (NGR).** A full NGR consists of two six-digit numbers, an Easting and a Northing and is accurate to 1m. In practice many locations are not known (or required) this accurately and a position is more usually given by an alphanumeric, e.g. NS 300 710. Such an NGR is only accurate to 100m; where possible SEPA will record a 10-digit alphanumeric NGR that is accurate to 10m.
- **Latitude/longitude position fixing** is routinely used for navigational purposes and is usually invoked as a marine/coastal site descriptor once the location is identified by other means, e.g. GPS, Range Position Fixing. The angle west or east from the meridian is given in degrees, minutes and seconds.

There are several reference systems against which the three-dimensional position in space may be recorded. Although these may use the same reference units, i.e. degrees of latitude and longitude, there are differences between datums and the idealised reference shapes (geoids) used to approximate the surface of the earth. Thus any one location may have significantly different co-ordinates under different systems or conversely one co-ordinate pair may refer to positions that may be up to 1km apart when different datums are used. So, it is important to include the name of the reference datum or co-ordinate system when quoting positions.

GPS receivers are commonly set to WGS84 (equivalent to ETRS89) as a default. Positions on OS maps and Admiralty charts for British waters, and National Grid References are specified with respect to OSGB36, which is often provided as an alternative datum option on GPS receivers sold in the UK. A software tool for conversion between the WGS84 and OSGB36 and further information about geodesy is available from [www.gps.gov.uk](http://www.gps.gov.uk).

### Methods of position fixing

- **GPS (Global Position System) and DGPS (Differential GPS)** are satellite navigation systems. Transmissions from satellites are detected by a receiver and calculated into positional data. GPS accuracy is around 10m, depending on the receiver, the number of satellites in view and other factors. High accuracy (<1m) may be achieved by using a differential correction system. It is important to be aware of the datum against which a receiver is referencing the positions it produces (see above).
- **Range Position Fixing Systems.** These are normally microwave devices that display the distance from a master transmitter to a set of onshore 'slaves' at precisely known locations. The accuracy depends on the accuracy of the position of these slaves. Accuracy of 25m - <1m can be achieved.

## Appendix 2 Biological Data Analysis

### Primary measures and Univariate methods

Primary measures are simply the number of taxa and number of individuals at each site, whereas univariate methods refer to the calculation of diversity indices. A variety of indices have been proposed but the widely used Shannon-Wiener index ( $H'$ , using logs to the base 2) is recommended by Rees *et al* (1990) and Pearson and Rosenberg (1978). Diversity is considered to have two components; species richness and equitability. Equitability can be measured using Pielou's evenness index (Pielou 1975).

These single figure measures represent a severe simplification of the biological data and consequently, if used without other complementary methods, may lead to misinterpretation. They can be extremely useful, however, if a gradient of effect is apparent or expected. In addition their simplicity suggests that the derivation of biological standards may be practical.

### Multivariate Methods

These methods have the advantage that the maximum amount of information in the original 'species at sites' data matrix is maintained because species identity is not lost during the analysis. Multivariate techniques are known to be very sensitive methods for analysing changes in community structure (Warwick and Clark, 1991).

There are two main types of multivariate methods as follows:

- a) Ordination, which condenses the biological information down to a two dimensional graph of the relationship between sites. The widely used techniques are: Multi-Dimensional Scaling (MDS) and Dendrograms available in the statistics software package PRIMER and De-trended Correspondence Analysis (DECORANA) available in the Cornell Ecology Programme (CEP).
- b) Classification, where the sites are sorted into groups on the basis of the similarity of the biological information between replicates and/or stations. Two widely used classification techniques are Two-way Indicator Species Analysis available in the CEP package as TWINSPAN and Group Average Sorting of Bray-Curtis Similarity Coefficients available in PRIMER.

All four of these methods are described by Rees *et al* (1990).

Before any multivariate analysis of the biological data takes place, it is common practice to transform the data. The purpose of transformation is to alter the balance between the contribution from common and rare species in the analysis. Effectively all transformations increase the contribution of rare species and decrease the contribution of common species (see Rees *et al*, 1990, for further details).

Multivariate methods are particularly useful in examining the relationship between environmental data and the biological data, using for example the BIOENV programme in PRIMER. BIOENV selects environmental variables, which best explain the community pattern by "maximising a rank correlation between their respective similarity matrices" (Carr, 1996). Spearman Rank Correlation coefficients (Siegel and Castellan, 1988) can be used in conjunction with DECORANA to elucidate the relationship between environmental variables and sites. Data on the environmental variables in question must be available for each sampling station before any relationship can be established. Furthermore multivariate analysis cannot prove a relationship exists but it is strong evidence that one may exist.

The application of standards to multivariate methods is complex but there is no reason why a predictive system similar to the RIVPACS Scheme used in freshwater (Moss *et al*, 1987) could not be developed for marine systems. Rees *et al* (1990) have outlined the approach necessary.

**Multivariate methods form an essential part of the consideration of benthic data due to their sensitivity and must be used to confirm the conclusions of the univariate methods applied.**

### **Infaunal Trophic Index**

**The Infaunal Trophic Index (ITI) is a biotic index, which has been developed by the Water Research Centre (WRc). It relies on the assessment of the changes in the feeding (trophic) mode of benthic organisms in areas subject to elevated levels of organic enrichment. Details of the index and its use can be found in WRc (Codling and Ashley 1992). The index was developed from a system originally devised for use in California (Word, 1979). This index differs from the preceding essentially numerical methods because knowledge of the ecology of the taxa involved is required. Thus it forms a useful complement to the numerical methods in that the data are considered from an alternative perspective. The index was found to respond satisfactorily to pollution gradients from a variety of sources including sewage and industrial discharges.**

**The ITI has great potential with regard to the setting of standards. The index has values that range from 0 to 100 and results can be interpreted as follows:**

<u>Index Value</u>	<u>Assessment</u>
60 to 100	Community 'Normal'
30 to 60	Community 'Changed'
< 30	Community 'Degraded'