

**MASQ: Monitoring and Assessing Soil Quality in  
Great Britain.  
Survey Model 6: Soils and Pollution**

**Technical Record  
E1-063/TR**

**MASQ: Monitoring and Assessing Soil Quality in Great Britain.  
Countryside Survey Module 6: Soils and Pollution**

R&D Technical Report E1-063/TR

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## **Keywords**

Countryside Survey 2000, monitoring, soil quality, heavy metals, soil pH, organic matter content, organic pollutants, microbial diversity, invertebrate diversity.

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## EXECUTIVE SUMMARY

The MASQ (Monitoring and Assessing Soil Quality) project was carried out by the Centre for Ecology and Hydrology and funded jointly by The Department of Environment, Food and Rural Affairs (DEFRA), The Environment Agency (EA), Scotland & Northern Ireland Forum for Environmental Research (SNIFFER) and the Natural Environment Research Council (NERC). The project was started in 1998 and completed in 2001.

The MASQ project undertook a nationwide survey of soil biological and chemical properties as part of The Countryside Survey 2000 (CS2000). The overall objective was to provide good quality datasets for soil invertebrate and microbial communities, soil pH and organic matter, as influences on soil biology and characteristic properties of British soils, and two groups of potentially widespread chemical pollutants (heavy metals and persistent organic pollutants). CS2000 was a cost-effective framework for integrating an assessment of soil properties with detailed landscape, land-use, soils and vegetation data since all data were obtained from the same plots.

This report provides summary information on methods and descriptions of individual datasets. Results from analyses by national scale stratifications used within CS2000 illustrate the range in values and variability within the British countryside for each soil property and highlight the potential for future exploration of the datasets.

1071 soil samples were analysed for pH in water with 769 from the same location as the Ecological Survey of 1978. Soil pH from both surveys corresponded with expected patterns. Soil pH ranged from 3.2 to 8.71 with acidic soils predominating. Upland soils were most acidic while arable/horticultural areas, mainly in the lowlands of England and Wales, were the most alkaline. Preliminary results suggest an overall increase in soil pH since 1978. Variation in pH across the British countryside can be used to explore the influence of site factors (soil type, land use, management, geology etc).

1067 soil samples were analysed for loss-on-ignition (LOI) as a measure of soil organic matter content (SOM); 744 from the same location as the 1978 Survey. In both surveys, SOM showed a bi-modal distribution. The highest SOM values were recorded from wetland habitats and upland, more acidic soils and the lowest in lowland agricultural habitats. Preliminary results suggest an increase, or at least no change, in SOM over the last twenty years. Further analyses are required to determine whether these results are consistent with *in-situ* accumulation, method differences and/or small-scale heterogeneity. The conversion of SOM to soil organic carbon contents (SOC) was not reliable using available equations. Reliable estimates could be obtained by analysing total carbon in a sub-set of CS2000 soil samples across the range of SOM values.

1080 soil samples were analysed for the total concentrations of seven heavy metals by ICP-OES; Cadmium (Cd), Copper (Cu), Lead, (Pb), Nickel (Ni), Vanadium (V) and Zinc (Zn). Non-normal distributions of concentrations were observed for all metals. Similar distribution patterns were obtained for associated metals (Cd/Pb, V/Cr/Ni and Cu/Zn). Mean metal concentrations were significantly higher in England and Wales than in Scotland while median concentrations were lowest in podzols and peat soils and heath and bog sites and highest in lowland agricultural areas.

A novel analytical method was developed using GC-MS for the analysis of polyaromatic hydrocarbons (PAHs), polychlorinated biphenyls (PCBs) and organochlorine pesticides (OCPs) in soils. Over 120 samples have been analysed, to date. 35 PCB congeners and 14 PAHs have been quantified, along with 7 OCP pesticides or their persistent metabolite, and include at least one pesticide in current use. Preliminary results revealed that the distribution of persistent organic pollutants (POPs) was not homogenous in British soils. A study of within field heterogeneity showed that PCB 180 concentrations could differ by more than an order of magnitude over 10 m while PAH benzo[*a*]pyrene concentrations were less variable.

Soil invertebrates were extracted and identified from 1052 soil samples. 25 major taxonomic Orders were recorded and species identified for Coleoptera, Diplopoda, Isopoda, Chilopoda, Pseudoscorpions and Oribatid mites. Collembola species identification has been initiated. The Oribatid mite identification has produced several new records for Britain, with potential new species and a new genus to science. Two groups of soil invertebrates, Collembola and Acari, were recorded in sufficient numbers to further examine distribution patterns and relationships with other soil properties and the wider environment and investigate whether variation corresponds to site-specific environmental factors or higher-level spatial and/or temporal factors.

940 soil samples were analysed for numbers and functional diversity of heterotrophic bacteria. The mean number of viable cells was  $2.51 \times 10^7$  colony forming units (cfu)  $\text{g}^{-1}$  dry weight of soil while the average global activities ranged from 17.1 to 302.9 OD<sub>590</sub> (Optical Density). The data were skewed towards those with low numbers or activities. The next stage will be to compare how published data fit into these distribution patterns. The responses to the 95 BIOLOG GN substrates were analysed as a whole for the purpose of this report. Responses for each set of carbon sources should be analysed separately so that the discriminating power of different substrate groups can be evaluated. Further analyses of the BIOLOG data will also have to account for the correlation between global activities at low inoculum densities.

All MASQ data and metadata have been fully integrated into The Countryside Survey 2000 Integrated Data System (CIDS), an ORACLE based data management system that ensures safe storage, access to data for future analyses and links among data from CS2000 and earlier surveys. CIDS is maintained by CEH Computer support Staff via CS2000 Module 13.

The data and summary statistics presented for the individual soil properties, demonstrate that the MASQ project has produced good quality national datasets that can be used to investigate the distribution patterns of, and relationships between, soil biological and chemical properties and their environment. The project also established that soil biological properties could be assessed, alongside other soil properties, within national scale soil monitoring programmes.

Future research priorities are presented in the over-view. These are not a comprehensive list but a starting point from which research priorities, that address research and policy requirements, can be explored, identified and undertaken. A priority for the next stage of data analyses must be the investigation of relationships between the invertebrate and microbial properties of soils and their environment e.g. other soil properties, soil type, habitat, land use and geographical location.

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# 1. INTRODUCTION

## 1.1 Scientific and policy background

### **Countryside Survey and Soils – a historical perspective**

The Royal Commission on Environmental Pollution (RCEP) raised the importance of soils in the U.K. with the publication of its nineteenth report, *Sustainable Use of Soil* (RCEP, 1996). In this, the Commission recommended that soil should have equal status with air and water in environmental legislation. The report stressed the need for the assessment and monitoring of soil quality that included measurements of certain chemical and physical attributes, specific pollutants, and some biological attributes and included all major soil types and a representative range of land uses. It was noted that it would be sensible to base an assessment on sites for which some baseline data was already available. The Countryside Survey of Great Britain was suggested as an existing and suitable baseline scheme with such sites.

The RCEP report (RCEP, 1996) also identified the development of indices of soil biological activity and diversity as a key research priority. The major difficulty in developing such indices is the need for baseline data from which a suitable set of standards can be formulated. For example, a review for the Environment Agency highlighted that existing data were inadequate for an assessment of the potential for developing indicators from soil invertebrates since methodologies were inconsistent and lack sufficient geographical coverage (Weeks, 1996).

A nationwide survey was proposed to establish a framework for comprehensive baseline datasets of soil biological properties. The Countryside Survey 2000 (CS2000) provided a cost-effective framework for integrating an assessment of soil biological properties with detailed landscape, land-use, soils and vegetation data. The MASQ (Monitoring and Assessing Soil Quality) project commenced in March 1998 with the overall objective to provide datasets that directly link soil biological and chemical properties to a range of other environmental parameters available from the Countryside Surveys.

### **Soil biodiversity**

Following the 1992 Earth Summit in Rio de Janeiro, Hagvår (1998) highlighted three reasons why soil biodiversity should be protected. The first, and most researched, is the fundamental role that soil organisms have in maintaining soil processes that are essential to the functioning of all terrestrial ecosystems; their “ecological” significance. The second is their usefulness; the “utilitarian” reason. Soil organisms have been used widely in biotechnology, e.g. to improve nitrogen fixation, for the bioremediation of contaminated soils (Graham and Vance, 2000; Lynch, 1997) and in screening for potential pharmacological compounds (Lynch, 1997; Turner, 1996).

The immense genetic pool within the soil suggests that there is, still, significant potential for novel products and applications. The third reason is ethical. The soil contains some of the oldest organisms on earth that should, as identified by the Convention on Biological Diversity, have value in their own right. The broad acceptance of the significance of soil organisms, in particular their ecological significance, has led to increasing consideration of soil biological properties within the

wider context of sustainable management of our soil resources and the assessment of soil health (Doran and Zeiss, 2000). It is within this context that an assessment of soil biodiversity was deemed timely within the Countryside Survey of Great Britain.

While it is acknowledged that the scale of effort and taxonomic expertise imposes serious limitations on our ability to assess soil biodiversity (Lawton *et al.*, 1998), some of these limitations are being tackled by the development and application of novel genetic and functional analytical techniques for the characterization of soil communities (e.g. Grayston *et al.*, 1998; Widmer *et al.*, 2001). One simple approach to evaluating functional diversity of a microbial community is based on the ability of members of the community to utilize different substrates.

Garland and Mills (1991) first introduced the use of community-level carbon source utilization patterns for comparison of microbial communities from different habitats. These authors used the Biolog<sup>TM</sup> microtitre plate system (BIOLOG Inc., Haywood, CA, USA) that is relatively inexpensive and does not require technically skilled staff to perform the assay. Statistical analysis of the results can provide information on the number of substrates utilized by different samples (diversity of metabolic potential) and cluster samples that have similar values (high or low activity) for certain substrates or certain groups or guilds of substrates (Zak *et al.* 1994).

Invertebrate diversity assessments still rely heavily upon morphological identification by trained specialists that can be costly and time-consuming. The potential to use molecular genetic techniques has been recognized but remains, as yet, largely undeveloped. Soil invertebrates have also been proposed as potential bioindicators and most studies have concentrated on the dominant groups e.g. nematodes, Acari, Collembola, earthworms (Van Straalen, 1998).

Two groups of microarthropods, Collembola and Acari, have received significant attention, as they are often highly abundant in a wide range of soil types and habitats while being relatively easy to extract from soil. Within the Acari, species richness of the Oribatid mites has also shown potential for assessing the impacts of land use and pollution (Vu *et al.*, 2000; Alvarez *et al.*, 2001; van Straalen and Verhoef, 1997).

## **1.2 Assessment of soils within CS2000 – an overview**

This programme has provided unique datasets that directly link soil biological and chemical properties to each other and a range of other environmental parameters that are available from the Countryside Surveys (e.g. ITE Land Class, soil series, land use, vegetation, etc). This report provides summary information on methods, quality control and a description of each MASQ dataset.

Summary statistics have been used to describe the datasets with analyses with respect to national scale stratifications used by CS2000 i.e. Environmental Zone, Broad Habitat, Major Soil Group and CVS Aggregate Vegetation Class. These illustrate the range in values and variability within the British countryside and highlight the potential for future exploration of the datasets.

### **1.2.1 Overall objectives**

To provide good quality data about soil chemical and biological properties for the development of national databases and to improve the understanding of links between soil biology, chemistry and the wider environment to support the development of suitable, effective strategies and policies relating to soil protection.

### **1.2.2 Specific Objectives**

- 1 To provide a national overview of chemical and biological soil properties and a baseline against which specific sites can be compared by carrying out a programme of soil sampling, at the locations used in the CS2000, by the field surveyors operating under the CS2000.
- 2 To measure pH and soil carbon content and carry out a range of chemical analyses and a laboratory evaluation of faunal diversity and microbiological status to provide a baseline for the monitoring and assessment of soil quality in England and Wales.
- 3 To integrate information on chemical and biological properties and to look at it in terms of soil quality assessment and the wider terrestrial environment.

### **1.2.3 Work Programme**

The project was divided into four stages.

#### Stage 1: Protocol development and training (December 1998 to April 1999)

Work during this phase included:

- trials of sampling and transport methods suitable for sampling a range of soils;
- increasing the capacity of existing laboratory facilities;
- development of tailor-made sampling kits;
- identification of appropriate existing, and development of new, protocols for field sampling and subsequent laboratory processing of samples.
- training of field survey teams, explanation of project rationale will be explained to the surveyors, sampling collection and packing (for posting) procedures demonstrated and potential problems discussed; sampling kits issued; Health and Safety aspects identified and appropriate training provided;
- field sampling and return of samples to Merlewood;
- preparation of a short scoping study for the heavy metals and organic pollutants to consider potential substances for analyses, suitability of analytical methods, quality assurance methods to be employed and cost.

#### Stage 2: Countryside Survey sampling and immediate sample processing (April 1999 to August 2000)

Work during this phase included:

- field sampling by CS2000 surveyors
- return of samples to CEH Merlewood;
- immediate sample processing at CEH Merlewood; extraction of soil fauna, determination of pH and loss of ignition, air drying, 2 mm sieving and storage

of samples for metal analysis; storage of sub-samples, as received at Merlewood, at -86°C prior to microbial evaluation and organics analyses.

### Stage 3: Biological and chemical analyses of samples (April 1999 to July 2000)

Work during this phase included:

- laboratory examination of soil fauna to identify and enumerate taxa;
- laboratory examination of soil microbial diversity using BIOLOG technology;
- analysis of heavy metal concentrations in soil; total Cd, Cr, Cu, Ni, Pb, V & Zn
- analysis of select organic compounds

### Stage 4: Evaluation and reporting (June 2000 to August 2001)

Work during this phase included:

- development and population of databases for the following parameters:
  - locational and associated soil data
  - soil acidity
  - soil organic matter and carbon contents
  - soil faunal diversity
  - soil microbial diversity
  - soil heavy metal contents
  - soil organic compound contents
- analysis of results, particularly in terms of identifying and interpreting temporal and spatial change;
- reporting and integration of results.

#### **1.2.4 Proposed outputs from the project**

The project outputs were modified as the project progressed with the emphasis on the production of good quality data. Additional requirements were identified for method validation and development, as discussed in the relevant sections, while samples were included from Scotland to complete the coverage of Countryside Survey sample locations. Summary analyses also highlighted that key issues should be addressed before interrogative analyses commenced to address specific objectives outlined at the start of the project. These objectives are re-addressed under future priorities. The specific objectives and outputs for the individual chemical and biological properties are detailed in the relevant sections.

1. Protocols for sampling, sample handling, faunal extraction, sample storage and analyses developed
2. Summary tables of samples received and archived by soil type, geographical location, ITE Land Class and land use/cover
3. Provision of material for CS2000 Newsletter and Website
4. Project reports
5. Scoping study for heavy metals and organic pollutants
6. ORACLE and GIS spatially referenced datasets in digital form that are Countryside Information System (CIS) compatible. Soil data will be provided for and incorporated within the data management systems established within Countryside Survey 2000 Module 13. Arrangements for access to data will follow the data access policy agreed by the Countryside Survey 2000 Advisory Group.

The datasets:

- Soil acidity
- Soil organic matter and carbon
- Heavy metal contents in soil
- Organic pollutants in soil
- Soil microbial diversity
- Soil invertebrate diversity

7. Production of summary statistics:

- *Soil acidity and organic matter content*
  - Quantification of soil pH and organic matter content in 1978 and 1998/9 and changes over this period.
  - Definition of patterns with respect to geographical area, habitat, soil type and vegetation type.
- *Heavy metal contents in soil*
  - Quantification of seven heavy metals in soil from samples taken in 1998/9. Definition of patterns with respect to geographical area, habitat, major soil type and vegetation type.
  - Evaluation in terms of current guide values and limits for metals.
  - Comparison with other regional or national data sources.
- *Organic pollutants in soil*
  - Quantification of selected organic compounds from a sub-set of samples taken in 1998/9.
  - Definition of patterns with respect to geographical area, habitat, major soil type and vegetation type.
- *Soil microbial diversity*
  - Quantification of metabolic potential and biodiversity of soil microflora from samples taken in 1998/9.
  - Definition of patterns with respect to geographical area, habitat, major soil type and vegetation type.
- *Soil invertebrate diversity*
  - Quantification of invertebrate diversity (taxa richness and diversity indices) from samples taken in 1998/9.
  - Definition of patterns with respect to geographical area, habitat, major soil type and vegetation type.

## **2. THE COUNTRYSIDE SURVEYS OF GREAT BRITAIN**

### **2.1 Overview**

The Countryside Surveys provide a national network of sites across Great Britain, representing the main types of landscape, land cover and soil groups (Haines-Young *et al.*, 2000). Each site comprises of a 1 km square. These sites are surveyed at intervals for land cover, landscape features and vegetation species composition. This framework is used to obtain information necessary for reporting on biodiversity in the wider countryside, measuring progress towards sustainable development and detecting the impacts of human activities and global environmental change (Firbank *et al.*, in press). Many of the sample sites were first visited in 1978 and subsequently in 1984 and 1990, thus providing a time series of changes in the countryside. The first survey in 1978 was based on 256 squares, representing the main types of landscapes, land cover and soil groups in Great Britain (Barr *et al.*, 1993). Some of the sites have been now been surveyed a number of times, beginning in 1978.

### **2.2 Sampling the British Countryside**

The sampling methods used for the Countryside Surveys are based on a rigorous, statistical approach (Barr *et al.*, 1994). The sampling sites form a restricted, stratified random sample. The stratification used in the Countryside Surveys is adapted from the approach developed for the classification of vegetation in British woodlands (Bunce, 1982 & 1989), which was also based on a stratified random sample. The strata were derived, in 1978, from a multi-variate analysis, using Twinspan, of available data on relief, climate, geology and settlement for a sample of 1228 1 km squares that lay at the intersections of a 15 x 15 km grid of the OS grid for GB; the number of squares used in this analysis being constrained by available computing power. The multi-variate analysis was stopped at the 32 class level and these classes have subsequently been referred to as the ITE Land Classes. Subsequently, all 240,000 1 km squares in GB were allocated to one of the 32 Land Classes. Since then the number of Land Classes has increased to 40.

For the first Ecological Survey of Great Britain in 1978, eight squares were chosen at random from each of the 32 Land Classes, using the 15 x 15 km grid, giving a total sample of 256 squares for the field survey. In 1984, the sample size was increased to 12 per Land Class, giving total of 384 survey squares. By, 1990 additional squares were added according to the size of the Land Classes. In the 1978 survey, the 1 km squares were selected randomly within the various sample strata.

For CS2000, a number of the Land Classes were further subdivided, giving a total of 40 Land Classes, to facilitate selection of additional sampling squares to allow national estimates, of cover for example, to be derived for Scotland and for England and Wales based on squares located within the relevant country. The number of survey squares was increased to 569, with 366 in England and Wales and 203 in Scotland; 366 in England and Wales, and 203 in Scotland (Figure 2.1). Only 256 of these were sampled for soils.



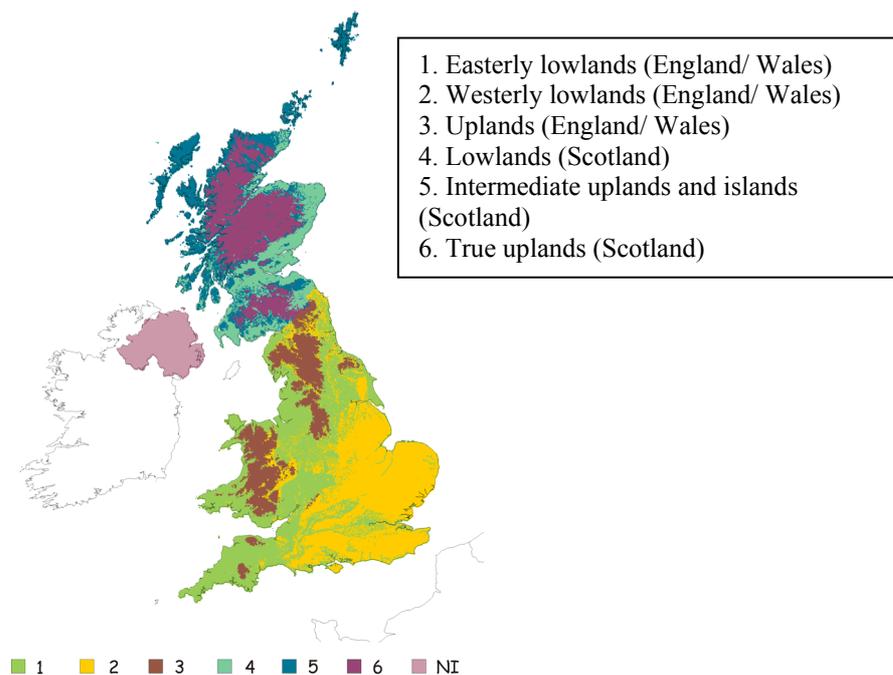
## 2.3 Reporting Units

Both the survey techniques and the data storage mechanisms employed within the Countryside Surveys allow information to be re-aggregated later at the data analyses and interpretation phase. These facilitate reporting of different types of statistics at different scales from national to specific vegetation types. The following provides a summary description of the main forms of reporting units.

### Environmental Zones

In addition to the national and separate country estimates, more detailed geographical breakdowns of the field survey data are possible. The six major Environmental Zones used to present the results of CS2000 are shown in Figure 2.2. Although it is difficult to find simple names to describe them, as this figure shows they cover the range of environmental conditions that we find in the UK from the lowlands of the south and east, through to the uplands and mountains of the north and west. Throughout this report, each Environmental Zone is referred to by its representative number.

Figure 2.2. Environmental Zones of Great Britain



The Zones are based on combinations of the underlying sampling units, or land classes, used for the CS2000 field survey. Presentation of the results of CS2000 using these Zones is helpful for identifying some of the key geographical contrasts within the UK. However, it is important to note that the information that was collected by the field survey is stored in a very disaggregated way so that different types of report can be made.

Table 2.1. List and brief definitions of the BAP Broad Habitats included in the Countryside Survey 2000 (excluding marine habitats)

1. Broadleaved, mixed & yew woodlands	Dominated by mature trees > 5 m high. Distinct canopy, cover > 20%. Includes native broadleaved trees, non-native broadleaved trees, and yew trees (> 20% of the total tree cover). Scrub (< 5 m high) included if cover of woody species > 30%.
2. Coniferous woodland	Dominated by mature trees > 5 m. Distinct canopy, cover > 20%. Includes native conifers (Scots pine but not yew) and non-native conifers (e.g. larch and Sitka spruce) where cover > 80%. Recently felled if intention to return to Coniferous
3. Boundary & linear features	Diverse range of linearly landscape features. Occur separately or in combinations forming multi-element boundaries.
4. Arable & horticultural	All arable crops, orchards, market gardening and commercial flower growing. Freshly ploughed & fallow areas, short-term set-aside and annual grass leys included.
5. Improved grassland	Fertile soils. Few fast-growing species e.g. rye-grass and white clover. Used for grazing and silage, also managed for recreational purposes.
6. Neutral grassland	Soils not very acid or alkaline. No calcifuge or calcicole plants. May be hay meadows, pastures or silage. Less fertile and more herbs & grasses than improved grassland. Rye grass < 25% cover
7. Calcareous Grassland	Dominated by grasses and herbs on shallow, well-drained, alkaline soils, from weathering of chalk, limestone or other types of base-rich rock. Characteristically include a range of calcicoles or 'lime-loving' plants
8. Acid Grassland	Dominated by grasses and herbs on a lime-deficient soils derived from acidic bedrock or from superficial deposits such as sands and gravels. Characteristically include a range of calcifuge or 'lime-avoiding' plants.
9. Bracken	Stands of vegetation greater than 0.25 ha in extent which are dominated by a continuous canopy cover (>95% cover) of bracken at the height of the growing season.
10. Dwarf Shrub Heath	Comprises of vegetation with > 25% cover of plant species from the heath family or dwarf gorse species. It generally occurs on well-drained, nutrient-poor, acid soils.
11. Fen, Marsh and Swamp	Permanently, seasonally or periodically waterlogged ground as a result of ground water or surface run-off. Peat, peaty soils or mineral soils. Wide range of wetland vegetation, fens, flushes, marshy grasslands, rush-pastures, swamps and reedbeds.
12. Bog	Wetlands, peat-forming vegetation, receives mineral nutrients from rain not ground water. Acid tolerant plants where not modified by surface drying/aeration or heavy grazing. Purple moor-grass/hare's-tail cotton-grass can dominate on modified bogs.
13. Standing Waters and Canals	Includes lakes, meres and pools and man-made water bodies e.g. reservoirs, canals, ponds, gravel pits and water-filled ditches. Aquatic vegetation (free-floating or rooted in the sediments), and vegetation found in the shallower water of the margins.
14. Rivers and Streams	Includes rivers and streams from bank top to bank top; where there are no distinctive banks or banks are never overtopped, includes the extent of the mean annual flood, and the channel that may support aquatic vegetation and water fringe vegetation.
15. Montane habitats	Exclusively above the former natural tree-line on mountains. Prostrate dwarf shrub heath, snow-bed communities, sedge and rush heaths, and moss heaths. Species characteristic of the arctic and alpine regions and the vegetation is often 'wind-clipped' or prostrate.
16. Inland Rock	Habitat types that occur on both natural and artificial exposed rock surfaces, such as inland cliffs, caves, screes and limestone pavements, as well as various forms of excavations and waste tips, such as quarries and quarry waste.
17. Built-up areas and Gardens	Covers urban and rural settlements, farm buildings, caravan parks and other man-made built structures such as industrial estates, retail parks, waste and derelict ground, urban parkland and urban transport infrastructure. Includes domestic gardens and allotments.
Coastal habitats	Supralittoral rock; Supralittoral sediment; Littoral sediment coastal Broad Habitats were included in the CS2000 field surveys but the data collected are not considered to be representative of the UK resource

## Broad Habitats

The habitats of Britain were first characterised by ecologists in the early part of the 20th Century. They recognised that there are major vegetation types that have developed in response to climate, soil, natural succession and people's activities. This work has been the foundation of more refined vegetation classifications, which have been used to shape many conservation initiatives. For the purposes of the *UK Biodiversity Action Plan* (UK BSG, 1995), a new system of Broad Habitat categories has been developed which together account for all the terrestrial, freshwater and marine ecosystems of the UK. The list of the BAP Broad Habitats used in CS2000 is given in Table 2.1.

## Aggregate Vegetation Classes of the Countryside Vegetation System

Following a detailed analysis of the vegetation data collected during Countryside Survey 1990, a classification of the vegetation types commonly found in the countryside has been developed and published as the Countryside Vegetation System (Bunce et al., 1999). The system is based on a statistical analysis of the botanical records, which has created a hierarchical classification of the different vegetation types found in the sample plots.

The classification of vegetation is made on the basis of the species composition of the sample plots. At the highest level, eight 'aggregate' vegetation types are defined, describing the groupings of plants commonly found in the countryside. The classes are not equivalent to the BAP Broad Habitats. The latter are more general in character and describe larger tracts of land that may contain a range of vegetation communities. For example, an area of the *Broadleaved Woodland* Broad Habitat could contain dense, well-shaded woodland vegetation plus patches of tall grass and herbs along ride edges and infertile grassland in glades. The eight major vegetation types used by CS2000 are described in Table 2.2. They are useful because they allow comparisons within and across Broad Habitats and give us an overview of vegetation found at the national scale.

Table 2.2. List and summary description of the eight aggregate vegetation classes (AVC) of the Countryside Vegetation System (CVS).

AVC	Description
1. Crops and weeds	Weedy communities of cultivated and disturbed ground, including species-poor arable and horticultural crops.
2. Tall grass and herb	Less intensively managed tall herbaceous vegetation typical of field edges, roadside verges, streamsides and hedge bottoms.
3. Fertile grassland	Improved or semi-improved grassland. Often intensively managed agricultural swards with moderate to high abundance of perennial rye-grass.
4. Infertile grassland	Less-productive, unimproved and often species-rich grasslands in a wide range of wet to dry and acid to basic situations.
5. Lowland wooded	Vegetation dominated by shrubs and trees in neutral or basic situations, generally in lowland Britain. Includes many hedgerows.
6. Upland wooded	Vegetation of broadleaved and conifer woodland often in more acidic situations, generally in upland Britain.
7. Moorland grass mosaics	Extensive, often unenclosed and sheep grazed hill pastures throughout Britain.
8. Heath and bog	Vegetation dominated by heathers. Includes drier heaths as well as bog. Mostly in the uplands.

### Major soil groups

To enable GB wide comparisons, a unified classification of GB soils was derived by CEH and MLRUI. This classification based on a comprehensive soil classification system in which classes are differentiated by characteristics that can be evaluated in the field or inferred with reasonable assurance from field examination, either by comparison with “bench-mark” soils or by reference to geological data (Avery, 1980). The system is hierarchical, with classes termed as major soil groups, soil groups, soil sub-groups and soil series defined at four successive categorical levels. A list and brief description of the major soil groups used to examine soil relationships in this project are presented in Table 2.3.

Table 2.3. List and brief descriptions of the major soil groups used to classify the Countryside Survey X-plots from Avery, 1980.

Major Soil Group	Description
1. Terrestrial raw soils	Mineral soils - no diagnostic horizons or disturbed fragments of horizons, unless buried beneath a recent deposit more than 30 cm thick
2. Raw gley soils	Gleyed mineral soils with no diagnostic surface horizon and/or unripened (soft and muddy) at 20 cm or less
3. Lithomorphic	Distinct, humose or peaty topsoil and no diagnostic subsurface horizon. normally with bedrock or little altered unconsolidated material within 30 cm depth
4. Pelosols	Slowly permeable non-alluvial clayey soils with a distinct topsoil, weathered or argillic B horizon and no non-calcareous gleyed subsurface horizon < 40 cm
5. Brown soils	Other mineral soils with a weathered or argillic B horizon and no gleyed sub-surface horizon < 40 cm depth
6. Podzolic	Mineral soils with a podzolic B horizon (Bs, Bh and/or thin ironpan)
7. Surface-water gley	Non-alluvial, non podzolic soils with non-calcareous gleyed subsurface horizon (slowly permeable); gleyed E and/or B horizons below a humose or peaty topsoil and little/ no gleying in underlying horizons or both
8. Ground-water gley	Other non-podzolic soils with a distinct humose or peaty topsoil and gleyed subsurface horizon < 40 cm depth
9. Man-made soils	Other mineral soils with a thick man-made A horizon and/or a disturbed subsurface layer
10. Peat (organic )	> 40 cm of organic material within the upper 80 cm, or more, than 30 cm of organic material resting on bedrock or extremely stony material

### 2.4 Soil monitoring as a component of Countryside Surveys

As part of the 1978 survey, annotated soil descriptions and soil samples were taken from five, fixed vegetation plots within each square, giving a total of 1256 vegetation plots sampled on this occasion (Figure 2.3). Details of slope, aspect, vegetation and altitude were also recorded while soil pH and organic matter content were measured on each soil sample. As part of the 1990 Countryside Survey, SSLRC and MLURI mapped the soils in each 1 km<sup>2</sup> according to both the English & Wales soil series and the Scottish soil series using existing Soil Survey maps and limited field surveillance.

As outlined previously, the Countryside Survey of Great Britain was suggested by the RCEP (1996) as an existing and suitable baseline scheme for an assessment of soil

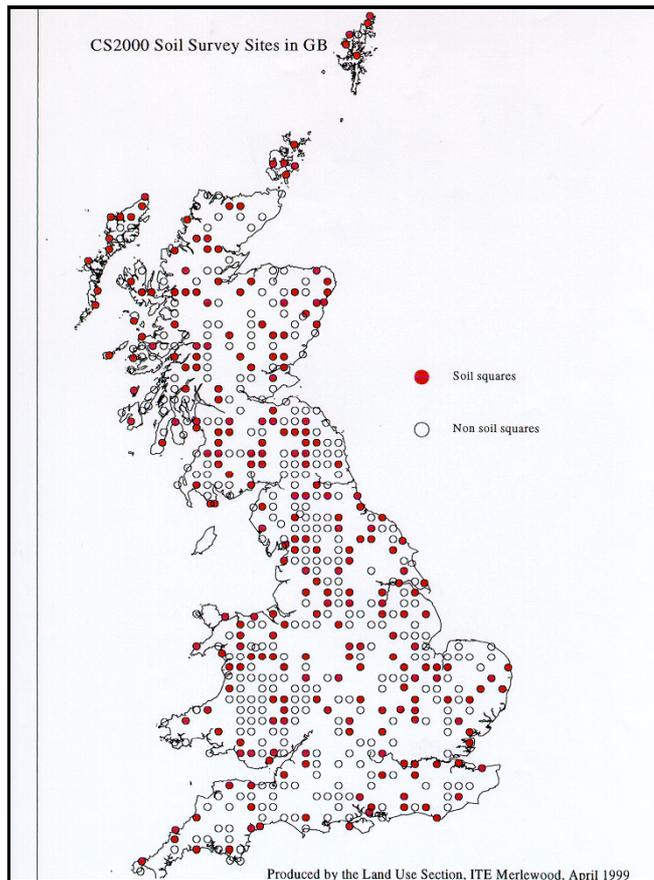
quality that included measurements of certain chemical and physical attributes, specific pollutants, and some biological attributes and included all major soil types and a representative range of land uses. The major issues were the production of baseline data as context for local, regional and national information and the development of suitable, effective strategies and policies relating to soil protection.

A nationwide survey was undertaken as a component of Countryside Survey 2000, to establish a framework for comprehensive baseline datasets that would directly link soil properties (chemical and biological) with their environment e.g. land use, vegetation, geographical location, climatic zone, etc. This project was titled “Monitoring and Assessing Soil Quality” (MASQ) and commenced early in 1998, prior to the start of the CS2000 field season, and was completed in 2001.

The following sections detail the process by which these data were obtained from the field sampling to the production of summary statistics. The focus of the project was to provide sufficiently detailed datasets from two dominant soil biological groups (soil invertebrate and microbial communities), soil chemical properties known to influence soil biology and be characteristic of GB soils; pH, organic matter content and two groups of chemical pollutants that could be widespread in the environment (heavy metals and persistent organic pollutants).

Statistics for soil properties can be produced at a range of scales and for different localities. However, careful consideration should be given to the significance of such results. For example, for soils where organic matter is typically low, most results from CS2000 were obtained from England and Wales while for the more organic soils, most results were obtained from Scotland. Results from agricultural systems (crops and weeds, tall grass and herbs, fertile grassland) were mainly from England and Wales while results from semi-natural upland systems were mainly from Scotland. However, England and Wales contain significant areas of soils with high organic matter while Scotland has large agricultural areas. Although statistics can be produced at these higher levels of resolution, they must be considered in the national context to gain a representative assessment of their characteristics. Greater resolution at these lower levels may be achieved with the amalgamation of groupings or by increasing the sample size in future surveys. The potential for amalgamation is discussed where relevant.

Figure 2.3. Locations of original 1978 soil sampling plots shown as “soil squares” from Countryside Survey 2000



## 2.5 Access to Countryside Survey data

At the most general level, national and regional statistics from the Countryside Surveys can be downloaded from web sites. This service provides tables, graphical material and map data for the UK, by country and by Environmental Zone, for each of the main themes covered by this report. Technical reports and other documents relating to these will be provided as they become available. Detailed data from CS2000 will also be accessible through the Countryside Information System (CIS). This provides the capability to map field and satellite land cover data, at the 1km square level, and to derive statistics for any geographical area defined by the user. These include Environmental Zones, landscape character areas, administrative regions, and designated areas such as National Parks, Areas of Outstanding Natural Beauty, Sites of Special Scientific Interest, and Environmentally Sensitive Areas. CIS also enables CS2000 data to be combined with other geographic data that can be summarised at the 1km x 1km grid square level. Datasets already available include information from Ordnance Survey, such as altitude, slope, roads, rivers and other specially constructed data sets relating, for example, to soils and climate.

### 3. SCOPING STUDY

A wide range of metals, metalloid and organic compounds are considered significant environmental pollutants. A select set was identified by the Environment Agency for relevance to the Agency's priorities in the UK and as potential substances for analyses in the CS2000 Soils Module project (MASQ). Since it was not feasible to analyse all these compounds due to limits on cost and sample size, a Scoping Study was carried out to produce a priority list of compounds based on national relevance, reliance of analytical methods (including quality assurance and quality control methods), sample storage and relative costs. A copy of the Scoping Study is available in the Project Record.

#### 3.1 Metals/semi metals/metalloids

Table 3.1. lists the metals, semi-metals and metalloids produced at the outset of the study.

Table 3.1. List of metals, semi-metals and metalloids addressed in the MASQ Scoping Study

Cadmium	Copper	Nickel	Vanadium
Lead	Mercury	Arsenic	Beryllium
Zinc	Antimony	Boron	Chromium
Manganese	Selenium	Sulphur	Cyanide

An extensive literature search was carried out using BIDS, BIOSIS, the WWW and the CEH library service to prioritise and group substances for analyses from CS2000 soils. The literature was examined for data on emissions/sources, toxicity (human, plant, microbial and animal) and current recommendations/legislation (soil protection, human exposure, critical loads and land remediation) for each of the listed substances. It was recognised that certain substances, potentially significant pollutants, may have been excluded, principally due to a lack of appropriate risk assessments and/or adequate toxicological data. Analytical techniques and sample requirements were also used to further prioritise these substances.

Inductively Coupled Plasma-Optical Emission Spectrometers (ICP-OES) and Inductively Coupled Plasma-Mass Spectrometry (ICP-MS) can be regarded as complementary methods for analyses and each has its own advantages and disadvantages. ICP-MS offers the potential for better sensitivity with lower detection limits. However, with ICP-MS there are matrix problems associated with the analysis of acid digests of solid samples, as there is a limit on the dissolved salt content. This means that the working detection limit may not be as good as the analysis by ICP-OES, due to the requirement to work with lower solid to liquid ratios. ICP-MS was available if analysis revealed concentrations below detection by ICP-OES.

The analysis of total metal or metalloid contents ( $\text{mg kg}^{-1}$ ) will remain the most reliable and meaningful environmental assessment until there are recognised standard methods for determining the available reactive content of metals or metalloids and the bioavailability of these "available" components is fully understood (de Vries and Bakker, 1998). In support of this, almost all current legislation and recommendations are based on total concentrations of metals and metalloids in soils since total

concentrations are relatively easy to determine and incorporate into soil quality standards, compared to available concentrations. Working in the precautionary principle, totals at least identify soils with potential risks: the first stage of a risk assessment is to identify presence of a chemical and the next is to identify a compound's behaviour (de Haan, 1996). Data from McGrath and Loveland (1992) indicate that relatively good relationships exist between total and available concentrations for several metals and that these relationships, once more fully understood, may prove useful in predicting available metal contents in soils.

From this process, six metals were identified for analyses by ICP-OES from aqua-regia digests (Pb, Zn, Cu, Cd, Ni, Cr) plus Hg by cold vapour AAS. Provisional analytical costs were provided for all options. Total metal concentrations were identified are the most appropriate given current status of standard analytical methods. In discussion with the EA, Vanadium and Arsenic were added as compounds of increasing pollution interest and it was agreed at CEH would carry out the analyses of process, six metals were identified for analyses by ICP-OES on all CS2000 soil samples, where sufficient sample was available, and that the EA Llanelli laboratories would carry out the analyses of Hg and As by ICP-MS, as well as a sub-set (ca 100) of the aqua regia digests for Pb, Zn, Cu, Cd, Ni, Cr by ICP-MS and all samples where the concentrations were too low for ICP-OES analyses. Full details of these analyses are provided in the heavy metals in soil section.

### **3.2 Organic compounds**

Existing analytical capabilities in both CEH and the EA were examined and assessed in relation to quantitatively analysing soil samples without undue time being devoted to method development. Approaching the problem in this way maximised the number of samples that could be analysed and minimised (but did not eliminate) method development. Select compounds were identified from the initial "wish-list" (Table 3.2) as meeting the overall aims of the project.

Analytical methods and their costs were then outlined. Consideration was then given to the various factors that need to be taken into account in selecting which of the 1,000 or so soil samples should be analysed, and to the scientific strategy of the organic analyses. Finally, because the resources will not permit all available soil samples to be analysed, a work plan was outlined to form the basis for a discussion with the Agency. This included proposals for a modest amount of method development that might considerably increase either the number of squares analysed or the number of determinands. Full details of the method development and organic compounds selected after this process are provided in the organic pollutant in soil section.

Table 3.2. Full wish-list of organic compounds

Polycyclic Aromatic Hydrocarbons (PAHs)			
CAS No.	PAH	CAS No.	PAH
120-12-7	Anthracene	129-00-0	pyrene
86-73-7	Fluorene	218-01-9	chrysene
91-20-3	Naphthalene	56-55-3	benzo[a]anthracene
85-01-8	Phenanthrene	205-99-2	benzo[b]fluoranthene
191-24-2	benzo[ghi]perylene	207-08-9	benzo[k]fluoranthene
208-96-8	Acenaphthylene	50-32-8	benzo[a]pyrene
83-32-9	Acenaphthene	53-70-3	dibenz[ah]anthracene
206-44-0	Fluoranthene	193-39-5	indeno[1,2,3-cd]pyrene
	Benzo[e]pyrene		Benzo[j]fluoranthene
Organic pesticides			
<u>Drins, including:</u> Aldrin Dieldrin Endrin	<u>OCs, including:</u> Chlordane DDT (and products?) HCH, (total/gamma)	<u>Azoles</u>	Triazines
Aromatic halocarbons	Chlorinated phenols	Organometallics	Other organics:
Chlorobenzenes Chlorotoluenes	Chlorophenols (no4tpenta) Pentachlorophenol	Organolead + Organotin compounds	Chlordecane, Hexabromobiphenyl, Mirex, Toxaphene, Acetone, Phenol, Oil/Fuel hydrocarbons
Polychlorinated Biphenyls (PCBs)			
IUPAC NO.	Structure	TEF	
non-ortho			
77	3,3',4,4'-tetrachlorobiphenyl	0.0005	
126	3,3',4,4',5-pentachlorobiphenyl	0.1	
169	3,3',4,4',5,5'-hexachlorobiphenyl	0.01	
mono-ortho			
105	2,3,3',4,4'-pentachlorobiphenyl	0.0001	
114	2,3,4,4',5-pentachlorobiphenyl	0.0005	
118	2,3',4,4',5-pentachlorobiphenyl	0.0001	
123	2',3,4,4',5-pentachlorobiphenyl	0.0001	
156	2',3,3',4,4',5-hexachlorobiphenyl	0.0005	
157	2,3,3',4,4',5-hexachlorobiphenyl	0.0005	
167	2,3',4,4',5,5'-hexachlorobiphenyl	0.00001	
189	2',3,3',4,4',5,5'-heptachlorobiphenyl	0.00001	
di-ortho			
170	2,2',3,3',4,4',5-heptachlorobiphenyl	0.0001	
180	2,2',3,4,4',5,5'-heptachlorobiphenyl	0.00001	
Dioxins			
Congener	I-TEF	Congener	I-TEF
2,3,7,8-TCDD	1	1,2,3,7,8-PeCDF	0.05
1,2,3,7,8-PeCDD	0.5	1,2,3,4,7,8-HxCDF	0.1
1,2,3,4,7,8-HxCDD	0.1	1,2,3,7,8,9-HxCDF	0.1
1,2,3,7,8,9-HxCDD	0.1	1,2,3,6,7,8-HxCDF	0.1
1,2,3,6,7,8-HxCDD	0.1	2,3,4,6,7,8-HxCDF	0.1
1,2,3,4,6,7,8-HpCDD	0.01	1,2,3,4,6,7,8-HpCDF	0.01
OCDD	0.001	1,2,3,4,7,8,9-HpCDF	0.01
2,3,7,8-TCDF	0.1	OCDF	0.001
2,3,4,7,8-PeCDF	0.5		

(Where: T=tetrachloro; Pe=pentachloro; Hx=hexachloro; Hp=heptachloro; and O=octachloro)

## **4. COUNTRYSIDE SURVEY SOIL SAMPLING AND INITIAL PROCESSING**

### **4.1 Ecological Survey of Great Britain in 1978**

The objective of this survey was to record land cover, land use, landscape features and vegetation in each 1 km square to obtain an overall ecological in a consistent manner. The collection of data was designed to take, on average, a single day per 1 kilometre square. Full details of the data collection procedures are available in the unpublished handbook for field surveyors. Most of the procedures were the same as those used in CS2000, unless otherwise stated. Five main plots (“X-plots”) were located within each 1 km square. These were located using random co-ordinates in five segments of the square. Each plot was characterised for slope, aspect, land use, vegetation, boundaries, animals and topography. Soil pits were then prepared in each plot. Information was then recorded on a pre-prepared record sheet. The following details the collection of soils data and samples.

#### **4.1.1 Location of the soil pit**

The pit was located at the centre of the quadrat in the quarter containing face number one on the wooden square on top of the centre pole. If the position fell on bare rock, a wall, a fence, a hedgerow or hedgebank, or a road, the pit was then sited at the end of the string between faces 1 and 2 of the wooden square. If still on an “obstruction” the pit was transferred to the ends of the strings between faces 2 and 3, then 3 and 4, then 4 and 1. If all the above sites fell on obstructions – no pit was dug. When the pit position was moved, the new position was recorded and the reason for moving detailed on the record sheet. The exact orientation depended on the site type, time of day etc. The pit was sited to allow maximum light to fall on one face for the horizon description. The depth varied with soil group and type but was carried out to a minimum of C or R horizon, or a depth of 75 cm, whichever was reached first. Once completed, the horizon face was cleaned up and the constituent horizons were identified and recorded with associated notes. Completion of the data sheet included the following; stratum and plot number, horizon thickness, moisture status, colour (using a Munsell Chart), mottles, texture, structure, stones, roots, carbonates, earthworms, iron pan, soil group (from list on sheet), “parent” material, solid geology, additional comments.

#### **4.1.2 Soil sampling**

A small sample (c. 200g) was collected from the surface horizon where the O<sub>1</sub> (fresh litter) and/or O<sub>f</sub> (partially decomposed litter) was less than 5 cm thick. Where these were > 5cm, the sample was taken from the next layer below. If the surface horizon was less than 5 cm thick, the soil sample was collected from the upper 5 cm. These samples were stored in bags and returned to the either ITE Bangor or ITE Merlewood for soil pH and LOI analyses.

#### **4.1.3 Chemical analyses**

Soil pH was determined on a homogenised sub-sample of the fresh soil. Loss-on-ignition was determined using the same methodology as CS2000 (see protocol) with a furnace combustion temperature of 550°C.

## 4.2 Countryside Survey 2000

### 4.2.1 Preparation prior to the field survey

Before the field survey commenced, all the necessary equipment and protocols were prepared at CEH Merlewood by 3 staff of the Soil Ecology Section. A field sampling kit was prepared for each survey team that consisted of sampling items additional to those already in the CS2000 field kit. Each team leader was issued with complete packs for all squares to be sampled in their area. Each pack contained stamped addressed envelopes to ITE Merlewood for each X plot which also contained two white cores plus plastic stoppers in labelled plastic bags, one each for faunal and microbial samples, and a black plastic core for the soil chemistry sample.

*Laboratory space:* A large store at Merlewood was renovated to provide the necessary space for processing, extracting and storing the large number of soil samples expected from CS2000.

*Soil faunal extraction equipment:* Tullgren funnels (72 in 6 x 12 banks) were housed in the dedicated laboratory and used in the dry extraction of soil mesofauna. Standard dry extraction protocols were tested in May 1998 and modified to ensure optimum extraction efficiency from each core.

*Soil Microbial Cores:* A processing and storage protocol was developed along with the soil faunal protocols, since these two cores should arrive at the same time. Freezers were purchased for the storage of CS2000 soil cores for microbial assessments.

### 4.2.2 Protocols and training

The field surveyors were all trained, at the CS2000 training course (May 1998), to collect soil samples for MASQ using protocols developed in the preceding months at CEH Merlewood. These protocols were tailored to be comparable with the 1978 soil samples and to enable soil samples for biological analyses to be returned to CEH Merlewood in a relatively short period of time e.g. by hand or first class post. The full protocols are available in the CS2000 Field Handbook. A summary of the sampling process is provided below.

### 4.2.3 Locating and sampling X-Plots

During 1998 and 1999, the surveyors collected soil samples from each of the “X-plots” first sampled in 1978 (where still in existence). Table 4.1. provides a description of the “X-plots”.

Table 4.1. Description of Countryside Survey X-plots

Code	Name	Other names	Where	Size	Number/square
X <sup>1</sup>	Large	Main	Random points in open polygons	200 m <sup>2</sup>	5

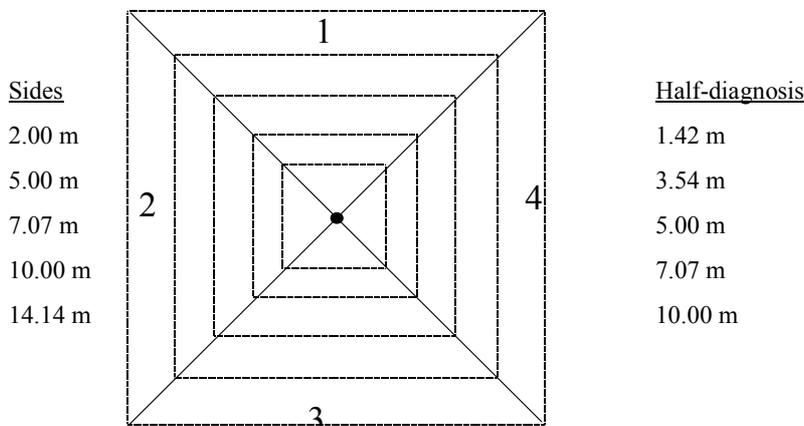
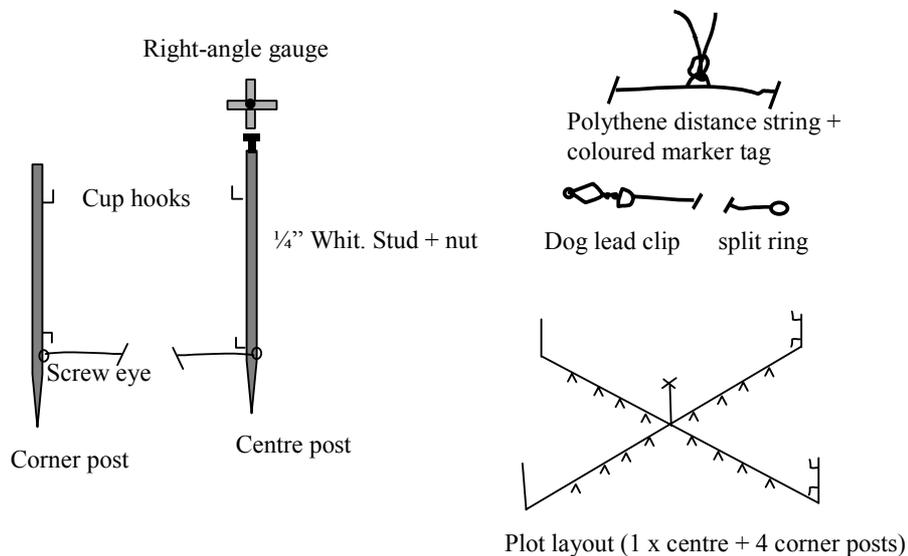
<sup>1</sup> first recorded in 1978

The X-plots were marked on maps in advance of each field visit and then located on the ground as accurately as possible using the position shown on the map and/or via the

markers placed in 1990 using a metal detector. There were some instances where the land use had changed so that a vegetation plot was no longer appropriate. Soil would not have been sampled from these.

Once the location was established, the plot was laid out using survey poles with the strings forming the diagonal of the square. The diagonals were orientated carefully at right angles and the plot was orientated with the strings on the North/South, East/West axes. Nested plots (Figure 4.1.) were then marked out using different coloured strings on the appropriate position of the diagonal. Detailed plant species data were recorded from the inner nested (4 m<sup>2</sup>) plot first, either by ticking the species names on the "top 200" list or by adding species names at the bottom of the recording form. The field surveyors then carried out the soil sampling.

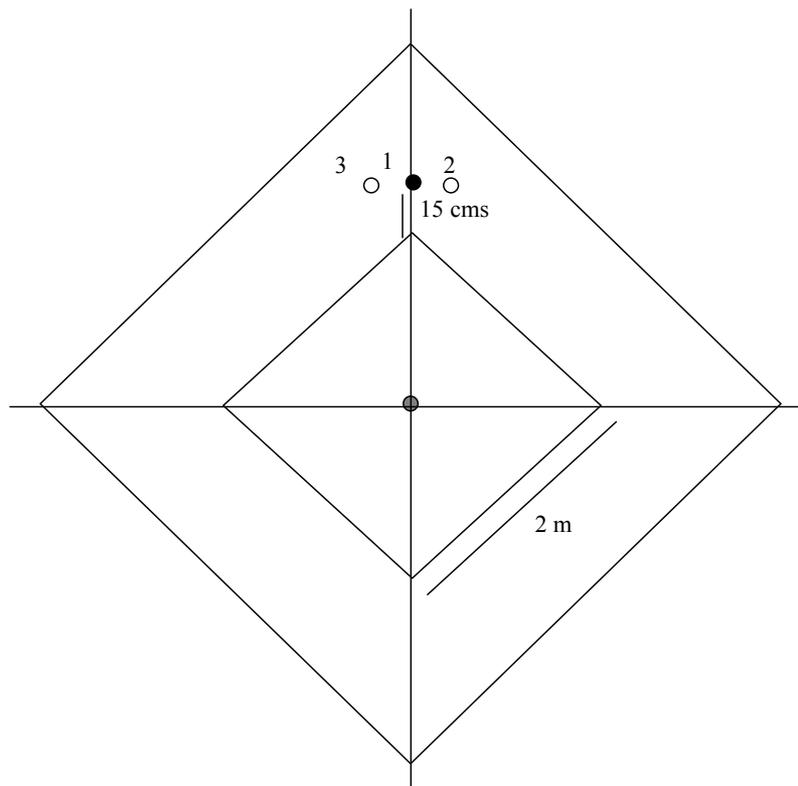
Figure 4.1. Sampling equipment, plot layout and design of X Plot.



#### 4.2.4 Sample collection

The soil sampling location, 15 cm to the N of the 2 x 2 m inner quadrat, was identified using measuring tapes and a ruler (Figure 4.2.). Three samples were taken, when ever possible, at this point. One, central, core was taken using a 15 cm deep x 8 cm diameter “**black core**”. Two smaller “**white core**” were taken (8 cm deep x 4 cm diameter) at 10 cm to either side of this black core. All samples were taken using a knife to cut around the edge of the core, where vegetation prevented adequate coring. A mallet and plate were used to knock the plastic cores into the soil and a trowel to dig the cores out. The smaller white cores were capped. These cores were for soil biological and organics analyses. These were posted (1<sup>st</sup> class) or returned to CEH Merlewood by hand. The smaller size allowed these white cores to fit into rural post-boxes, in the majority of cases. The larger central black cores (for all other chemical analyses) were returned to Merlewood by the regional field co-ordinators and/or field surveyors.

Figure 4.2. Location of MASQ soil sampling in the centre of the X-plot. 1 = black core location, 2 + 3 = white core locations. Adapted from the CS2000 field handbook



#### 4.2.5 Initial sample processing

A protocol was developed and followed by all staff for the initial processing of all soil cores as they arrive at Merlewood. This was developed and tested in May 1998. A working practice was established that used two recording systems. The first system was used to log-in all the soil biota samples, monitor the processing of these samples and

record storage details. The second system was used to log in all the soil chemistry cores and again monitor their processing progress to storage. Record sheets were up-dated every day by the staff processing the samples and the data were transferred to Excel spreadsheets at regular intervals.

As cores arrived at CEH Merlewood, they were collected from reception by the person responsible for sample processing. All samples were taken to the MASQ sample processing area and all samples entered into the log in sheets. The samples were then processed appropriately. Where two white cores were present for an X-plot, one was immediately placed in the freezer in an appropriate location; freezer trays were organised by 1 km<sup>2</sup> number and each tray contained plastic bags marked with a relevant 1 km<sup>2</sup> number. These cores were then processed in date order.

The remaining white core was then used for a 5-day dry extraction in Tullgren apparatus. The resulting invertebrate sample in 70% ethanol was stored in fire-proof cupboards, in 1 km<sup>2</sup> square organised storage boxes, until identification was initiated. The black soil cores were immediately weighed and then processed to allow wet and dry pH to be assessed, loss-on-ignition to be determined and the remaining sample to be stored for future chemical analyses.

## **5. DATA MANAGEMENT AND STRUCTURE OF MASQ DATABASE**

### **5.1 Introduction**

The development and management of an appropriate, user-friendly, database was a key stage in the MASQ project to ensure the long-term storage of all data and meta data and the provision of all datasets in formats suitable for investigative statistical and spatial analyses using standard protocols for dataset population and metadata production. The process ensured that all MASQ datasets were fully compatible with each other and also with other Countryside Survey data taken from the 1 km squares. Since all data are geo-referenced, comparative analyses could be carried out with other geo-referenced data. Meta-data describing the data collected by the MASQ survey, data products and the data stored within the integrated database are integral parts of the MASQ database. Each dataset and related metadata file contains a WWW-searchable catalogue describing the data and data products.

The specific requirements of the MASQ data management system were:

- To ensure safe long-term storage of meta-data as well as analytical results.
- To provide a mechanism to link-up all MASQ datasets with each other and with other Countryside Survey data (and other national datasets, in future)
- To provide a system to produce amalgamated datasets in formats for further analyses
- To provide data in formats suitable for use in CIS

To meet these objectives, a common data management structure was devised in accordance with The Countryside Survey 2000 Integrated Data System (CIDS). This section of the report describes the process of establishing the MASQ database within CIDS.

#### **Countryside Survey 2000 Integrated Data System**

The Countryside Survey 2000 Integrated Data System (CIDS) is an ORACLE based data management system that enables secure storage and access to datasets via different toolsets, including data entry facilities, analytical packages and Web based data catalogues. The CIDS is maintained by CEH Computer support Staff. Data security is ensured via off-site backup and internet fire wall facilities. By storing MASQ data within the CIDS, safe storage of the data, links to CS2000 and ease of data access for current and future analyses is ensured.

#### **The Countryside Information System**

The Countryside Information System (CIS) is a Microsoft Windows-based program developed to give policy advisers, planners and researchers easy access to spatial information about the British countryside. CIS contains a wide range of environmental data, including landscape features, vegetation habitats and topography for each 1 km square of Great Britain. As part of the CIS, the Environmental Catalogue provides information that enables users to identify and obtain available datasets and a forum for data suppliers to promote their datasets.

## 5.2 Methods

### **Integration of MASQ data into CIDS**

Integration of the MASQ data with other Countryside Survey data sets was enabled through the data management activities of CS2000 Module 13. Module 13 is the Scientific Support and Information Management service for the CS2000 Programme. A key objective of Module 13 was to design, implement and manage a single overarching database located at CEH Merlewood to integrate and link the data collected in each of the CS2000 Modules, and earlier surveys (e.g. Countryside Survey 1990 and ITE's National Ecological Survey 1978).

One of the activities of Module 13 was to develop a data dissemination strategy for the programme as a whole (reference). It is intended that the MASQ programme followed this strategy. The key elements of this strategy are

- Dissemination of results tables via reports and web-based data tables and of data descriptions via web based data catalogues e.g. The CS2000 data catalogue (Level 1 data)
- Dissemination of Land Class summary data via the Countryside Information system (CIS) and it's associated websites (Level 2 data)
- Access to bespoke analysis of the base data sets via the CS2000 Scientific Support Service (Level 3 data).

### **Development of MASQ data management system**

The following packages were used to establish the MASQ data management system. All packages are held under license, maintained and supported by CEH Computer Support at CEH Merlewood.

ORACLE Developer 6i under NERC Licence

The SAS system version 8

Microsoft Excel 2000 (Microsoft Office 2000) licensed through Microsoft Select

Minitab Version 13.2 for Windows

Statistica for Windows

The process of setting up the MASQ data management system is outlined below.

#### ***Stage 1. Development of MASQ database structure and links to the Countryside Information System (CIS)***

- The structure of the MASQ database and data connections to CIS were established with the Countryside Survey Database Manager
- A development plan was agreed within CS2000 Module 13
- Datasets and metadata from each of the analytical groups were identified
- Links to the Countryside Survey data were identified Broad habitat (BH), Environmental Zone (EZ), Major Soil Group (SOIL), Aggregate Vegetation Class (AVC) and 1998 Land Class (LC)
- Links to Countryside Survey 2000 Integrated Data System (CIDS) were established through ORACLE Query Builder

### *Stage 2. Population of the ORACLE database*

- MASQ data were validated by each of the analytical groups (soil pH, organic matter, heavy metals, invertebrates, microbes, and organics)
- The data was sent electronically to the MASQ data manager
- The datasets and metadata (data descriptions) were inserted into ORACLE storage database by the MASQ database manager through ORACLE Schema Builder and ORACLE Query Builder

### *Stage 3. Creation of data views in ORACLE for analyses*

- Views were created by connecting MASQ data with Countryside Survey data (e.g. Broad Habitat) in ORACLE Query builder (see Project Record: MASQ Data management protocol)
- A SAS library (which connects directly into the Oracle database) was created for each MASQ user with access to connected views
- All files entered onto the MASQ ORACLE database can be accessed through SAS (currently SAS System for Windows Version 8), a statistical analysis tool which can analyse large databases, produce summary tables systematically and store summary results as HTML (web page of results) if required. Data and results can easily be exported for further analysis and into presentation programmes
- Two views per data set were created, one with links to EZ and BH and the other with links to EZ, SOIL, LC and AVC.

### *Stage 4. Preparation of summary tables*

- Summary statistics (number of non-missing values, mean, median, maximum, minimum, standard deviation) were produced by each group in SAS or Excel
- Summary tables were compiled by each group in Excel for incorporation into the Final Report
- Between group data checks were carried out for data validation (see MASQ ORACLE Data Validation Protocol)

### *Stage 5. Further analyses of selected data sets*

- Further connections were requested for more detailed analyses of the data.
- New views were created in ORACLE Query Builder connecting loss on ignition and pH with data from other groups

## **5.3 Production of summary statistics**

The data manager established the procedures for producing the required data views from ORACLE via SAS. MASQ project staff were then shown how to produce summary tables for their component. SAS created an HTML version (see Table 5.1) of summary statistics that was pasted directly into Excel (see Project Record: MASQ Data Analysis Protocol). This Excel sheet was reformatted and sorted. Minitab was used to produce Normal distribution plots for the data and box and whisker plots were created using Statistica 98 Edition. These results are presented within the relevant sections for each group.

### **Integrative analyses**

Selected MASQ datasets were connected with soil organic matter content and soil pH. Further links can be made for future analyses using this approach.

Table 5.1 An example of a SAS HTML summary output.

	Number of non-missing values
	<b>EXAMPLE</b>
<b>SOIL_TYPE</b>	
<b>3</b>	90
<b>4</b>	24
<b>5</b>	335
<b>6</b>	148
<b>7</b>	187
<b>8</b>	117
<b>10</b>	122
<b>Total</b>	1023

### **5.4 Maintenance of the MASQ data management system**

The MASQ database manager maintains day-to-day running to the MASQ database and holds sole write access to the MASQ ORACLE database. All data additions and/or changes are carried out by this person on request from project staff responsible for key components. This person also liases with CS2000 Module 13 for any further data requirements on behalf of MASQ project staff.

Table 5.2. MASQ catalogue entry of data sets, metadata and views.

**Monitoring and Assessment of Soil Quality (MASQ)**

**Database Short Name (Schema): DB\_MASQ**

**User ID masq has READ ONLY access to the following tables**

Table Name	Description
MAJOR_SOIL_DESC	Using Classification of soils in England and Wales (Avery, 1980)
MASQ_ALL_MICRO_BH_VIEW	View contains selected MASQ microbiology data connected to selected CS2000 data
MASQ_ALL_MICRO_NO_BH_VIEW	View contains selected MASQ microbiology data connected to CS2000 data (including Environmental Zone, ITE Land Class, CVS Aggregate Vegetation Class and Soil Type)
MASQ_AS_HG_DATA	Author: Environment Agency Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: As and Hg (mg/kg) with limit of detection data
MASQ_AS_HG_EA_METADATA	Author: Environment Agency Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Explanation of column headings found in: MASQ_AS_HG_EA_DATA
MASQ_AS_HG_NO_BH_VIEW	View contains Environment Agency arsenic and mercury data connected to selected CS2000 data
MASQ_AS_HG_VIEW	View contains Environment Agency arsenic and mercury data connected to selected CS2000 data
MASQ_BIOLOGCOUNT_BH_VIEW	View contains MASQ_BIOLOGCOUNT_DATA connected to selected CS2000 data
MASQ_BIOLOGCOUNT_NO_BH_VIEW	View contains MASQ_BIOLOGCOUNT_DATA connected to selected CS2000 data
MASQ_BLACKCORE_BH_VIEW	View contains MASQ_BLACKCORE_INFO connected to selected CS2000 data
MASQ_BLACKCORE_INFO	Author: RE Creamer Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: soil cores sampled in 1998 Explanation of column headings found in: MASQ_BLACKCORE_METADATA
MASQ_BLACKCORE_METADATA	Author: RE Creamer Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> File contains explanation of column headings for: MASQ_BLACKCORE_DATA
MASQ_BLACKCORE_NO_BH_VIEW	View contains MASQ_BLACKCORE_INFO connected to CS2000 data (excluding BH)
MASQ_FAUNA_BH_VIEW	View contains MASQ_FAUNA_DATA connected to CS2000 EZ and BH data
MASQ_FAUNA_DATA	Author: Jacky Garnett Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: Total numbers of soil fauna identified by major invertebrate group. Explanation of column headings found in: MASQ_FAUNA_METADATA
MASQ_FAUNA_EXTRACTION_INFO	Author: Jacky Garnett Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: Dates and notes regarding arrival of samples and extraction start and stop dates. Explanation of column headings found in: MASQ_FAUNA_EXTRACTION_METADATA
MASQ_FAUNA_EXTRACTION_METADATA	Author: Jacky Garnett Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> File contains explanations of column headings for: MASQ_FAUNA_EXTRACTION_DATA
MASQ_FAUNA_METADATA	Author: Jacky garnett Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> File contains explanation of column headings found in: MASQ_FAUNA_DATA
MASQ_FAUNA_NO_BH_VIEW	View contains MASQ_FAUNA_DATA connected to CS2000 data (excluding BH)
MASQ_FAUNA_PH_BH_VIEW	View contains MASQ_FAUNA_DATA connected to selected MASQ_PHLOI_DATA and selected CS2000 data

Table 5.2. MASQ catalogue entry of data sets, metadata and views. *Cont.*

MASQ_FAUNA_PH_VIEW	View contains MASQ_FAUNA_DATA connected to selected MASQ_PHLOI DATA and CS2000 data
MASQ_GENERAL_SITE_BH_VIEW	View contains MASQ_GENERAL_SITE_INFO connected to selected CS2000 data
MASQ_GENERAL_SITE_INFO	Author: RE Creamer Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description:1998 Explanation of column headings can be found in: MASQ_GENERAL_SITE_METADATA
MASQ_GENERAL_SITE_METADATA	Author: RE Creamer Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> File contains explanation of column headings found in: MASQ_GENERAL_SITE_INFO
MASQ_GENERAL_SITE_NO_BH_VIEW	View contains MASQ_GENERAL_SITE_INFO connected to selected CS2000 data
MASQ_METALS_DATA	Author: Phil Rowland Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Total Cr, Cd, Cu, Ni, Pb, V and Zn in soil, determined on ball milled sample, digested in aqua-regia and analysed on the JY 38+ ICP-OES
MASQ_METALS_METADATA	Author: Environmental Chemistry Section Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> File contains explanation of column headings found in: MASQ_METALS_DATA
MASQ_METALS_NO_BH_VIEW	View contains MASQ_METALS_DATA connected to selected CS2000 data
MASQ_METALS_VIEW	View contains MASQ_METALS_DATA connected to selected CS2000 data
MASQ_METAL_PH_BH_VIEW	View contains MASQ_METAL_DATA connected to selected MASQ_PHLOI_DATA and CS2000 data
MASQ_METAL_PH_VIEW	View contains MASQ_METAL_DATA connected to selected MASQ_PHLOI_DATA and CS2000 data
MASQ_MICRO_BIOLOG_DATA	Author: Nisha Parekh Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: Determination of respiration of 95 different BIOLOG-GN substrates and a substrate free control by extracted soil microbial communities after 0, 4 and 7 days of incubation at 20oC. Samples collected as part of the MASQ project in 1998. Explanation of column headings can be found in: MASQ_MICRO_BIOLOG_METADATA
MASQ_MICRO_BIOLOG_METADATA	Author: Nisha Parekh Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Explanation of column heading for: MASQ_MICRO_BIOLOG_METADATA
MASQ_MICRO_COUNTS_BH_VIEW	View contains MASQ_MICRO_COUNTS_DATA connected to selected CS2000 data
MASQ_MICRO_COUNTS_DATA	Author: Nisha Parekh Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: Determination of number of colony forming units of bacteria per ml of soil extract solution and gram dry weight equivalent of soil after 4 and 7 days of incubation on 10% nutrient agar at 20oC. Also information on number of cells in dilution used for BIOLOG tests. Explanation of column headings found in: MASQ_MICRO_COUNTS_METADATA
MASQ_MICRO_COUNTS_METADATA	Author: Nisha Parekh Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Explanation of column headings for: MASQ_MICRO_COUNTS_DATA file
MASQ_MICRO_SIEVEMOIST_DATA	Author: Nisha Parekh Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: Dates and notes for thawing and sieving samples, moisture contents and dry weight equivalent calculations for each sample. Explanation of column headings found in: MASQ_MICRO_SIEVEMOIST_METADATA
MASQ_MICRO_SIEVEMOIST_METADATA	Author: Nisha Parekh Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Explanation of column headings: MASQ_MICRO_SIEVEMOIST_DATA file.
MASQ_MICRO_SUBSTRATE_DATA	Author: Dr. N Parekh Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: abundance, activity and functional diversity of heterotrophic soil bacterial communities with relation to land use, vegetation, soil type, soil physical characteristics and pollutant levels.
MASQ_MIC_COUNT_NO_BH_VIEW	View contains MASQ_MICRO_COUNT_DATA connected to selected CS2000 data

Table 5.2. MASQ catalogue entry of data sets, metadata and views. *Cont.*

MASQ_ORGANICS_DATA	Author: D Osborne Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: Organic compound concentration data. Analyses performed by CEH Monkswood. Explanation of column headings found in MASQ_ORGANICS_METADATA
MASQ_ORGANICS_METADATA	Author: D Osborn Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Explanation of headings found in: MASQ_ORGANICS_DATA
MASQ_ORGANICS_VIEW	View contains MASQ_ORGANICS_DATA connected to selected CS2000 data
MASQ_ORIBATID_FAMILY_DATA	Author: FM Monson Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Occurrence (presence = 1) of oribatid mite family in MASQ soil fauna cores collected in 1998. Explanation of column headings found in: MASQ_ORIBATID_FAMILY_METADATA
MASQ_ORIBATID_FAMILY_METADATA	Author: FM Monson Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Explanation of column headings for: MASQ_ORIBATID_FAMILY_DATA
MASQ_ORIBATID_FAM_BH_VIEW	View contains MASQ_ORIBATID_FAMILY_DATA connected to selected CS2000 data
MASQ_ORIBATID_SPECIES_DATA	Author: FM Monson Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Occurrence (presence = 1) of oribatid mite species in MASQ fauna soil cores collected 1998. Column heading explanations found in: MASQ_ORIBATID_SP_METADATA
MASQ_ORIBATID_SP_METADATA	Author: FM Monson Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Explanation of column headings found in MASQ_ORIBATID_SPECIES_DATA
MASQ_ORIBATID_SP_NO_BH_VIEW	View contains MASQ_ORIBATID_SPECIES_DATA connected to selected CS2000 data
MASQ_ORIBATID_SP_VIEW	View contains MASQ_ORIBATID_SPECIES_DATA connected to selected CS2000 data
MASQ_ORIBATID_TOTAL_DATA	Author: FM Monson Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data description: Total number of families and species of oribatid mites found in CS2000 soil core extractions.
MASQ_ORIBATID_T_BH_VIEW	View contains MASQ_ORIBATID_TOTAL_DATA connected to selected CS2000 data
MASQ_ORIBATID_T_VIEW	View contains MASQ_ORIBATID_TOTAL_DATA connected to selected CS2000 data
MASQ_ORIBAT_FAM_VIEW	View contains MASQ_ORIBATID_FAMILY_DATA connected to selected CS2000 data
MASQ_PHLOI_BH_VIEW	View contains MASQ_PHLOI_DATA connected to selected CS2000 data
MASQ_PHLOI_DATA	Author: RE Creamer Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: pH and loss on ignition data for MASQ soil cores collected in 1998. Column heading explanations found in: MASQ_PHLOI_METADATA
MASQ_PHLOI_METADATA	Author: RE Creamer Contact: <a href="mailto:j.garnett@ceh.ac.uk">j.garnett@ceh.ac.uk</a> Data: Explanation of column headings for MASQ_PHLOI_DATA file
MASQ_PHLOI_SITEINFO_NO_BH_VIEW	View contains MASQ_PHLOI_DATA connected to selected MASQ_GENERAL_SITE_INFO and CS2000 data
MASQ_PHLOI_NO_BH_VIEW	View contains MASQ_PHLOI_DATA connected to selected CS2000 data
MASQ_PHLOI_SITEINFO_BH_VIEW	View contains MASQ_PHLOI_DATA connected to selected MASQ_GENERAL_SITE_INFO and CS2000 data
MASQ_SIEVEMOIST_BH_VIEW	View contains MASQ_SIEVEMOIST_DATA connected to selected CS2000 data
MASQ_SIEVEMOIST_NO_BH_VIEW	View contains MASQ_SIEVEMOIST_DATA connected to selected CS2000 data

## **6. SOIL PH FROM THE ECOLOGICAL SURVEY OF 1978 AND CS2000**

### **6.1 Introduction**

The assessment of pH in soils is a relatively easy and inexpensive measurement. It provides a great deal of information on how a soil will transform and release chemicals as well as providing a relatively consistent method for assessing long-term trends in soil acidity while the behaviour and local/regional distribution of soil biota often correlates with soil pH. As such, it is one of the first measurements to be included when experimenting with or monitoring soils.

Soil pH was measured in soil samples collected during the first Ecological Survey of Great Britain in 1978 by the Institute of Terrestrial Ecology (now integrated within CEH). The re-analyses of soil pH from the same sample locations as 1978 presented a unique opportunity to investigate long-term trends in soil pH across the British countryside, especially in semi-natural and upland systems.

Recent nationwide assessments of changes in soil pH, and other soil properties, have been focussed on lowland, agricultural systems (Webb et al., 2001) and national assessments are lacking from semi-natural and upland systems. It has been proposed that impacts of atmospheric pollution may affect semi-natural, upland systems more than other areas of the British countryside.

Several site-specific and regional studies within GB have found increased soil acidity (decrease in soil pH) from the late 1940's to the early 1990's that have been related, in part, to acid deposition (Adamson et al., 1996; Billet et al., 1988; Kuylenstierna and Chadwick, 1991). Pollution deposition patterns, high rainfall, cloud cover and vegetation sensitivity are, in general, higher or more frequent in these systems. Recent results from the Countryside Survey 2000 suggest, however, that semi-natural systems may be experiencing reduction in soil acidity (increase in soil pH) while the fertilisation effect from pollution continues (Haines-Young et al., 2000). Soil pH was, therefore, an key measurement to include in the assessment of soils from CS2000.

This section provides an overview of the soil pH data produced from the 1978 and CS2000 surveys along with a description of methods. Summary statistics are provided for soil pH at national scale and within various stratifications of the British countryside; Environmental Zone, Broad Habitat, ITE Land Class, CVS Aggregate Vegetation Class and Major Soil Group.

#### **6.1.1 Specific Objectives**

There were to:

- To measure soil pH in soil samples collected during CS2000, from the same locations as the 1978 Ecological Survey of Great Britain.
- To produce digital ORACLE GIS spatially referenced datasets for soil pH for the 1978 and CS2000 surveys.
- To examine the differences in soil pH between 1978 and CS2000.

- To interpret differences in soil pH in terms of soil type, geographical location, land use/cover and vegetation.
- To compare the soil pH data with other relevant national datasets.

## **6.2 Methods**

### **6.2.1 Ecological Survey of Great Britain 1978**

Soil sampling within the 1978 Ecological Survey has been summarised earlier. A full description is available in the Project Record. In 1978 soil samples were taken from the upper 15 cm of the soil profile in a soil pit. There was one important difference in this sampling compared to CS2000. In soils where the surface litter layer was greater than 5 cm, the litter layer was removed and the soil sample taken below this. The implications for comparing results between the two surveys are discussed in the text below. This removal of litter did not occur in all samples and by referring back to the field record sheets it should be possible to identify both how many and which samples were involved. These record sheets detail soil horizon depths, including the depth of the upper soil horizons from which the soil samples were obtained.

Soil pH in water was measured on all soil samples in 1978 using the same method detailed below for CS2000 soil samples. During CS2000, a single dataset was compiled from 1978 soil pH data recovered from the Countryside Information System and other Countryside Survey datasets. These data were checked against printouts from the analytical phase in 1978. Sample locations (X-plot numbers) were checked against record sheets. The X-plots that had not changed location from 1978 to CS2000 (through 1984 and 1990 field surveys) were identified within the datasets. These samples are referred to as “repeats” in the text and were used to determine differences in soil pH from 1978 to CS2000 (1998/9). The CS2000 data manager entered the 1978 dataset into ORACLE once data checks had been completed.

### **6.2.2 Countryside Survey S2000**

Soil sampling by the CS2000 field surveyors has been presented earlier. Full protocols for soil sampling, initial processing and analyses for soil pH are available in the Project Record. The following summarises the main procedures.

#### **Soil Sampling**

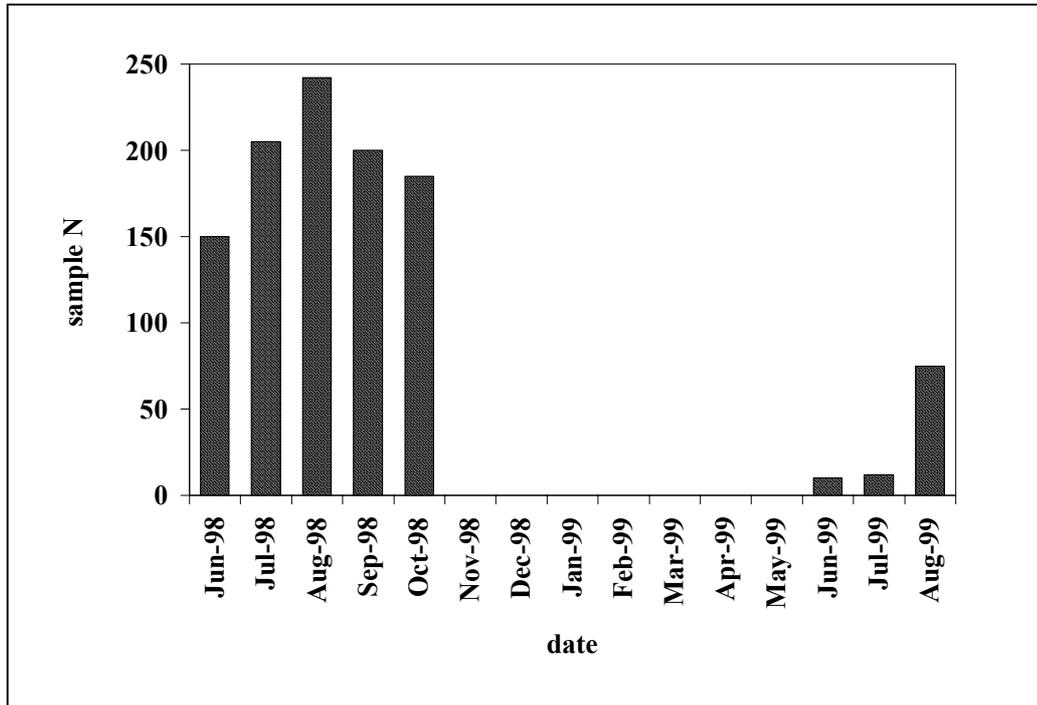
The field surveyors were instructed to take an 8 cm diam. x 15 cm soil core from each X-plots in all 256 1 km squares; the same squares sampled in 1978. Each core was stored in a labelled, un-sealed plastic bag in a cool-box and returned to CEH Merlewood by the field surveyors or by the field co-ordinators at regular intervals (see Figure 6.1).

#### **Measurement of pH**

On arrival at CEH Merlewood, all soil cores were followed through the various analytical phases via the daily diary and laboratory log sheets. In the first instance, a vertical slice was taken from each newly arrived soil sample for the measurement of pH (“fresh” soil pH). The soil was then processed to produce a 2 mm sieved, air-dried sample for long-term storage. Soil pH was measured on a homogenised sub-sample of this before storage. This “dry” soil pH can be used, in future, to determine effects of

length of storage and methodology. Soil sampled were weighed at the various analytical stages and, therefore, the amount of soil in storage is known as well as the amount of more than 2mm soil and water content. Soil pH was measured by electrode from a soil/water mixture using a standard technique (Allen et al., 1989).

Figure 6.1. Histogram showing the processing rate of soil cores for chemical analyses at CEH Merlewood during CS2000.



### Quality control

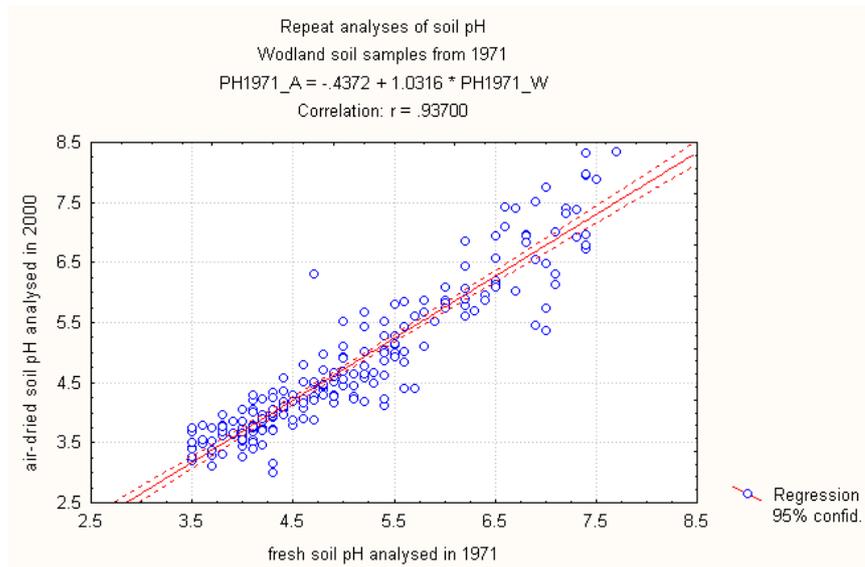
The pH electrode was calibrated using standard buffer solutions and re-calibrated after every 10<sup>th</sup> sample and a sub-set of samples was re-analysed to confirm results. All QC data are available from the ORACLE soil pH dataset.

### 6.2.3 Reproducibility of methods between 1978 and CS2000

Soil samples taken in 1971 from woodland habitats across Great Britain were used to determine whether differences in the analytical methods used in 1978 and CS2000 introduced significant variation in soil pH results. These air-dried, 2 mm sieved, soil samples are stored at CEH Merlewood.

The original pH results from 1971 were obtained using the same methods as the 1978 Ecological Survey. During CS2000, soil pH was re-analysed on 216 of these soils using the same method and equipment for the CS2000 soil samples (Smart et al., 2000). Results are presented in Figures 6.2. Both measurements gave highly comparable results. There were no statistically significant differences between these analyses (dependent sample T-test;  $P < 0.005$ ).

Figure 6.2. Soil pH taken in 2000 from stored samples collected in 1971 plotted against the original pH from the field-fresh soil samples. Methods in 2000 were the same as those from CS2000 and the methods in 1971 were the same as those from the 1978 Survey.



#### 6.2.4 Data management and analyses

All data from CS2000 were entered into log-sheets, from where they were transferred to Excel spreadsheets by project staff. These datasheets were checked by the project data manager and/or another person, with summary statistics carried out to check for errors (e.g. distribution graphs, max, mean, min.). Approximately 10% of all Excel data were checked against the log-sheets. Once completed, the final Excel spreadsheets were sent to the data manager for transfer into the MASQ database, along with relevant meta-data. These datasets are maintained in an ORACLE database at CEH Merlewood. All data are archived on a regular basis and copies maintained off-site under secure conditions. Summary datasets for loss-on-ignition were extracted from ORACLE.

Summary statistics on SOM produced in tabular form using SAS and Statistica software maintained, under license, by CEH. Unless otherwise stated, all results from CS2000 soil pH were calculated on the soil pH from the “fresh” (non-dried soil) since this was equivalent to the soil samples analysed in 1978.

*Regression to the mean:* This phenomenon occurs in all datasets where there is less than 100% correlation between factors. It does not affect the overall results of the data but can result in spurious associations when changes in a factor are plotted against the initial values (Hill, 1974). These spurious associations have been highlighted in previous studies of changes in soil pH over time (Adamson, et al., 1996). To prevent this artefact, changes should be plotted against the average values over the timespan (Streiner, 2001). Therefore, differences in soil pH from 1978 to CS2000 have been plotted against the mean of 1978 and CS2000 soil pH where relevant.

### 6.3 Results

Table 6.1. presents summary statistics for the complete datasets and for a sub-set of “repeat” X-plots from 1978 to CS2000 i.e. where the X-plots have maintained the same location from 1978 to CS2000. Distribution plots of soil pH values in 1978 and CS2000 and the difference in soil pH between these surveys are plotted in Figure 6.3. Figure 6.4 illustrates that there is a strong correlation in soil pH values between 1978 and CS2000, with few inconsistent values.

Table 6.1. Summary statistics for soil pH from 1978 and CS2000.

year	dataset	Sample N	Mean	Median	Min	Max	Range	Std.
1978	all data	1148	5.43	5.2	3.2	8.65	5.45	1.27
CS2000	all data	1071	5.72	5.58	3.4	8.71	5.31	1.32
1978	repeats	833	5.30	5.1	3.2	8.65	5.45	1.19
CS2000	repeats	783	5.59	5.47	3.4	8.71	5.31	1.22

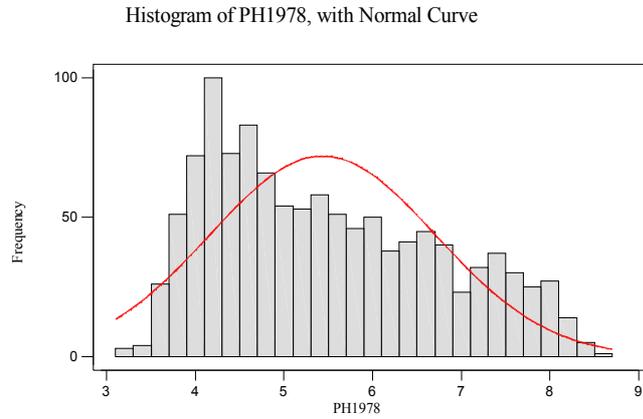
A total of 1148 and 1071 soil samples were measured for soil pH in 1978 and CS2000, respectively. The selection of the X-plots that maintained their locations from 1978 to CS2000 reduces the number of samples by 27% in both surveys. Of these, 768 were exactly the same X-plots in 1978 and CS2000 (Table 6.2). Sample numbers were less than optimal in both surveys for several reasons; samples were not collected due to sampling difficulties; too little soil was available for analyses; in CS2000, a small number of samples were collected from the wrong location.

#### 6.3.1 Soil pH in the British Countryside

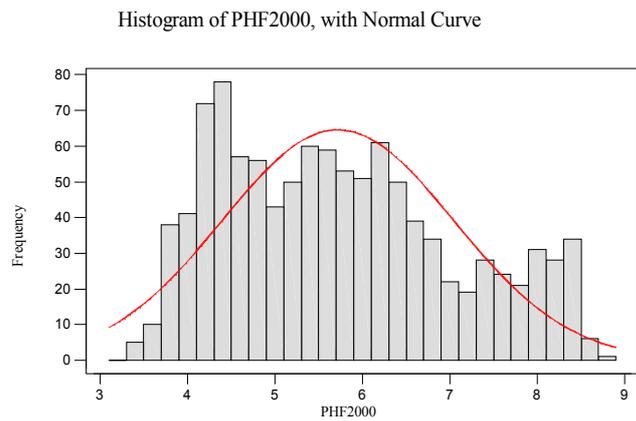
The results indicate that soils in the British countryside were predominately acidic with a median pH of less than 5.6 in both surveys (Table 6.1; see also Figure 6.3). The distribution plots of the soil pH data reveal that the soil pH pattern for 1978 was skewed towards the lower pH values while for CS2000, soil pH was more evenly distributed with a greater number of samples with pH more than 7. This trend was reflected in the summary statistics for the complete and repeat datasets. In both, the mean, median, minimum and maximum soil pHs in CS2000 were greater than those in 1978 while there was a narrower range of pH values in CS2000.

Statistical analyses (Table 6.2) showed that the difference in soil pH between 1978 and CS2000 was significant in the complete datasets (T-test for independent samples and in the repeat dataset (T-test for dependant samples). The mean increase in soil pH over this time was 0.27 pH units. The frequency histogram (Figure 6.3c) shows that the differences in soil pH from 1978 and CS2000 range from -3 to + 3 pH units with the majority of samples ( more than 90%) between -0.5 and 1.25 pH units; repeats dataset only. The data are being analysed further to examine reasons for values to occur outside the 90% intervals. Figure 6.5. presents the mean differences in soil pH between the two surveys for five pH intervals from pH 3.5 to 8.5; calculated from the average of 1978 and CS2000 pH values to negate the influence of the regression of the mean (as detailed above). The results indicate that increases in soil pH occurred in all soil pH intervals and mainly at pH 4.5-5.5 and pH 7.5-8.5.

Figure 6.3. Frequency histograms of soil pH data for (a) 1978, (b) CS2000 and (c) difference in pH from 1978 to CS2000  
 (a) 1978



(b) CS2000



(c) difference between 1978 and CS2000

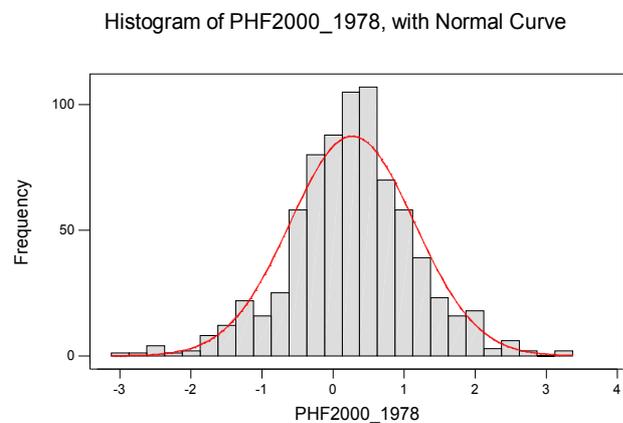


Table 6.2. Results from T-test of soil pH from 1978 and CS2000 using all data and repeats only; differences significant at  $P < 0.05$

Year	Datasets	Mean	Std.	N	Diff.	Std diff.	t	df	P
1978	all	5.43	1.288						
CS2000	all	5.72	1.322	1051	0.278	0.873	10.33	1050	6.8E-24
1978	repeats	5.30	1.210						
CS2000	repeats	5.59	1.218	768	0.274	0.873	8.71	767	1.9E-17

Figure 6.4. Scatterplot of soil pH from CS2000 plotted against soil pH from 1978.

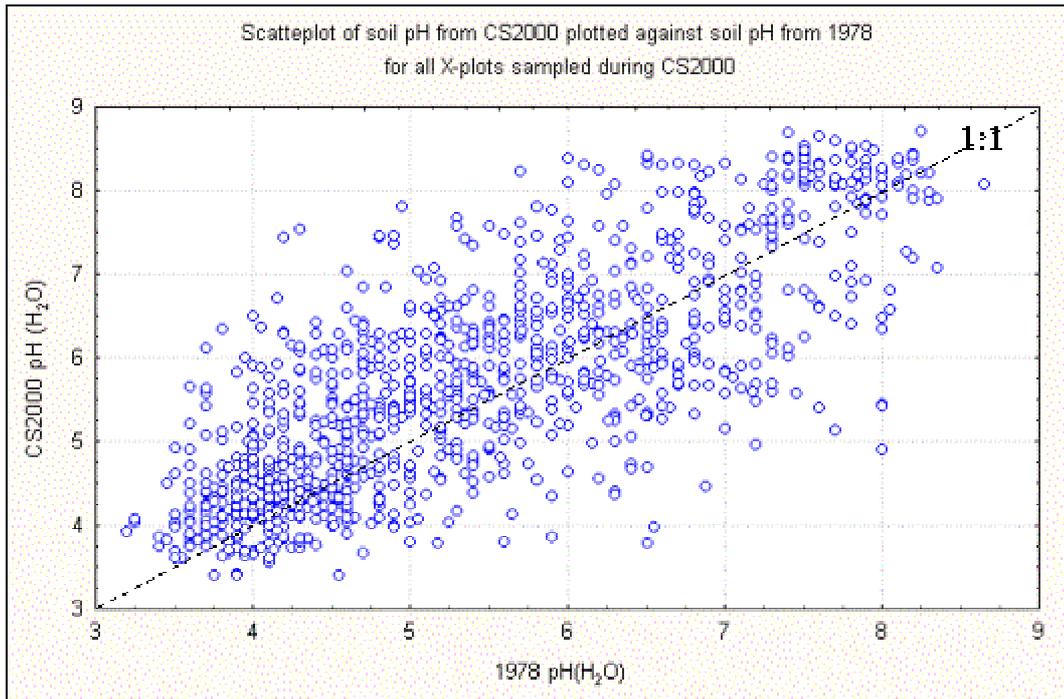
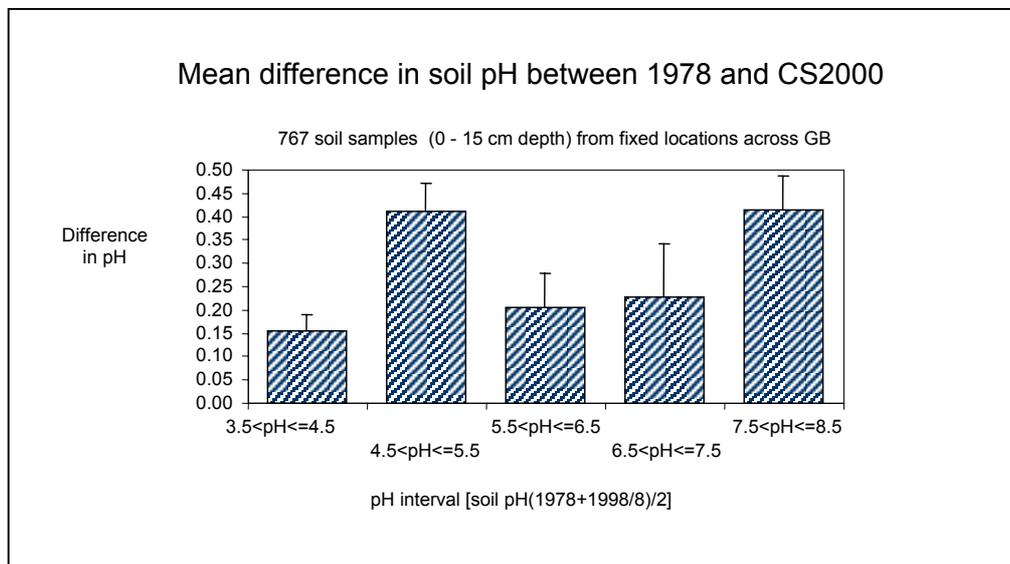


Figure 6.5. Mean difference in soil pH between 1978 and CS2000 for average soil pH intervals between the two surveys; repeat X-plots only.



### 6.3.2 Environmental Zones

Summary statistics for soil pH from 1978 and CS2000 within each Environmental Zone (EZ) are presented in Tables 6.3, 6.4 and 6.5. The total number of soil pH values obtained from each Environmental Zone ranged from 153 (EZ6) to 287 (EZ2) from the 1978 dataset (Table 6.3). Over 88% of all 1978 X-plots samples were retrieved in CS2000 (Table 6.4). Of these, 52 to 82% were repeated X-plots (Table 6.5).

Soil pH from 1978 and CS2000 followed predictable trends with the lowest pH values recorded in EZ 3, 5 & 6 (upland areas) and highest pH values recorded in EZ 1, 2 & 4, predominately lowland and agricultural areas. There were differences in the minimum and maximum pH values between the two surveys. In CS2000, the minimum pH values were lower than in 1978 in EZ 1, 2 and 6 and greater than in 1978 in EZ 3, 4 and 5. Maximum pH values were greater in CS2000 in EZ 1, 2, 3 and 4 and lower in EZ 5 and 6 than in 1978.

Table 6.3 Summary statistics for soil pH in 1978 for Environmental Zones.

Environmental Zone	N.	mean	median	min	max	Std
1	251	6.77	6.88	3.75	8.65	1.08
2	287	5.68	5.7	3.45	8.05	1.14
3	117	4.51	4.3	3.2	6.6	0.82
4	161	5.5	5.5	3.75	8	0.89
5	179	4.56	4.35	3.4	7.7	0.73
6	153	4.46	4.38	3.5	7	0.59

Table 6.4. Summary statistics for soil pH in CS2000 for Environmental Zones.

Environmental Zone	N	Mean	median	min	max	std	%1978
1	229	7.02	7.43	3.4	8.71	1.270	91.24
2	270	6.12	6.125	3.4	8.53	1.133	94.08
3	117	4.92	4.78	3.62	7.03	0.946	100.00
4	142	5.58	5.64	3.8	8.16	0.925	88.20
5	176	4.94	4.705	3.62	7.46	0.737	98.32
6	137	4.60	4.47	3.43	6.59	0.670	89.54

Summary statistics for the difference in soil pH between repeat X-plots from 1978 and CS2000 (Table 6.5) indicate that soil pH was greater in all zones in CS2000 compared to 1978. The greatest increases were recorded in EZ 2 and 3 (England and Wales) and EZ 5 (Scotland) and the lowest in EZ 1 with intermediate increases in EZ 4 & 6. These results also show that differences in soil pH has declined, as well as increased, in some X-plots.

These data will be further explored to determine whether the differences in soil pH between 1978 and CS2000 correspond to particular vegetation types and/or changes land uses within an Environmental Zone.

Table 6.5. Summary statistics for difference in soil pH between 1978 and CS2000; repeat X-plots only.

Environmental Zone	N	mean	median	min	max	std
1	130	0.03	0.01	-3.1	2.66	1.09
2	210	0.43	0.47	-1.65	3.25	0.87
3	88	0.43	0.42	-1.55	2.57	0.78
4	108	0.14	0.08	-2.56	2.54	0.88
5	138	0.36	0.4	-2.57	2.5	0.7
6	93	0.12	0.14	-2.72	2.08	0.76

### 6.3.3 Broad Habitats

Summary statistics are presented in Tables 6.6, 6.7 and 6.8. Broad Habitat classifications are such that an X-plot could be assigned to more than one Broad Habitat. The total N may, therefore, be more than the number of samples taken. The number of pH results ranged from 329 to 1 within each Broad Habitat and seven Broad Habitats contained less than 10 soil pH results. Improved grassland and arable/horticultural, followed by bog, contained most pH values from 1978 and CS2000. Future analyses should consider appropriate combinations of Broad Habitats to increase sample N for statistical analyses. Results are discussed for the Broad Habitats with more than 7 samples.

Median soil pH across the Broad Habitats ranged from 4.1 to 6.8 in 1978 and from 4.29 to 7.07 in CS2000. In both surveys, the highest median pH was recorded in arable/horticultural and the lowest pH was recorded in coniferous woodland. In CS2000, median soil pH values were acidic in all but arable/horticultural and neutral grassland habitats. The trend for higher median soil pH values in CS2000 compared to 1978 occurred in all Broad Habitats. Minimum soil pH values were greater in CS2000 than 1978 in 9 out of 10 Broad Habitats (where sample N more than 10) while maximum soil pH values were greater in 8 out of 10. Coniferous woodlands and dwarf shrub heath were the two habitats where minimum and/or soil pH values were greater in 1978 than CS2000.

These results were further confirmed by the summary statistics for differences in soil pH between 1978 and CS2000 (Table 6.8). The greatest increase in soil pH occurred in Fen, marsh & swamp and neutral grassland, followed by acid grassland. The exception was coniferous woodland where, unlike all other habitats, the median difference in soil pH was negative (-0.04); Table 6.10. The increase in soil pH was reflected in the distribution of the median soil pH. In CS2000, the median soil pH was less than 5.0 in four habitats compared to six habitats in 1978.

Table 6.6. Summary statistics for soil pH in 1978 for each Broad Habitat; all data.

Broad Habitat	No.	mean	median	min	max	st. dev
Improved grassland	329	5.75	5.6	3.6	8.2	0.96
Arable and horticultural	255	6.73	6.8	4.4	8.65	0.98
Neutral grassland	44	5.46	5.25	3.5	8	1.14
Broadleaved, mixed and yew woodland	69	5.09	4.75	3.45	7.8	1.2
Coniferous woodland	86	4.29	4.1	3.5	7.2	0.69
Bog	143	4.43	4.3	3.2	7	0.55
Dwarf shrub heath	97	4.34	4.2	3.25	6.5	0.64
Acid grassland	66	4.31	4.2	3.5	6.35	0.5
Fen, marsh and swamp	39	4.86	4.7	3.4	6.9	0.85
Bracken	20	4.2	4.13	3.5	5.1	0.47
Calcareous grassland	6	7.23	7.4	6.5	7.5	0.37
Inland rock	1	4.2	4.2	4.2	4.2	.
Built-up areas and gardens	9	6.01	6.3	3.95	7.5	1.15
Boundary and linear features	1	5.4	5.4	5.4	5.4	.
Supralittoral sediment	1	6.65	6.65	6.65	6.65	.
Supralittoral rock	2	4.85	4.85	4	5.7	1.2
Littoral sediment	7	7.33	7.5	5.3	8.05	0.96

Table 6.7 Summary statistics for soil pH in CS2000 for each Broad Habitat; all data

Broad Habitat	No.	mean	median	min	max	st. dev
Improved grassland	310	6	5.82	3.99	8.53	0.94
Arable and horticultural	227	7.1	7.07	4.55	8.71	0.94
Neutral grassland	42	5.98	6.01	3.81	8.32	0.95
Broadleaved, mixed and yew woodland	62	5.4	5.07	3.63	8.41	1.45
Coniferous woodland	84	4.39	4.29	3.4	7.02	0.67
Bog	130	4.7	4.5	3.67	6.85	0.68
Dwarf shrub heath	94	4.51	4.39	3.4	6.51	0.67
Acid grassland	64	4.57	4.48	3.55	6.72	0.63
Fen, marsh and swamp	39	5.5	5.41	3.75	7.41	1
Bracken	18	4.81	4.36	3.89	6.6	0.87
Calcareous grassland	6	7.81	8.29	5.7	8.49	1.08
Inland rock	1	4.16	4.16	4.16	4.16	.
Built-up areas and gardens	10	6.45	6.48	4.46	8.24	1.38
Boundary and linear features	1	5.39	5.39	5.39	5.39	.
Supralittoral sediment	1	7.41	7.41	7.41	7.41	.
Supralittoral rock	3	5.43	4.57	4.11	7.62	1.91
Littoral sediment	6	7.67	7.7	6.93	8.3	0.44

Table 6.8. Summary statistics for differences in soil pH between 1978 and CS2000 for each Broad Habitat; repeat X-plots only.

Broad Habitat	No.	mean	median	min	max	st. dev
Improved grassland	262	0.26	0.33	-2.57	3.25	0.94
Arable and horticultural	123	0.23	0.21	-3.1	2.66	0.96
Neutral grassland	32	0.34	0.5	-1.79	1.88	0.9
Broadleaved, mixed and yew woodland	44	0.35	0.25	-1.93	2.32	1.07
Coniferous woodland	56	0.01	-0.04	-2.24	1.43	0.73
Bog	100	0.26	0.21	-1.84	2.5	0.71
Dwarf shrub heath	69	0.22	0.27	-2.72	2.1	0.78
Acid grassland	38	0.4	0.44	-1.02	2.57	0.7
Fen, marsh and swamp	29	0.57	0.51	-0.56	1.8	0.52
Bracken	14	0.54	0.38	-0.52	1.63	0.62
Calcareous grassland	4	0.18	0.62	-1.5	0.99	1.17
Inland rock	1	-0.04	-0.04	-0.04	-0.04	.
Built-up areas and gardens	6	1.2	1.07	-0.27	2.64	1.05
Boundary and linear features	1	-0.01	-0.01	-0.01	-0.01	.
Littoral sediment	.	.	.	.	.	.
Supralittoral sediment	.	.	.	.	.	.
Supralittoral rock	1	0.11	0.11	0.11	0.11	.

#### 6.3.4 ITE Land Classes

Tables 6.9 and 6.10 present summary statistics for 1978, CS2000 and differences in soil pH between 1978 and CS2000. From 40 ITE Land Classes, there were 13 Land Classes with less than 20 pH values from 1978 and CS2000. Future analyses should consider appropriate combinations of ITE Land Classes to increase sample N for statistical analyses. In both years, the lowest pH was recorded in ITE Land Class 22s and the highest in 12e for 1978 and 3e for CS2000. In 1978, there were a greater number of Land Classes with a median soil pH less than 5 than in CS2000 while in CS2000 there were three more Land Classes with soil pH more than 6 than in 1978 (Table 6.12). The difference data indicate that, in Land Classes with more than 20 samples, the median soil pH was higher in 17 Land Classes and lower in 4 Land Classes in CS2000 compared to 1978. The greatest increase was found in 7e and the greatest decrease in 11e.

Table 6.9. Summary of differences in soil pH between 1978 and CS2000 by ITE Land Class; all pH data included.

Property	1978	CS2000
Median soil pH	Number of classes	Number of classes
<5	12	9
5 to 6	8	8
6 to 7	5	6
>7	2	4
Min. pH	ITE Land Class 22s	ITE Land Class 22s
Max. pH	ITE Land Class 12e	ITE Land Class 3e

Table 6.10. Summary statistics for soil pH from 1978, CS2000 and pH difference from 1978 to CS2000 by ITE Land Class.

1978							CS2000						DIFFERENCEE					
ITE LC	N	mean	median	min	max	sd	N	mean	median	min	max	sd	N	mean	median	min	max	sd
10e	54	5.9	5.9	3.6	7.7	0.9	52	6	6.2	3.4	7.6	1	40	0.1	-0	-1.7	2.4	1
11e	57	6.6	6.6	4.2	8.3	0.9	49	6.7	6.8	3.8	8.4	1.3	27	-0.6	-0.5	-3.1	1.9	1.1
12e	20	7.3	7.3	5.7	8.3	0.8	20	7.4	7.6	5.6	8.4	0.8	8	-0	-0.2	-1.3	2.5	1.3
13e	27	5.7	5.7	3.6	7.4	1.1	27	6	5.9	4.1	7.8	1	24	0.4	0.3	-1.4	3.2	1.1
13s	15	4.9	4.9	4	6	0.6	15	4.8	4.8	3.8	6.3	0.7	11	0	-0.1	-0.6	0.9	0.5
15e	25	5.1	5.1	3.5	7.2	1.2	17	6	5.8	4.1	7.6	1.1	14	0.8	0.7	-0.3	1.7	0.6
16e	30	5.2	5.2	3.6	8.1	1.1	30	6.1	6	4.1	8.3	1.2	24	0.7	0.6	-1.6	2.2	0.9
17e	20	4.9	4.9	3.2	6.6	1	20	5.1	5.3	3.8	6	0.7	18	0.2	0.1	-1.3	1.9	0.8
17w1	10	5.1	5.1	4	5.9	0.7	10	5.6	5.7	4.4	7	0.8	10	0.5	0.5	-1.6	1.5	0.9
17w3	18	4.9	4.9	3.6	6.4	0.8	18	5.5	5.5	3.8	6.9	0.9	15	0.5	0.4	-1	2.2	0.9
18e	25	4.3	4.3	3.5	6.2	0.8	25	5	4.9	3.6	6.7	1	16	0.8	0.9	-0.5	2.6	0.8
18s	5	3.8	3.8	3.6	4.2	0.2	3	4.4	4.1	4	5	0.6						
19e	15	4.3	4.3	3.3	5.5	0.8	15	5	4.5	3.6	7	1.1	10	0.5	0.5	-0.8	1.3	0.6
19s	5	4.4	4.4	3.6	5.4	0.7	5	4.9	4.7	4.1	5.8	0.7	4	0.7	0.7	-0	1.4	0.7
1e	28	5.9	5.9	3.9	7.4	1.2	28	6.4	6.4	3.7	8.4	1.3	24	0.4	0.3	-1.1	3.3	0.9
21s	44	4.3	4.3	3.7	5.7	0.5	37	4.5	4.4	3.4	5.9	0.5	30	0.1	0.2	-1.5	1.1	0.6
22e	14	4	4	3.6	4.7	0.3	14	4.3	4.1	3.6	5.8	0.6	11	0.4	0.3	-0.6	1.3	0.5
22s	35	4.3	4.3	3.5	6.2	0.5	32	4.5	4.2	3.7	6.1	0.6	21	0.1	0.2	-0.9	1	0.5
23e	15	4.1	4.1	3.6	5	0.4	15	4.1	3.9	3.7	5.5	0.5	8	0.1	0.2	-1.2	1.4	0.7
23s	34	4.5	4.5	3.7	6.5	0.6	28	4.4	4.4	3.6	5.5	0.5	14	0	0.1	-2.7	1.7	1.1
24s	40	4.8	4.8	3.7	7	0.6	40	5	4.8	4	6.6	0.8	28	0.2	0.1	-1.8	2.1	0.9
25e	9	6.1	6.1	4.4	7.1	0.9	9	6.4	6.1	5.7	7.5	0.7	9	0.4	0.1	-1	2.7	1.3
25s	46	5.2	5.2	3.8	6.5	0.8	41	5.6	5.8	3.8	7	0.9	34	0.4	0.3	-1.4	2.5	0.9
26s	35	5.8	5.8	4	7.5	0.8	31	5.6	5.7	4	6.8	0.8	22	-0.1	-0.2	-2.6	2	1.1
27s	40	5.9	5.9	4	8	1	34	5.9	5.9	4.1	8.2	1.1	27	-0.1	-0.2	-1.5	1.8	0.8
28s	36	4.4	4.4	3.5	6.5	0.6	36	4.8	4.5	3.6	6.3	0.7	32	0.4	0.4	-0.8	1.4	0.5
29s	43	4.7	4.7	3.6	7.7	0.8	40	5	4.7	3.7	6.5	0.7	35	0.3	0.2	-2.6	1.9	0.9
2e	46	6.7	6.7	3.8	8.2	1.2	45	6.9	7.7	3.8	8.5	1.5	30	0	0	-2.6	2.3	1.2
30s	35	4.4	4.4	3.7	5.4	0.4	30	4.7	4.6	4.1	6.2	0.6	23	0.4	0.6	-1.1	1.3	0.6
31s	26	4.9	4.9	4	7.3	0.8	32	5.3	5.2	4.2	7.5	0.9	17	0.6	0.6	-1.7	2.1	0.8
32s	29	4.7	4.7	3.4	7.1	1	30	4.9	4.5	3.8	6.7	0.8	27	0.2	0.3	-1.5	2.5	0.7
3e	51	7.1	7.1	4.1	8.7	1	47	7.5	8	5.1	8.7	1	20	0.4	0.2	-1	2.5	0.9
4e	19	7.4	7.4	5.5	8.4	0.9	14	8	8.2	6.5	8.7	0.7	11	0.3	0.2	-0.7	1.8	0.7
5e	11	5.4	5.4	4.3	7	0.9	11	5.8	5.9	4.9	6.7	0.5	9	0.6	0.6	-1	1.7	0.9
6e	39	4.7	4.7	3.5	6.2	0.8	35	5	5.1	3.8	6.7	0.7	26	0.5	0.5	-0.7	1.5	0.6
7e	42	6.4	6.4	4.4	8.1	0.9	39	6.8	6.6	4.8	8.5	1	28	0.3	0.6	-1.6	1.4	0.9
7s	25	5.3	5.3	3.8	6.7	0.8	21	5.4	5.6	3.9	7.2	0.8	14	0.4	0.3	-1.2	1.8	0.9
8e	31	6.4	6.4	4.6	8	1	31	6.9	7	5	8.3	0.9	21	0.5	0.6	-0.4	1.6	0.6
9e	49	6.3	6.3	3.8	8.2	1.1	45	6.7	6.9	3.4	8.4	1.3	25	0.3	0.4	-1.2	2.6	0.9

### 6.3.5 CVS Aggregate Vegetation Class

Summary statistics for 1978, CS2000 and difference in soil pH from 1978 to CS2000 are presented in Tables 6.11, 6.12 and 6.13. Figures 6.6 and 6.7 present further summary statistics for soil pH in 1978 and CS2000, for all data and for the repeats dataset only and show the range of pH values with medians and outliers. In both years, the highest median soil pH was recorded in Crops/weeds and Tall grass/herbs and the lowest median soil pH was recorded in Upland woodland and Heath & Bog (Tables 6.11 and 6.12). In all but one AVC, the median soil pH in CS2000 was higher than 1978. In Lowland Wood, the median soil pH was lower in CS2000 compared to 1978. Overall the distribution of median soil pH was different between CS2000 and 1978, with an additional class with a median soil pH less than 4 (Lowland Wooded), only two classes with a median soil pH between 5 and 7, and two classes with a median soil pH more than 7.

Table 6.11. Summary statistics for soil pH in 1978 for CVS Aggregate Vegetation class

Aggregate Vegetation class	Code	No.	mean	median	min	max	st. dev
Crops and weeds	1	207	6.72	6.8	4.4	8.65	0.98
Tall grass and herb	2	63	6.48	6.6	3.95	8.25	1.06
Infertile grassland	3	220	5.51	5.35	3.6	8.2	1.02
Fertile grassland	3	207	5.98	5.9	3.8	8.2	0.95
Lowland wooded	5	29	5.48	5.18	3.45	7.8	1.57
Upland wooded	6	78	4.22	4.1	3.5	5.5	0.53
Moorland grass mosaics	7	137	4.44	4.35	3.2	6.5	0.64
Heath and bog	8	207	4.33	4.2	3.25	7	0.51

Table 6.12. Summary statistics for soil pH in CS2000 for CVS Aggregate Vegetation class; all data

Aggregate Vegetation class	Code	No.	mean	median	min	max	st. dev
Crops and weeds	1	185	7.02	7	3.99	8.71	0.99
Tall grass and herb	2	59	7.01	7.17	3.81	8.53	1.11
Fertile grassland	2	192	6.24	6.13	4.17	8.33	0.95
Infertile grassland	3	211	5.92	5.71	3.89	8.53	0.98
Lowland wooded	5	29	5.53	4.96	3.4	8.39	1.69
Upland wooded	6	69	4.45	4.26	3.4	6.36	0.71
Moorland grass mosaics	7	137	4.75	4.58	3.55	7.02	0.74
Heath and bog	8	189	4.49	4.4	3.4	6.5	0.56

Figures 6.6 and 6.7 show that there was a wide range in soil pH values in all Aggregate Vegetation Classes for 1978 and CS2000, with outliers in several classes. These values require further analyses to determine whether they are representative of the classes they are currently recorded against. In the classes with the most acidic soils, Heath & bog, Upland wood and Moorland grass mosaic, over 50% of the pH values fall within a relatively narrow range of pH units less than 1.5 for both years. The greatest range in pH in both 1978 and CS2000 was observed in Lowland wood. Figure 6.8 highlights the presence of outlier values in the repeat dataset that require further examination to

determine whether these samples are truly representative of the class they are currently recorded against.

Figure 6.6. Box-plots of soil pH from 1978 and CS2000 by Aggregate Vegetation Class;

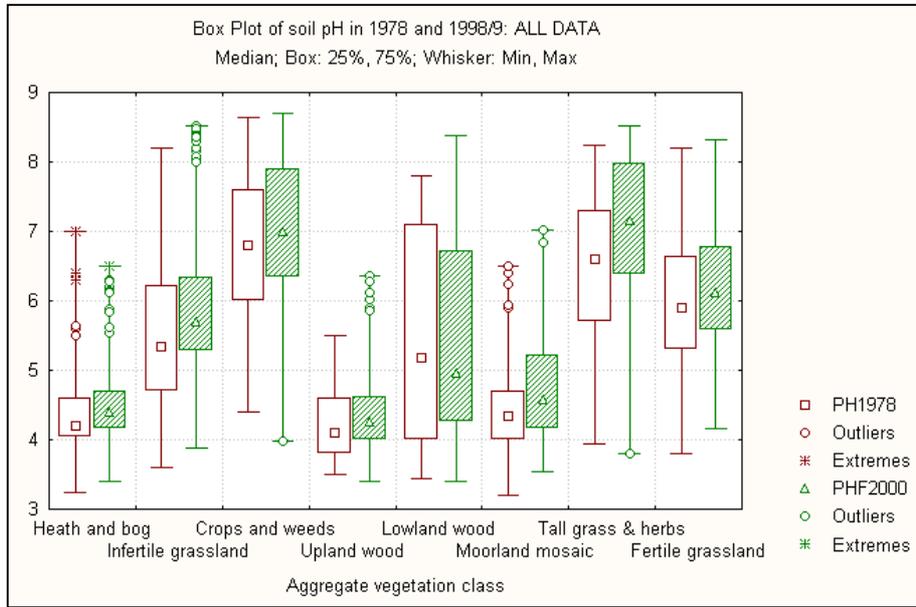
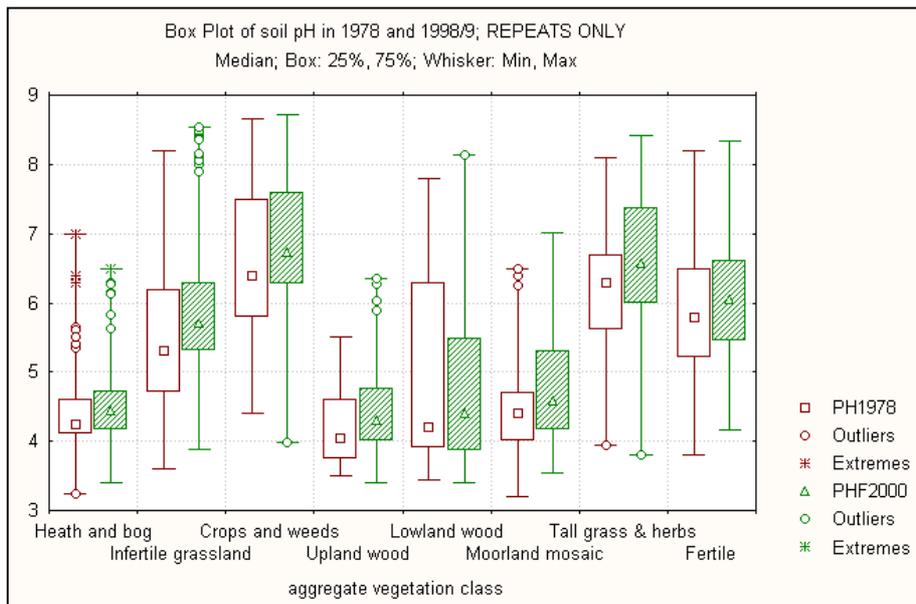


Figure 6.7. Box-plots of soil pH from 1978 and CS2000 (1998/9) by Aggregate Vegetation Class. Repeat X-plots only.

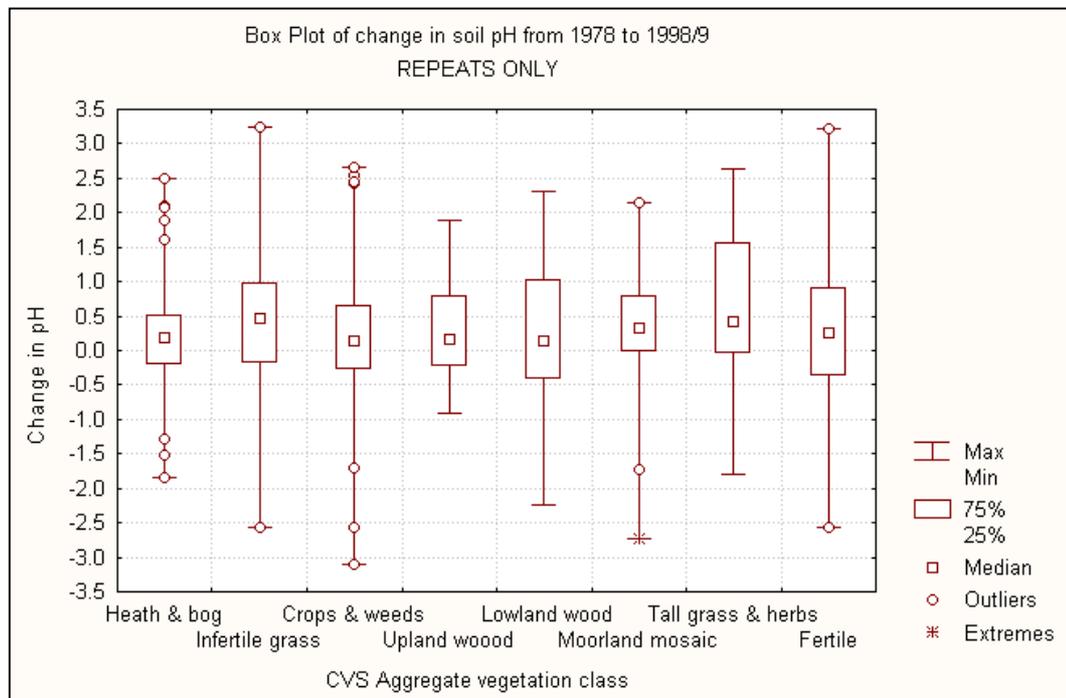


Differences in soil pH between 1978 and CS2000 (Table 6.13) were in general agreement with the overall trend for an increase in soil pH from 1978 to CS2000. The greatest increases in soil pH occurred in infertile grassland and tall grass/herbs. The lowest increases occurred in lowland wooded areas, crops/weeds and heath/bogs. Contrary to all other AVC data, there was an overall decrease in soil pH in lowland wooded areas.

Table 6.13. Summary statistics for difference in soil pH between 1978 and CS2000 for CVS Aggregate Vegetation class; repeat X-plots only.

Aggregate Vegetation Class	Code	No.	mean	median	min	max	st. dev
Crops and weeds	1	110	0.15	0.15	-3.1	2.66	0.92
Tall grass and herb	2	27	0.48	0.41	-1.8	2.64	1.2
Fertile grassland	3	149	0.25	0.26	-2.57	3.23	0.97
Infertile grassland	4	176	0.41	0.46	-2.57	3.25	0.9
Lowland wooded	5	19	-0.04	0.13	-2.24	2.32	1.2
Upland wooded	6	43	0.3	0.17	-0.92	1.9	0.71
Moorland grass mosaics	7	98	0.33	0.33	-2.72	2.15	0.75
Heath and bog	8	145	0.17	0.18	-1.84	2.5	0.66

Figure 6.8. Box-plots of difference in soil pH between 1978 and CS2000 (1998/9) by Aggregate Vegetation Class. Repeat X-plots only.



### 6.3.6 Major Soil Groups

Summary statistics for 1978, CS2000 and difference in soil pH from 1978 to CS2000 are presented in Tables 6.14, 6.15 and 6.16. In 1978 and CS2000, the highest median soil pH was recorded in Pelosols and the lowest median pH recorded in Peats and Podzolic soils. In all, the median soil pH was higher in CS2000 than in 1978. In 1978, there were two major soil groups with a median pH less than 5, four major groups with median pH 5 to 6 and one major group with median pH more than 7. In CS2000, median pH was less than 5 in the same two major groups (peat and podzolic soils) and between pH 5 and 6 for lithomorphous soils and surface-water gleys (although the median values were greater than those in 1978). In ground-water gleys and brown soils, the soil pH was greater in CS2000 with the median pH now greater than 6. Although pelosols still recorded a median soil pH of more than 7 in CS2000, this median value was less than 1978.

There was a wide range in soil pH values in all Major soil groups for 1978 and CS2000 with outliers in several classes (Figure 6.9). Outliers and extremes are also present in the repeat data only (Figure 6.10). In the groups with the most acidic soils, Peats and Podzolics, over 50% of the pH values fall within a relatively narrow range of pH units less than 1.5 for both years. Soil pH values in both gleys and lithomorphous soils typically exhibited a range of 1.5 to 2 pH values. The values require further analyses to determine whether these samples are truly representative of the class they are currently recorded against.

The median values for differences in soil pH from 1978 to 2000 by Major Soil Groups (Table 6.16) concur, in all but one Group, with the overall trend for increased soil pH from 1978 to CS2000; a decrease in soil pH was noted in pelosols. The highest median increase in soil pH appeared in surface water gleys and peat soils. The box plot (Figure 6.11) highlights the presence of outlier values in the repeat dataset that require further examination as outlined above.

Table 6.14. Summary statistics for soil pH from 1978 for Major Soil Groups

1978	Code	No.	mean	median	min	max	st. dev
Lithomorphous Soils	3	109	5.49	5.05	3.6	8.2	1.29
Pelosols	4	29	7.12	7.4	5.4	8.25	0.85
Podzolic Soils	5	164	4.56	4.4	3.25	7.45	0.82
Brown Soils	6	381	5.87	5.8	3.5	8.65	1.18
Surface-water Gley Soils	7	201	5.44	5.3	3.6	8.3	1.23
Ground-water Gley Soils	8	127	6.04	5.9	3.5	8.35	1.19
Peat (organic) soils	10	137	4.29	4.2	3.2	6.7	0.51

Table 6.15. Summary statistics for soil pH from CS2000 for Major Soil Groups

CS2000	Code	No.	mean	Median	min	max	st. dev
Lithomorphous Soils	3	95	5.85	5.7	3.68	8.53	1.46
Pelosols	4	24	7.19	7.27	4.4	8.65	1.2
Podzolic Soils	5	153	4.77	4.53	3.62	7.44	0.82
Brown Soils	6	346	6.14	6.06	3.4	8.69	1.2
Surface-water Gley Soils	7	191	5.77	5.73	3.4	8.53	1.3
Ground-water Gley Soils	8	126	6.43	6.3	3.7	8.71	1.2
Peat (organic) soils	10	136	4.65	4.49	3.43	8.24	0.71

Figure 6.9. Box-plots of soil pH from 1978 and CS2000 (1998/9) by Major Soil Group.

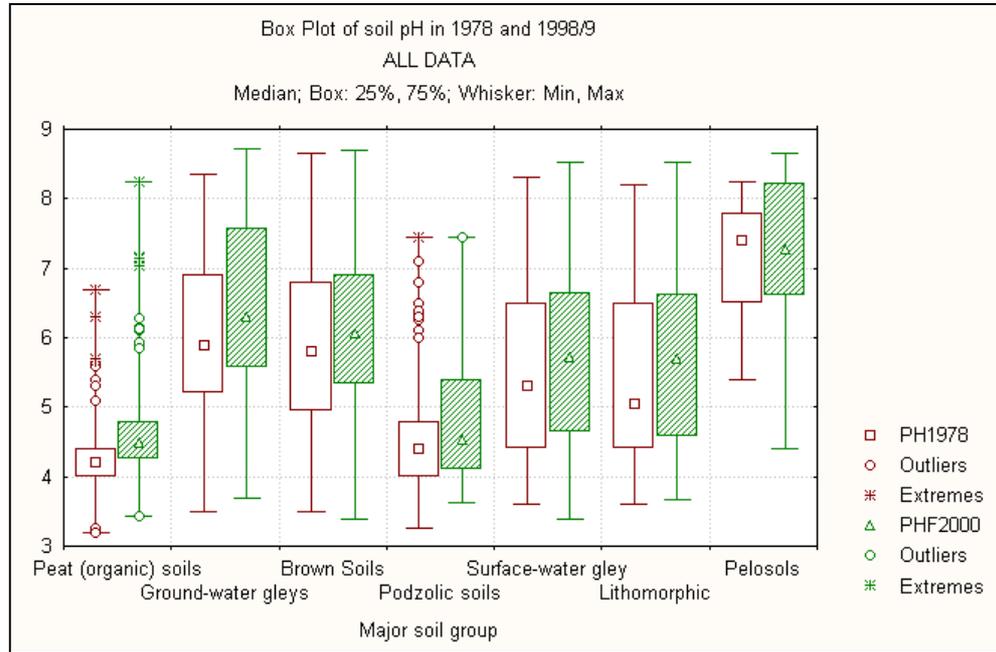


Figure 6.10. Box-plots of soil pH from 1978 and CS2000 (1998/9) Major Soil Groups. Repeat X-plots only.

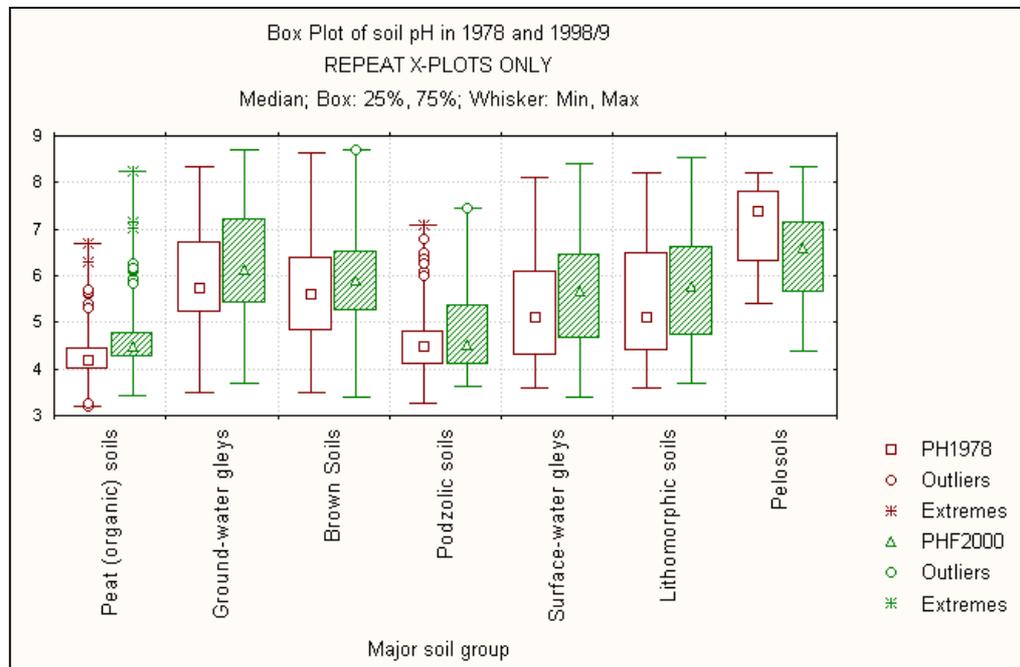
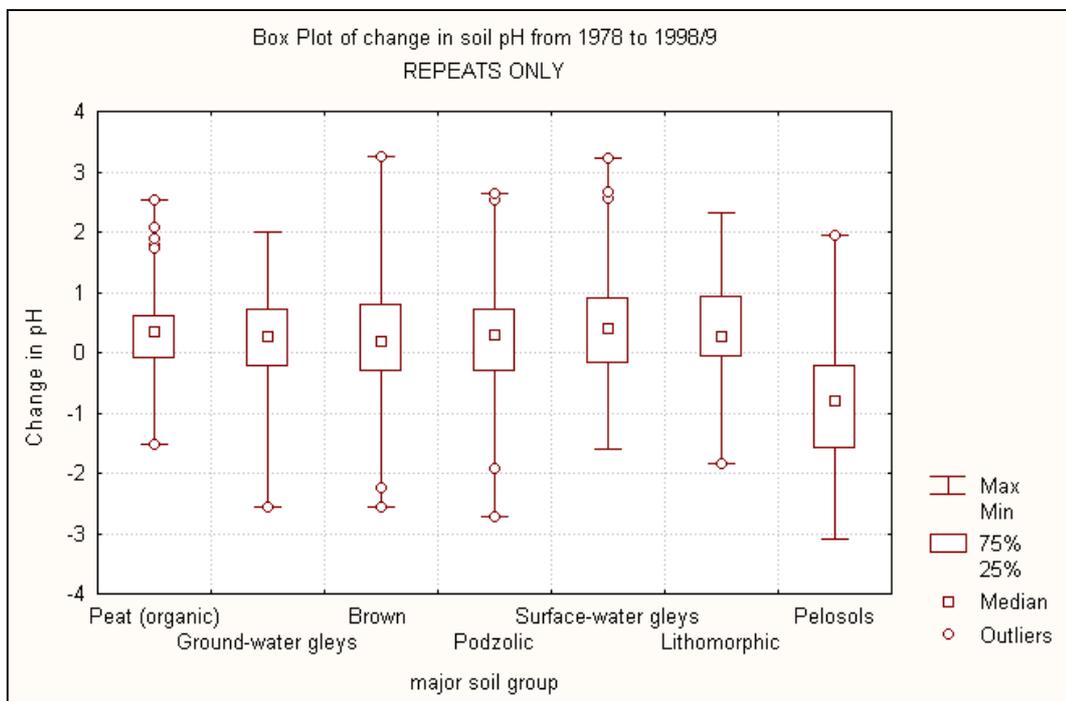


Table 6.16. Summary statistics for differences in soil pH between 1978 and CS2000 for Major Soil Groups; repeat X-plots only.

Difference 1978 to CS2000	Code	No.	mean	median	min	max	st. dev
Lithomorphic Soils	3	68	0.31	0.26	-1.84	2.32	0.86
Pelosols	4	12	-0.77	-0.8	-3.1	1.94	1.3
Podzolic Soils	5	112	0.24	0.29	-2.72	2.64	0.84
Brown Soils	6	247	0.24	0.2	-2.57	3.25	0.91
Surface-water Gley Soils	7	137	0.42	0.4	-1.61	3.23	0.9
Ground-water Gley Soils	8	92	0.22	0.27	-2.57	2	0.87
Peat (organic) soils	10	99	0.34	0.34	-1.51	2.54	0.64

Figure 6.11. Box-plots of difference in soil pH from 1978 and CS2000 (1998/9) by Major Soil Group.



### 6.3.7 Analyses of soil pH by Vegetation Aggregate Class and Major Soil Group

Summary statistics of soil pH in 1978, CS2000 (1998/9) and differences between 1978 and CS2000 for each AVC within each Major Soil Group are presented in Table 6.17. The results highlighted in bold are those with the highest sample N in each grouping. The higher median soil pH values in CS2000, compared to 1978, are repeated in all but 7 groupings. Only one of these (Upland wooded on Podzolic soils) contained more than 10 samples and only 6 of these groupings contained more than 30 samples. Further analyses should examine appropriate combinations to increase sample N.

The potential for examining pH within individual Aggregate Vegetation Classes is highlighted in Figure 6.12. This box-plot diagram presents summary statistics for pH within the Infertile Grassland AVC for each Major Soil Group in 1978 and CS2000 and the difference between the two years, using repeat data only. The number of samples in each major soil group ranged from 19 to 78 (excluding peats and pelosols where sample N = 2 and 0 respectively). The data from 1978 and CS2000 indicate that the typical range for soil pH in this AVC is from pH 5 to 6. There are however, exceptions to this, in particular for lithomorphous soils where the soil pH was generally much higher.

Figure 6.12. Box-plot of soil pH for 1978, CS2000 and difference in soil pH from 1978 to CS2000 in the infertile grassland Aggregate Vegetation Class split by Major Soil Group.

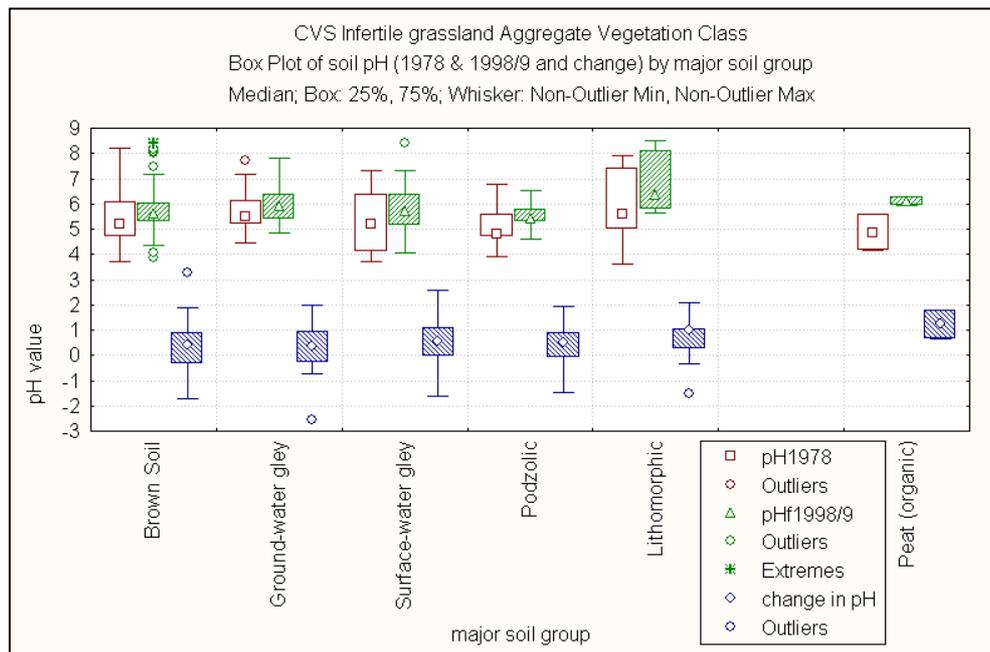


Table 6.17. Summary statistics for soil pH by Major Soil Group and Aggregate Vegetation Class in CS2000 and difference from 1978 to CS2000. Classes with more than 30 samples are highlighted in bold.

Major soil group	AVC	Soil pH CS2000			difference in soil pH from 1978 to CS2000		
		N	Median	Mean	N	Median	Mean
Brown soils	<b>Crops and weeds</b>	<b>56</b>	<b>6.57</b>	<b>6.67</b>	<b>55</b>	<b>0.10</b>	<b>0.09</b>
	<b>Fertile grassland</b>	<b>61</b>	<b>6.06</b>	<b>6.04</b>	<b>61</b>	<b>0.17</b>	<b>0.19</b>
	Heath and bog	3	4.70	4.53	3	-0.20	-0.44
	<b>Infertile grassland</b>	<b>78</b>	<b>5.61</b>	<b>5.78</b>	<b>78</b>	<b>0.42</b>	<b>0.32</b>
	Lowland wooded	8	4.57	4.54	8	-0.38	-0.40
	Moorland grass mosaic	14	4.86	5.05	14	0.49	0.51
	Tall grass and herbs	14	6.56	6.47	13	0.10	0.27
	Upland wooded	15	4.62	4.89	15	0.80	0.73
Ground-water gley	<b>Crops and weeds</b>	<b>23</b>	<b>7.70</b>	<b>7.23</b>	<b>22</b>	<b>0.23</b>	<b>0.13</b>
	<b>Fertile grassland</b>	<b>30</b>	<b>5.95</b>	<b>6.17</b>	<b>26</b>	<b>0.10</b>	<b>0.07</b>
	Heath and bog	1		3.70	1		0.20
	<b>Infertile grassland</b>	<b>30</b>	<b>5.90</b>	<b>6.03</b>	<b>29</b>	<b>0.38</b>	<b>0.30</b>
	Lowland wooded	1		4.27	1		0.47
	Moorland grass mosaic	5	5.20	5.41	5	0.61	0.62
	Tall grass and herbs	5	6.57	6.83	4	1.04	0.93
	Upland wooded	4	4.61	4.72	4	-0.17	-0.05
Lithomorphic	Crops and weeds	6	6.55	6.75	6	0.56	0.26
	Fertile grassland	10	6.68	6.73	10	0.22	0.16
	<b>Heath and bog</b>	<b>19</b>	<b>4.52</b>	<b>4.59</b>	<b>19</b>	<b>0.02</b>	<b>-0.06</b>
	<b>Infertile grassland</b>	<b>19</b>	<b>6.35</b>	<b>6.82</b>	<b>19</b>	<b>0.98</b>	<b>0.76</b>
	Lowland wooded	3	7.93	7.48	3	1.04	1.16
	Moorland grass mosaic	10	4.99	5.11	9	0.11	0.16
	Tall grass and herbs	1	4.16	4.16	1		-0.04
	Upland wooded	1	4.61	4.61	1		-0.39
Peat (organic)	Crops and weeds	1	8.24	8.24	1		2.54
	Fertile grassland	1	4.77	4.77	1		-0.63
	<b>Heath and bog</b>	<b>79</b>	<b>4.47</b>	<b>4.52</b>	<b>77</b>	<b>0.27</b>	<b>0.26</b>
	Infertile grassland	2	6.11	6.11	2	1.23	1.23
	Lowland wooded	0			0		
	Moorland grass mosaic	17	4.57	4.72	14	0.48	0.53
	Tall grass and herbs	2	7.10	7.10	2	0.60	0.60
	Upland wooded	2	3.85	3.845	2	0.05	0.05
Pelosols	<b>Crops and weeds</b>	<b>9</b>	<b>6.80</b>	<b>6.60</b>	<b>9</b>	<b>-0.70</b>	<b>-0.70</b>
	Fertile grassland	1		8.33	1		0.73
	Heath and bog	0			0		
	Infertile grassland	0			0		
	Lowland wooded	1	4.40	4.40	1		-1.90
	Moorland grass mosaic	0			0		
	Tall grass and herbs	1	4.70	4.70	1		-1.80
	Upland wooded	0			0		
Podzolic	Crops and weeds	2	5.96	5.96	2	-0.05	-0.05
	Fertile grassland	5	5.67	5.71	5	1.01	0.89
	<b>Heath and bog</b>	<b>26</b>	<b>4.29</b>	<b>4.43</b>	<b>26</b>	<b>0.07</b>	<b>0.09</b>
	<b>Infertile grassland</b>	<b>21</b>	<b>5.39</b>	<b>5.50</b>	<b>21</b>	<b>0.49</b>	<b>0.42</b>
	Lowland wooded	4	4.12	4.14	4	-0.11	-0.27
	<b>Moorland grass mosaic</b>	<b>35</b>	<b>4.48</b>	<b>4.58</b>	<b>34</b>	<b>0.36</b>	<b>0.18</b>
	Tall grass and herbs	3	6.42	6.70	3	2.02	2.00
	Upland wooded	17	4.09	4.17	17	-0.16	0.01
Surface-water gley	Crops and weeds	15	7.11	6.94	15	0.45	0.71
	<b>Fertile grassland</b>	<b>45</b>	<b>6.21</b>	<b>6.19</b>	<b>45</b>	<b>0.41</b>	<b>0.41</b>
	Heath and bog	19	4.29	4.52	19	0.17	0.26
	<b>Infertile grassland</b>	<b>27</b>	<b>5.67</b>	<b>5.74</b>	<b>27</b>	<b>0.58</b>	<b>0.45</b>
	Lowland wooded	2	4.68	4.68	2	0.68	0.68
	<b>Moorland grass mosaic</b>	<b>22</b>	<b>4.41</b>	<b>4.69</b>	<b>22</b>	<b>0.25</b>	<b>0.32</b>
	Tall grass and herbs	3	7.70	7.27	3	0.20	0.13
	Upland wooded	4	4.36	4.33	4	0.56	0.57

### 6.3.8 Comparisons with other studies of long-term changes in soil pH

Berden et al (1987) noted a general global decline in soil pH from published data upto the mid 1980's. A recent re-sampling of agricultural soils in England and Wales indicated that the proportion of arable soils with pH less than 6.0 decreased between 1980 and 1995 while the proportion of grassland soils with pH less than 6.0 increased in this time (Webb et al., 2001) while re-sampling of upland and semi-natural systems in Europe by various authors (Billet et al., 1988; Adamson et al., 1996; Hallbacken and Tamm, 1986; Gustafsson et al., 1993) have all recorded decreased soil pH over 25 to 55 years from 1927 to 1991.

Two non-soil surveys suggest, however, that an overall increase in soil pH may have occurred over the last thirty years in semi-natural systems of Great Britain. The first is a significant change in vegetation structure towards more nitrophilous, less acid tolerant species from 1990 to CS2000 (Haines-Young et al., 2000) and the second is the increase in pH of waters from various aquatic surveys.

## 6.4 Conclusion

Over 1000 soil samples from CS2000 were successfully analysed for soil pH. 769 of these soil samples were obtained from the same X-plots from which the 1978 soil samples were taken. An ORACLE dataset of soil pH data from 1978 and CS2000, plus relevant metadata, has been fully integrated into the Countryside Survey data management system that is maintained at CEH Merlewood. From here the soil pH data can be linked to any other data recorded from the same 1 km square and/or the same X-plot. The soil pH dataset is available for investigating the distribution patterns of the soil biological properties and pollutants from CS2000, as well as examining patterns of pH in soils of the British Countryside. The distribution patterns of soil pH in 1978 and CS2000 indicate that appropriate data transformations would be required for parametric analyses.

Summary results produced so far indicate that patterns of soil pH from 1978 and CS2000 correspond with expected patterns. Soils of Great Britain were, in the main, predominately acidic though pH ranged from 3.2 to 8.71. The lowest soil pH values were recorded in upland soils and the highest pH was recorded from arable/horticultural areas, especially in the lowlands of England and Wales. There were, however, wide ranges in soil pH values within the different stratifications. These values warrant further examination to establish that whether they correspond to particular site factors e.g. land use, management, geology etc. or whether the site has been correctly assigned to the different stratifications e.g. major soil group.

The summary statistics from 1978 and CS2000 and the results from differences in soil pH between 1978 and CS2000 for known repeat locations all suggest that there has been an overall increase in soil pH since 1978. These results concurred with the increase in soil pH shown in some agricultural soils in GB (Webb et al., 2001) but did not in the overall decrease in soil pH in grassland soils also shown by these authors. The only decrease in soil pH from 1978 to CS2000 was recorded from coniferous woodlands.

Differences in the soil pH values between surveys and samples must be explored in greater detail before any significance can be attributed to the trends highlighted in this

report. Further analyses are required to examine whether these results are consistent with *in-situ* changes in soil pH or whether they could be the result of methodology differences and/or small-scale heterogeneity. Potential ways of examining unusual values and the validity of the overall differences in soil pH recorded between 1978 and CS2000 are:

- Examine 1978 field records to determine whether litter removal can explain differences in soil pH.
- Examine effects of small differences in sampling depth on soil pH.
- Examine small-scale spatial heterogeneity in soil pH by referring to published data and *in-situ* study of small-scale heterogeneity.
- Examine change in land use cover by referring to 1978, 1984, 1990 and CS2000 land use and vegetation data.
- Confirm X-plot allocations to Broad Habitat, ITE Land Class, Major Soil Group and AVC.

## 7. SOIL ORGANIC MATTER AND CARBON CONTENTS FROM THE ECOLOGICAL SURVEY OF 1978 AND COUNTRYSIDE SURVEY 2000

### 7.1 Introduction

Soil organic matter (SOM) is key to a healthy soil (Doran and Zieff, 2001). Both the quality and quantity of SOM play an important role in sustaining soil functions e.g. supplying nutrients, retaining water, modifying pollutants, resisting degradation or releasing/consuming greenhouse gases. There is awareness for a need to optimise management of this important, but vulnerable, resource to maintain these functions (Carter, 2001). Since the late 1800's, there have been substantial losses in SOM associated with changes in land use and management practices while the soil's capacity to store carbon is considered an important component of climate change mitigation strategies through carbon sequestration (IPCC, 1996); the carbon content of soils is principally held in SOM.

Carbon is accumulating in the atmosphere at a rate of ca. 3.5 Pg yr<sup>-1</sup>, principally from burning of fossil fuels and land use changes (Siegenthaler and Sarmiento, 1993), with the, now widely accepted, consequences for global climate changes (IPCC, 2001). Mechanisms have been introduced that aim to reduce the amount of carbon in the atmosphere e.g. lowering greenhouse gas emissions and carbon sequestration (Brubb et al., 1999). The later mechanism is, however, controversial since there are large uncertainties in the size and behaviour of global sources and sinks of carbon while significant discrepancies have been uncovered in estimating the potential for changes in land use and/or management practices to increase carbon stored in terrestrial ecosystems, and in soils in particular (i.e. for these systems to act as C sinks).

A major difficulty is in quantifying current carbon stocks due to large field-scale temporal and spatial variability. As a consequence, there is a need for soil carbon databases that will increase our knowledge of how soil carbon contents correspond to topography, habitat and, especially, management history. Habitat-specific studies have shown that there has been an increase in the soil organic matter content of soils in semi-natural and/or upland areas of Great Britain (Adamson et al., 1996; Billet et al., 1990). In contrast, however, the carbon content of agricultural soils has declined (Webb et al., 2001). What is, as yet, unclear, is whether the carbon content of the SOM itself has changed over time.

Carter (2001) summarised the outcomes of several UK studies that have highlighted major concerns with specific regard to soil organic matter and sustainability (MAFF, 1970; HMSO, 1994; RCEP, 1996) which can also be directed to soil carbon content, since SOC is closely related to SOM. These concerns are:

- Improved methods to estimate soil organic matter
- Monitor soil organic matter concentrations over time
- Establish minimum soil organic matter concentrations for different soil below which adverse effects may occur

- Protect soil as a limited resource and as an essential part of life-support systems
- Ensure land management maintains soil functions by preventing irreversible declines in soil organic matter
- Soils should be given the same priority in environmental protection as air or water
- Future surveys of land should provide a measure of soil biological quality

The publication of the RCEP Report on Sustainable Use of Soils (RCEP, 1996) was a major driver in identifying Countryside Survey 2000 (CS2000) as cost-effective framework for integrating an assessment of soil biological properties with detailed landscape, land-use, soils and vegetation data. Soil carbon content was seen as a key property. Soil organisms and soil organic matter (and soil carbon) are inextricably linked. Soil organisms are primarily responsible for the biochemical transformations of SOM as a predominant food source (Ritz and Griffiths, 2001).

SOM is also known to have a significance influence over the behaviour and local/regional scale distribution of soil biota, as well as other soil properties that also influence the behaviour of soil biota e.g. form and concentrations of metals and organic compounds. Therefore SOM data from the same sampling location was deemed an important property for the assessment of soil biodiversity as part of CS2000.

The Countryside Surveys also presented a unique opportunity to investigate long-term trends in soil organic matter and carbon contents in the British countryside. Soil organic matter content was measured, by loss-on-ignition, from soil samples taken during the first Ecological Survey of Great Britain in 1978, the same locations as those to be sampled in CS2000. Data from semi-natural and upland systems are lacking. Recent data from GB on long-term trends in SOM have been concentrated on lowland, agricultural systems (Webb et al., 2001).

### **7.1.1 Specific Objectives**

These were:

- To measure soil organic matter content in soil samples collected during CS2000, from the same locations as the 1978 Ecological Survey of Great Britain.
- To calculate carbon content in soil samples from Countryside Survey 2000
- To produce digital ORACLE GIS spatially referenced datasets for organic matter content and carbon content for the 1978 and CS2000 surveys.
- To examine the differences in soil organic matter and carbon content between 1978 and CS2000.
- To interpret change date in terms of soil type, geographical location, land use/cover and vegetation.

The following reports on the SOM results from the 1978 and CS2000 surveys across the British Countryside and with reference to broad stratifications of the countryside; Environmental Zone, Broad Habitat, ITE Land Class, CVS Aggregate Vegetation Class and Major Soil Group. Definitions and descriptions of these were presented in the earlier section on the Countryside Surveys. Methods are discussed followed by an

overview of the summary statistics produced to describe the datasets with some potential uses of the datasets highlighted.

## **7.2 Methods**

### **7.2.1 Loss-on-ignition method**

Ball (1964) proposed loss-on-ignition (LOI) as a relatively simple and inexpensive method for determining SOM, and soil organic carbon content (SOC) using an appropriate conversion equation. Since then it has been used extensively although it has attracted various criticisms with regard to the relationships of LOI to SOM and SOC. Donkin (1991) summarised the criticisms of this technique as errors introduced by a mass loss of CO<sub>2</sub> from soil carbonates, loss of elemental C; loss of hygroscopic and structural water from clay minerals and the uncertain C:SOM ratio. Another source of potential error is the difference in inter-laboratory methods, including combustion temperatures. These may lead to uncertainties in the absolute amounts of SOM and SOC but not relative comparisons of LOI results from year to year.

Mass loss of CO<sub>2</sub> from carbonates during combustion has been examined in soils and sediments by many authors. Combustion at temperatures less than 900°C ensures negligible combustion of soil carbonates (Ball, 1964; Donkin, 1991; Heiri et al., 2001; Jovilet et al. 1998). The loss of elemental C is negligible and does not contribute to the humic cycle. Loss of hygroscopic and structural water from clay minerals may be more significant factors that would improve estimates of SOC from LOI. Both can introduce small errors into LOI values, especially at low organic carbon contents. However, even when these factors are ignored, the correlation of LOI to SOC, as estimated by dry combustion methods, is typically greater than 95% (Howard and Howard, 1990; Donkin, 1991; Soon and Abboud, 1991). Adamson et al. (1996) compared LOI results from the analyses of 100 soils at 375°C and 550°C using exactly the same methods as those used during the 1978 and CS2000 surveys, respectively. These authors concluded that there were negligible differences in LOI results from analyses at these two temperatures.

### **7.2.2 Ecological Survey of 1978**

Soil sampling within the 1978 Ecological Survey has been discussed in Section 2. A copy of the original field notes and record sheet are provided in the Technical Annex. The record sheets from the field sampling contain details of the soil horizon depths, including the depth of the upper soil horizons from which the soil samples were obtained. A relatively small percentage of the soils in 1978 were sampled without the litter layer. Consequences of this are outlined further in the text below. By cross-checking the field sheets, it would be possible to determine which soil samples were sample below the litter layer; where the surface litter layer was greater than 5 cm, a soil sample taken below this layer. Loss-on-ignition was determined on all soil samples using the same method detailed below for CS2000 soil samples at a furnace temperature of 375°C for CS2000. As outlined above, this difference should not produce a significant difference between LOI determined at 550°C (Adamson et al., 1996).

The data for soil organic matter content (loss-on-ignition) from 1978 were entered into ORACLE databases in the intervening years by staff from the Land Use Section at CEH

Merlewood. These data were recovered from the Countryside Information System and other Countryside Survey datasets in 1997. A single dataset was compiled from these sources while the validity of the database results was checked against printouts from the analytical phase in 1978. Sample locations were checked against the original record sheets. The X-plots that had not changed location between 1978 and CS2000 (through 1984 and 1990 field surveys) were identified within the datasets to enable differences in SOM between 1978 and CS2000 to be carried out on the same locations. These are referred to as “repeats” in the text. The data manager entered the 1978 dataset into ORACLE, once the data checks had been completed.

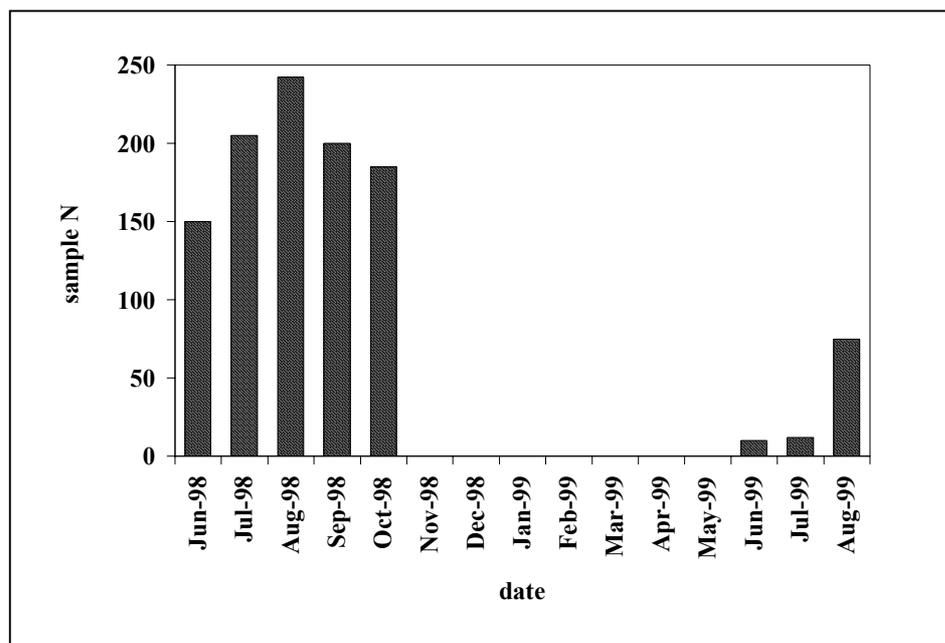
### 7.2.3 Countryside Survey 2000

Full protocols for soil sampling, initial processing and analyses for soil organic matter content by loss-on-ignition are available in the Technical Annex. The following summarises the procedures.

#### Soil Sampling

The sampling process by the CS2000 field surveyors is detailed in Section 4. In summary, where possible, the field surveyors collected an 8 cm diam. x 15 cm deep soil core from a specific location in each X-plot specific for every 1 km squares originally sampled in the 1978 Ecological Survey. Each core was stored in a labelled, un-sealed plastic bag in a cool-box and returned to CEH Merlewood by the field surveyors or by the field co-ordinators at regular intervals (see Figure 7.1).

Figure 7.1. Histogram showing the processing rate of soil cores for chemical analyses at CEH Merlewood during CS2000.



### **Initial soil processing**

Project staff logged soil cores in the daily diary on their arrival at CEH Merlewood, along with any notes provided by the field surveyors. The soil samples were removed from their cores and processed to obtain an air-dried 2 mm sieved soil sample in preparation for further analyses and long-term storage. Each sample was weighed at all stages in the initial processing while soil pH was obtained in the process. Soil organic carbon content was then determined on homogenised sub-samples of this air-dried sample by loss-on-ignition.

### **Loss-on-ignition**

Loss-on-ignition analyses were carried out in the laboratories of the Environmental Chemistry Section at CEH Merlewood using their standard method, including furnace combustion at 550°C (Allen et al., 1989). Samples were analysed in batches of 25, 23 MASQ samples and two standard materials for quality control.

### **Quality control**

Loss-on-ignition analyses were included in the Environmental Chemistry Section's laboratory QC procedures. As stated above, all analytical batches contained two standard materials that were cross-checked after analyses to validate the data. There were no significant deviations in the standard materials for the analyses of LOI of the MASQ samples. Results from LOI of the standard materials are included in the ORACLE SOM datasets for future reference.

#### **7.2.4 Reproducibility of methods**

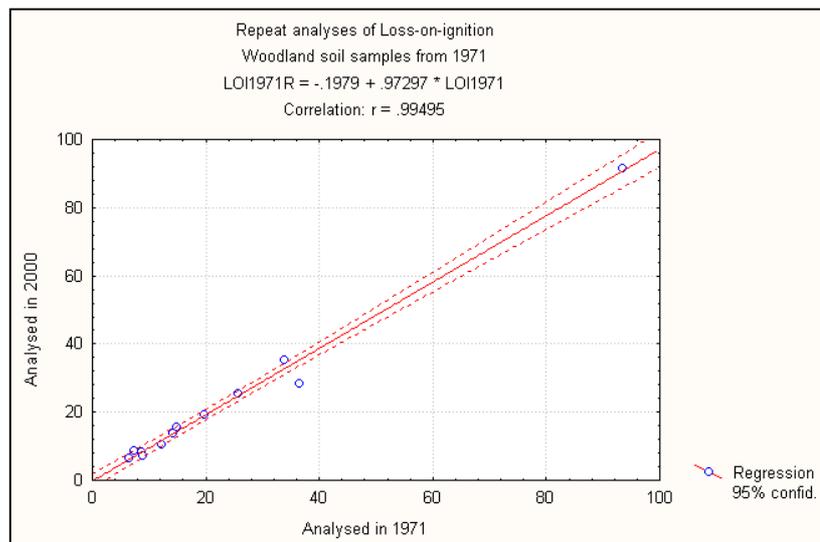
Soil samples from the 1978 Ecological Survey were, unfortunately, lost in the intervening period. It was, however, possible to use another set of long-term soil samples stored at CEH that were obtained from a range of woodland sites across Great Britain in 1971. The soil samples were taken in a similar manner to the Ecological Survey of 1978 and the methods used loss-on-ignition were the same as those used for the CS2000 soil samples (Smart et al., 2000). In 2000, a sub-set of these samples was identified across the range of original LOI values and these were re-analysed for LOI using the same methods as the MASQ project. The results were plotted against the original 1971 analyses (Figure 7.2). Both analyses give highly comparable results and there were no statistically significant differences between these samples (dependent sample T-test;  $P > 0.005$ ).

#### **7.2.5 Data management and analyses**

All data from CS2000 were entered into log-sheets, from where they were transferred to Excel spreadsheets by project staff. These datasheets were checked by the project data manager and/or another person, with summary statistics carried out to check for errors (e.g. distribution graphs, max, mean, min.). Approximately 10% of all Excel data were checked against the log-sheets. Once completed, the final Excel spreadsheets were sent to the data manager for transfer into the MASQ database, along with relevant meta-data. These datasets are maintained in an ORACLE database at CEH Merlewood.

All data are archived on a regular basis and copies maintained off-site under secure conditions. Summary datasets for loss-on-ignition were extracted from ORACLE. Results from CS2000 were rounded up for the calculation of SOM differences between 1978 and CS2000. Summary statistics on SOM produced in tabular form using SAS and Statistica software maintained, under license, by CEH.

Figure 7.2. Repeat analyses in 2000 of loss-on-ignition from woodland soils collected in 1971 plotted against the original loss-on-ignition from 1971. The methods were the same as those used in MASQ and the 1978 Ecological Survey, respectively.



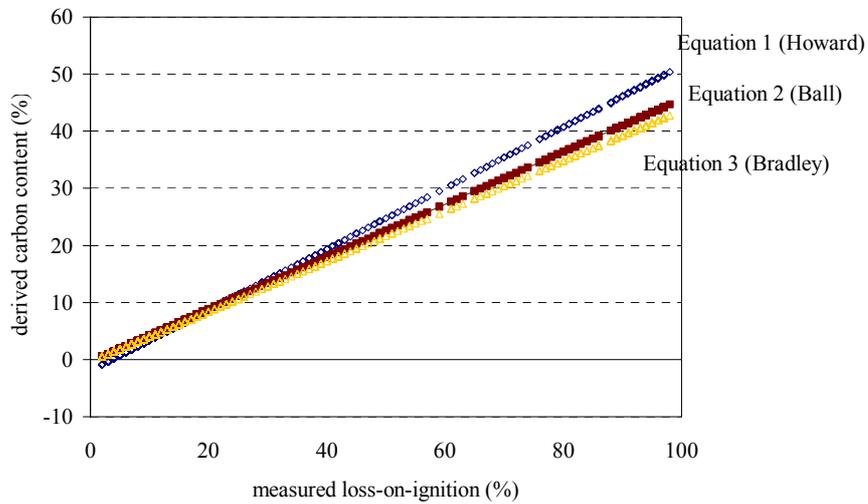
### 7.2.6 Conversion of loss-on-ignition data to carbon content

An objective of this project was to examine differences in soil organic carbon content (SOC) over time. This required the derivation of soil carbon contents from loss-on-ignition analyses using a suitable conversion equation. Howard and Howard (1990) highlighted that a single conversion factor to convert LOI to SOM has been questioned. Problems with the conversion equations may be due to differing C contents of SOM, as discussed by Donkin (1991), but also to differences in the carbon content of SOM between different soils. Various authors have developed conversion equations suitable for specific soils. Figure 7.3 presents the results of converting the 1978 and CS2000 loss-on-ignition data to SOC using three equations that were developed for soils of Great Britain (Howard and Howard, 1990; Ball, 1964 and Bradley, pers. comm.); only the repeat samples were used.

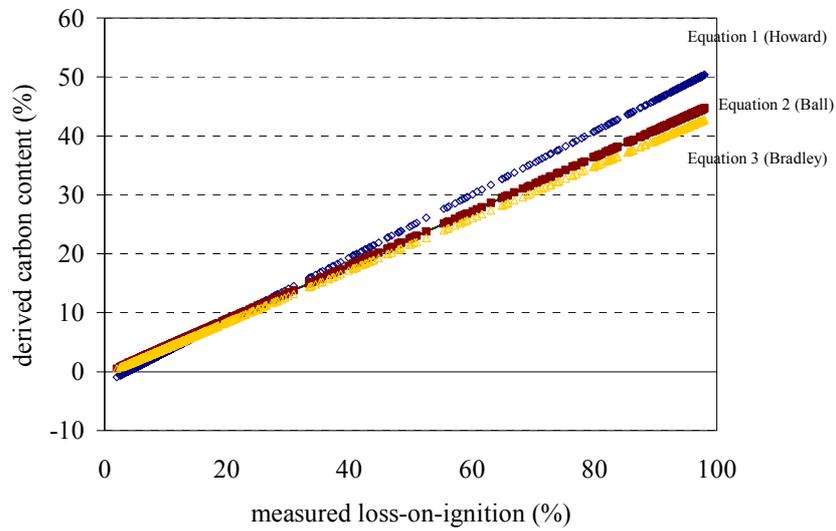
These results show that there are distinct differences in the results obtained from the three equations, especially at higher levels of loss-on-ignition. The Ball and Bradley equations offer the best-fit equations as they do not introduce negative C contents at lower LOI values. The production of summary statistics of SOC would be inappropriate using only one of these equations and, therefore, it is proposed that a more accurate estimate of SOC from the 1978 and CS2000 soil samples should be obtained. Countryside Survey specific equation(s) could be derived from the analyses of total carbon, and possibly clay content, in a sub-set of soil samples across the range of LOI values within all Major Soil Groups.

Figure 7.3. Derived soil carbon contents plotted against measured soil organic matter contents for 1978 and CS2000 soil samples.

**Organic carbon content of 1978 Ecological Survey soil samples derived from three different equations plotted against measured loss-on-ignition**



**Organic carbon content of CS2000 soil samples derived from three different equations plotted against measured loss-on-ignition**



## 7.3 Results

### 7.3.1 National Over-view

A total of 1122 and 1023 analyses were obtained for soil organic carbon content from 1978 and CS2000, respectively. A total of 744 X-plots were repeated from 1978 to CS2000 (Table 7.1). Summary statistics are presented in Table 7.1. Results are presented for all SOM data from 1978 and CS2000 and for two sub-sets of these data. The first is for SOM data from 1978 and CS2000 where the X-plots maintained the same location from 1978 to CS2000 (“repeats”) and the second sub-set is for 1978 and CS2000 SOM data where the X-plots maintained the same location from 1978 to CS2000 (“repeats”) and with X-plots with large variation from 1978 to CS2000 removed, as discussed below. Sample numbers were less than optimal in both years for various reasons. Samples may not have been collected due to sampling difficulties on site or collected from the wrong location while insufficient soil may have been collected for chemical.

Table 7.1. Summary statistics for soil organic matter content of soil samples collected in 1978 and CS2000 and for the difference\* between these two surveys.

year	DATASET	Sample N	Mean	Median	Minimum	Maximum	Std.Dev.
1978	all data	1122	26.70	10	2	98	31.79
CS2000	all data	1067	29.11	12.57	2	98.02	31.95
DIFFERENCE	all data	1023	2.84	2.09	-84.64	88.14	19.63
1978	repeats only	812	27.96	10	2	98	32.25
CS2000	repeats only	778	30.46	13.3	2	98.02	32.52
DIFFERENCE	repeats only	744	3.37	2.27	-84.64	88.14	20.04
1978	Sub-set <sup>1</sup>	640	25.02	9	2	98	31.66
CS2000	Sub-set	640	27.22	12.16	2.6	98.02	31.41
DIFFERENCE	Sub-set	640	2.21	2	-24	24	6.67

\*Difference results calculated using rounded CS2000 data

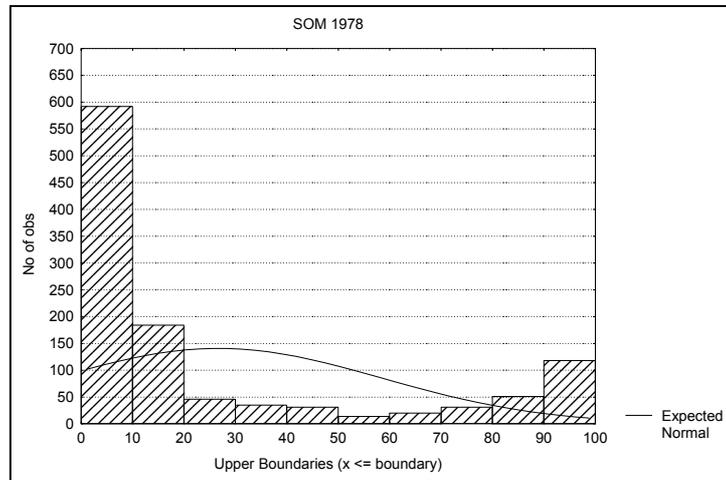
<sup>1</sup>Sub-set of repeat values derived from removal of extreme LOI values, see text for explanation.

The results indicate that median values for soil organic matter (SOM) were typically <14% while the values ranged from 2 to 98% for all samples (Table 7.1). The median soil organic matter content value was higher in CS2000 compared to 1978 although there was no difference in the minimum or maximum values. The same trends are present in the data from the repeat X-plots only, although the mean and median values were slightly higher in the repeats dataset for both years. The distribution plots (Figures 7.4 and 7.5) show little difference in the distribution of SOM values between the two surveys and highlights that the summary statistics were strongly influenced by the bi-modal distribution of the 1978 and CS2000 datasets.

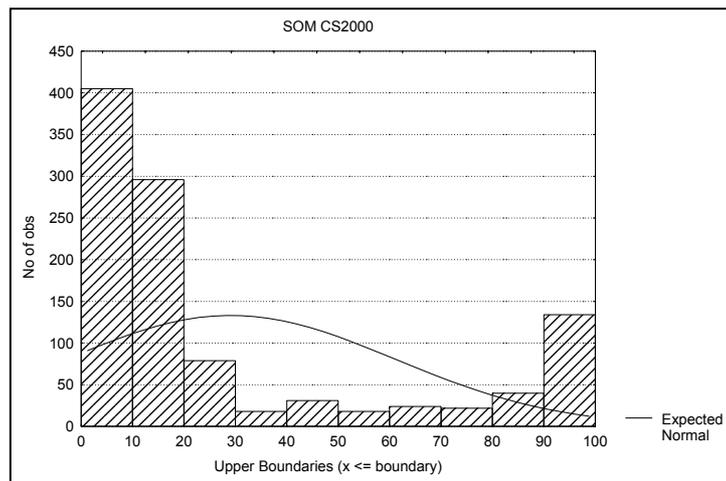
Regression analyses showed that there was no correlation between SOM values from 1978 and CS2000 from the same X-plots that maintained their location. Closer examination of the data indicates that there were large differences in SOM values between the two surveys for a limited number of X-plots.

Figure 7.4. Frequency histograms of soil organic matter contents; (a) 1978, (b) CS2000 and (c) difference between 1978 and CS2000 using a sub-set of 640 samples

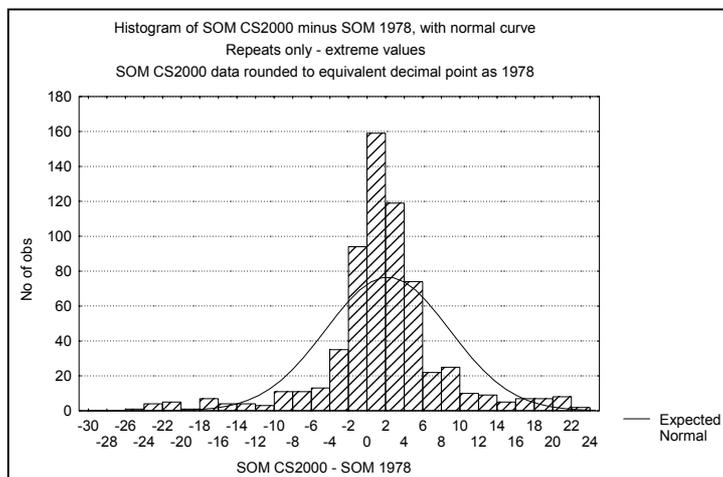
(a) 1978



(b) CS2000



(c) difference between 1978 and CS2000 for sub-set of 640 samples

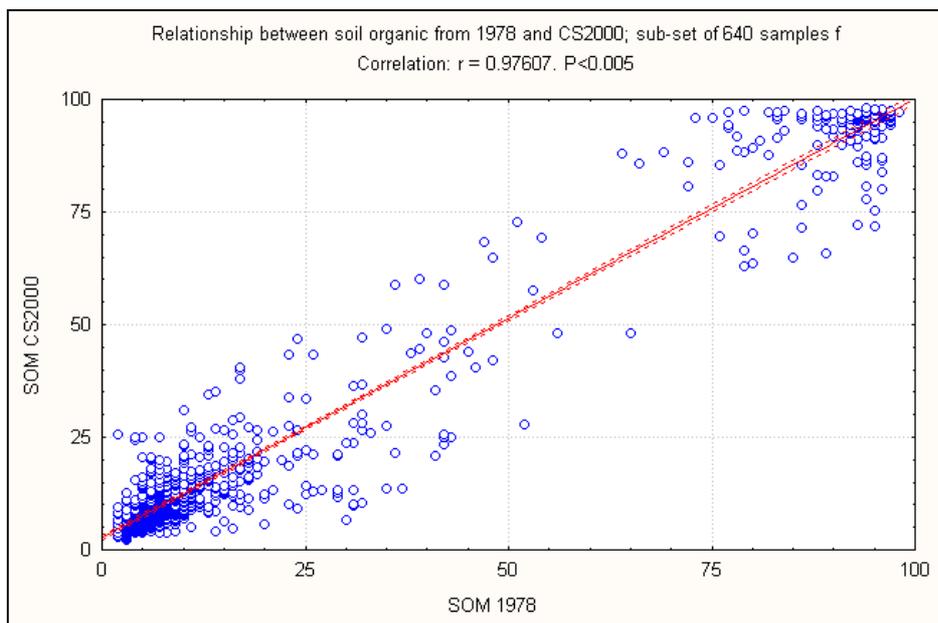


Although these values do not alter overall datasets from 1978 and CS2000, they influence the difference between the two surveys and may, in some instances, influence the summary statistics at the different levels of stratification. Possible reasons for this variability are outlined below.

For the purposes of reporting on the MASQ datasets, a sub-set of the 1978 and CS2000 data were identified to examine trends in SOM differences between the two surveys. **It must be stressed, however, that these results can only be substantiated once the variation between the 1978 and CS2000 datasets have been resolved.**

This sub-set was identified by carrying out a series of regression analyses between the 1978 and CS2000 SOM and pH, datasets to determine at which point a significant correlation between 1978 and SOM could without significantly affecting either the SOM or soil pH summary statistics. As a consequence, differences in SOM between 1978 and CS2000 were reported here on a sub-set of 640 data points where the SOM difference between 1978 and CS2000 was greater than -25% and less than 25%. (Figure 7.5 and Table 7.1). The distribution plot of the differences in SOM values between the two surveys for this sub-set was normally distributed (Figure 7.4c).

Figure 7.5. Scatterplot of soil organic matter from 1978 plotted against soil organic matter from CS2000 (1998/9) for repeat samples with extreme values excluded.



Soil pH was included to determine whether the variation in SOM between 1978 and CS2000 would influence the soil pH results. Analyses indicated that this was not the case and therefore summary statistics of soil pH was carried out on the full dataset of repeat X-plots.

Table 7.1 indicates that the median difference in soil organic matter between 1978 and CS2000 was c. 2%. This difference was significant using the sub-set of 640 samples (Table 7.2); T-test for dependant samples. The frequency histogram of differences in SOM (Figure 7.4c) shows that the majority of samples fall between -4% to +6%

difference in SOM between the two surveys, with most samples indicating no difference or a relatively small increase in SOM.

Table 7.2. Results from T-test for dependant samples between soil organic matter content from 1978 and CS2000; repeat samples with unusual values excluded.

Year	Mean	Std.	N	Diff	s.d.	T	d.f.	P
1978	25.02	31.66						
2000	27.23	31.39	640	2.21	6.67	8.38	639	.00000

The following sections present summary statistics and results for five different stratifications of the British countryside; Environmental Zone, Broad Habitat, ITE Land Class, Aggregate Vegetation Class and Major Soil Group, respectively, to illustrate the range and variability in soil organic matter contents across the British countryside. Results are presented for the complete datasets for 1978 and CS2000 and for the sub-set of 640 samples from the difference in SOM between the two surveys, as discussed above.

### 7.3.2. Environmental Zones

Soil organic matter content results from 1978 and CS2000 (Tables 7.3, 7.4 and 7.5) follow expected trends with the lowest mean and median values in EZ 1, 2 and 4, areas predominately lowland + agricultural, and highest values in EZ 3, 5 & 6, the upland areas. The maximum SOM in EZ1 was much lower than SOM values in all other Zones. The maximum values were higher in CS2000, especially in EZ 1 and 2 while the minimum value was much higher in CS2000 than 1978 in EZ3. Differences in SOM between 1978 and 2000 were mostly positive in all zones except EZ 6, as reflected in the mean and median values with the highest increase recorded in EZ 5, followed by EZ 2 and 3 (Table 7.5).

Table 7.3. Summary statistics for soil organic matter in 1978 by Environmental Zone.

Environmental Zone	Sample N	Mean	median	min	max	std
1	247	7.27	6	2	41	5.7
2	282	10.54	7	2	83	10.83
3	117	30.87	15	4	96	30.64
4	149	16.02	8	2	96	22.25
5	179	56.92	70	2	97	36.7
6	148	60.85	72	4	98	31.71

Table 7.4. Summary statistics for soil organic matter in CS2000 by Environmental Zone

Environmental Zone	Sample N	Mean	median	min	max	std
1	229	9.07	7.77	2.6	46.18	5.76
2	270	13.74	10.11	2	92.76	13.49
3	117	32.42	19.1	6.51	96.4	28.43
4	143	19.98	9.6	2.38	97.37	25.23
5	173	59.71	76.53	4.45	98.02	36.73
6	135	61.45	71.45	4.83	97.61	32.73

Table 7.5. Summary statistics for difference in soil organic matter between 1978 and CS2000 by Environmental Zones for 640 repeat X-plots only

Environmental Zone	Sample N	Mean	median	min	max	std
1	123	1.89	2	-14	16	3.7
2	196	2.81	2	-23	22	4.82
3	68	3.47	3	-21	23	8.11
4	88	3.3	2.5	-18	21	5.6
5	105	1.21	2	-23	24	8.81
6	60	-0.37	-0.5	-24	20	10.27

### 7.3.3 Broad Habitats

Summary statistics are presented in Tables 7.6, 7.7 and 7.8. Results are discussed for the Broad Habitats with more than 30 samples (sample N ranges from 1 to 320). Future analyses could consider combinations of Broad Habitats to increase sample N for statistical analyses. Most samples were obtained from improved grassland and arable/horticultural, followed by bog in both surveys. Nine Broad Habitats contained more than 30 samples. In the Broad Habitats with more than 10 samples, the highest median values in SOM were recorded in the habitats which would promote accumulation of SOM; bog, dwarf shrub heath, acid grassland, coniferous woodland and fen, marsh/swamp (median 35 to 82%). The lowest SOM was recorded in arable/horticultural areas, in both years, while SOM in neutral and improved grassland and broad-leaved woodland and bracken was intermediate to these two groups (median 8 to 19%). Median and mean differences in SOM between 1978 and CS2000 showed an increase in SOM in all habitats with more than 10 samples, although there was a wide range in difference results (Table 7.8). Median differences ranged from 1 to 5% across the Broad Habitats with the greatest difference in acid grassland and least in dwarf shrub heath, fen marsh swamp and arable/horticultural areas.

Table 7.6 Summary statistics for soil organic matter contents in 1978 by Broad Habitats

Broad Habitat	Sample N	mean	median	min	max	st. dev
Boundary and linear features	1	9.00	9	9	9	.
Inland rock	1	38.00	38	38	38	.
Supralittoral sediment	1	10.00	10	10	10	.
Supralittoral rock	2	52.00	52	12	92	6.57
Calcareous grassland	6	16.83	16	6	32	8.7
Littoral sediment	7	8.14	9	2	14	5.4
Built-up areas and gardens	8	9.00	7.5	2	20	6.39
Bracken	19	25.74	19	5	90	22.07
Fen, marsh and swamp	39	35.85	24	5	96	29.33
Neutral grassland	42	12.57	9	4	41	9.66
Acid grassland	65	49.54	43	4	97	32.85
Broadleaved, mixed and yew woodland	67	16.40	8	2	92	20.54
Coniferous woodland	84	41.81	28.5	4	97	33.93
Dwarf shrub heath	97	55.28	65	3	97	34
Bog	141	76.22	90	2	98	27.17
Arable and horticultural	249	5.69	5	2	39	3.28
Improved grassland	320	10.32	8	2	92	8.99

Table 7.7 Summary statistics for soil organic matter contents in CS2000 by Broad Habitats; all X-plots

Broad Habitat	Sample N	mean	median	min	max	st. dev
Boundary and linear features	1	6.47	6.47	6.47	6.47	.
Inland rock	1	13.30	13.30	13.3	13.30	.
Supralittoral sediment	1	11.50	11.50	11.5	11.50	.
Supralittoral rock	3	77.79	94.62	43.25	95.49	29.91
Calcareous grassland	6	28.47	27.27	22.67	36.61	5.44
Littoral sediment	6	8.36	5.99	4.18	15.38	5.15
Built-up areas and gardens	10	10.30	9.64	5.69	17.42	3.84
Bracken	18	29.78	15.79	6.89	95.37	28.53
Fen, marsh and swamp	39	37.88	27.32	4.83	97.03	29.62
Neutral grassland	43	13.58	10.13	2	49.36	10.42
Broadleaved, mixed and yew woodland	62	22.20	13.58	3.72	97.43	23.4
Acid grassland	64	49.65	40.28	6.35	97.29	32.18
Coniferous woodland	84	45.59	35.45	3.04	97.51	33.56
Dwarf shrub heath	93	58.22	62.73	7.24	98.02	30.64
Bog	126	81.68	93.33	11.36	97.89	24.51
Arable and horticultural	227	6.79	6.28	2.38	17.13	2.68
Improved grassland	310	12.32	10.79	3.18	92.50	8.05

Table 7.8 Summary statistics for difference in soil organic matter between 1978 and CS2000 for each Broad Habitat for 640 repeat X-plots only

Broad Habitat	Sample N	mean	median	min	max	st. dev
Acid grassland	26	4.00	5	-23	22	12.77
Arable and horticultural	116	1.44	1	-2	8	1.81
Bog	73	1.70	2	-23	24	8.42
Boundary and linear features	1	-3.00	-3	-3	-3	.
Bracken	11	1.18	2	-8	17	7.18
Broadleaved, mixed and yew woodland	34	5.44	3	-1	18	5.25
Built-up areas and gardens	6	0.17	2.5	-14	7	7.83
Calcareous grassland	4	8.75	8.5	5	13	4.35
Coniferous woodland	36	1.36	2	-24	21	11.26
Dwarf shrub heath	38	0	1	-23	23	13.26
Fen, marsh and swamp	23	2.70	1	-8	21	7.58
Improved grassland	249	2.52	3	-14	21	4.27
Neutral grassland	31	1.16	3	-21	21	8.27
Supralittoral rock	1	3.00	3	3	3	.
Acid grassland	26	4.00	5	-23	22	12.77
Arable and horticultural	116	1.44	1	-2	8	1.81
Bog	73	1.70	2	-23	24	8.42

### 7.3.4 ITE Land Classes

Summary statistics for 1978, CS2000 and the difference in SOM between 1978 and CS2000 are presented in Table 7.9. Of the 40 ITE Land Classes, 28 contained more than 20 SOM results from 1978 and CS2000. Future analyses could consider appropriate combinations of ITE Land Classes to increase sample N for statistical analyses. In both years, the lowest SOM values were recorded in ITE Land Class 3e and the highest in two Scottish Land Classes, 21s and 30s. The differences in SOM between the two

surveys indicated an increase in SOM in all but three Land Classes. Again there was a wide range in the differences within Land Classes.

Table 7.9. Summary statistics for soil organic matter content from 1978, CS2000 and difference between 1978 and CS2000 by ITE Land Class.

ITE LC	1978						CS2000						Difference 640 repeat X-plots only					
	N	mean	median	min	max	sd	N	mean	median	min	max	sd	N	mean	median	min	max	Sd
18s	5	60.2	63	24	90	28.2	3	41.8	21.7	12	91.7	43.5	2	0	0	-2	2	2.828
19s	5	40.8	14	12	86	37.7	5	45.6	26.8	15.1	91.7	35.8	4	3	3.5	-9	14	9.42
25e	9	7.33	6	5	13	2.6	9	10.7	8.87	5.84	20.8	4.78	9	3.56	4	0	8	2.46
17w1	10	12.2	8.5	5	41	10.8	10	12.3	11.2	9.17	20	3.23	9	3.44	2	-1	9	2.88
5e	11	9.27	7	5	26	6.15	11	10.5	8.21	5.73	21.2	4.9	9	1.22	1	-5	5	2.91
22e	14	55.1	46	15	96	30.4	14	57.3	54.4	11.9	95.7	32	6	7.83	5.5	0	17	7.41
13s	15	51.9	54	5	96	38.2	15	55.9	59.2	9.6	96.9	34.4	9	9.22	7	1	20	7.17
19e	15	38.3	15	6	96	38.6	15	38.5	17.5	8.67	95.2	34.9	5	1.8	2	-6	11	7.4
23e	15	59	65	13	94	32.2	15	70.4	79.8	25.8	96.4	27.1	6	2.33	0.5	-8	23	11.31
17w3	18	25.5	14	8	88	23.3	18	19	17.5	10.8	40.4	7.46	12	1	2.5	-17	9	6.3
4e	19	9.63	6	3	41	9.91	14	9.2	8.61	3.92	20.9	4.33	10	0.7	2	-8	4	3.68
12e	20	7.35	5.5	2	39	7.96	20	8.6	8.86	2.97	17.1	3.73	7	0.71	1	-1	2	1.11
17e	20	22.1	12	4	95	25.8	20	21.6	17.5	9.37	55.9	11	15	5.87	5	-8	20	8.39
15e	25	14.8	11	4	53	10.3	17	13.7	12.5	7.48	30.9	5.41	13	2.15	2	-6	21	6.38
18e	25	14.3	10	5	73	14.9	25	18.4	15	6.51	68.6	13.5	15	2.33	3	-21	13	10.26
7s	25	9.68	9	3	32	6.28	21	14.1	10.8	5.09	47.3	10.4	13	3.77	3	-1	11	3.72
31s	26	38.7	31	2	96	34.2	32	43	28	5.18	96.5	33.9	13	-2.38	-3	-23	24	11.49
13e	27	7.85	6	3	31	5.48	27	12.1	8.75	5.22	74.3	13.1	23	2.17	2	-7	7	2.67
1e	28	10.1	9.5	3	20	4.57	28	15	13.2	7.27	49.8	8.2	23	3.7	3	-2	14	3.7
32s	29	60.1	86	5	97	39.3	29	65.4	94.1	7.58	97.9	39.9	22	2.27	2	-4	11	2.83
16e	30	10.5	9	2	25	5.62	30	14.5	11.8	2	69.1	12.6	23	3.26	3	-8	21	6.38
26s	31	8.23	7	4	40	6.48	32	11	8.14	4.74	48.1	8.22	19	3.11	2	-1	9	2.69
8e	31	10.9	6	2	45	12.8	31	14.4	10.3	3.63	50.5	14.1	21	2.71	3	-4	7	2.69
23s	34	60.1	73	8	97	33.1	27	60.5	67	7.79	97.5	31.7	7	-9.43	-16	-23	9	13.4
22s	35	54.3	56	4	95	32.1	32	58.2	65.6	4.83	96.6	33.9	13	0.23	0	-24	17	11.97
30s	35	85	93	29	97	18.3	27	90.7	96.1	40.6	98	13.5	14	3.71	2	-3	21	6.56
28s	36	35.9	18	2	97	36.4	36	42.1	21.3	4.45	97.6	35.9	29	0.59	2	-20	21	9.75
27s	37	15.6	7	2	95	24.6	34	21.4	9.25	2.38	97.4	30.6	22	2.59	2	-11	10	4.15
7e	37	8.14	7	2	24	5.5	39	10.8	7.82	3.33	43.3	8.47	23	2.61	2	-4	12	3.83
21s	39	75.4	92	13	98	27.7	36	69.3	86.7	8.73	97.6	33.2	23	0.52	1	-17	20	7.54
6e	39	16.3	9	5	83	19.4	35	22.4	11.7	6.28	92.8	23.1	23	5.61	3	-2	22	6.83
24s	40	53	51	7	96	30	40	57.7	63.8	12.1	97.3	32	17	1.71	-1	-15	20	9.7
25s	41	13	8	3	55	12.5	41	15.7	8.47	2.57	86.3	19.5	25	1.68	1	-18	21	7.25
2e	42	9.98	8	2	32	6.65	45	13	10.6	3.75	36.6	7.57	27	3.07	3	-11	16	5.04
29s	43	62	75	9	96	33	41	66.8	80.7	8.2	97.4	32.5	23	1.17	1	-23	22	10.73
9e	49	6.51	5	2	20	3.84	45	9.73	8.07	3.51	46.2	7.26	24	2.33	2	-14	12	4.72
3e	51	4.88	4	2	14	2.29	47	5.71	5.03	2.78	12.1	2.03	20	1.1	1	-2	6	2
10e	54	7.7	6	3	70	9.52	52	10.1	7.53	3.33	76.4	11.4	38	1.47	2	-23	6	4.69
11e	57	7.23	6	2	38	5.1	49	7.95	7.64	2.6	17.1	3.3	26	1.04	1	-3	7	2.22

### 7.3.5 CVS Aggregate Vegetation Class

Summary statistics for 1978, CS2000 and the difference in soil organic matter content between 1978 and CS2000 are presented in Tables 7.10, 7.11 and 7.12 and Figure 7.6. Figure 7.6 illustrates that there is a wide range in soil organic matter content values in all Aggregate Vegetation Classes for 1978 and CS2000, with outliers in several classes, especially in upland woods, heath/bogs and moorland grass mosaics. These values require further analyses to determine whether they are representative of the classes they

are currently recorded against. In 1978 and CS2000, the highest median SOM results occurred in heath and bogs (>89%) followed by moorland grass mosaics (>30). The lowest median SOM results were recorded in crops and weeds, tall grass and herbs and fertile grasslands (5 to 10%). In all AVC's, except moorland grass mosaics, the median SOM values were higher in CS2000 than in 1978; the opposite was recorded in moorland grass mosaics (Figure 7.6). Summary statistics from the differences in SOM between 1978 and CS2000 did not show this trend in moorland grass mosaics. The results showed an increase in all median values with the greatest increase recorded in lowland wooded areas

Table 7.10. Summary statistics for soil organic matter content in 1978 by CVS Aggregate Vegetation Class

AVC	Code	Sample N	mean	median	min	max	st. dev
Crops and weeds	1	201	5.65	5	2	39	3.45
Tall grass and herb	2	63	8.95	6	2	45	10.09
Fertile grassland	3	200	8.75	7	2	42	5.45
Infertile grassland	4	215	13.73	9	3	94	14.26
Lowland wooded	5	27	15.48	9	3	83	19.96
Upland wooded	6	77	26.30	16	2	97	26
Moorland grass mosaics	7	136	46.89	42	2	97	32.24
Heath and bog	8	203	72.61	89	4	98	29.54

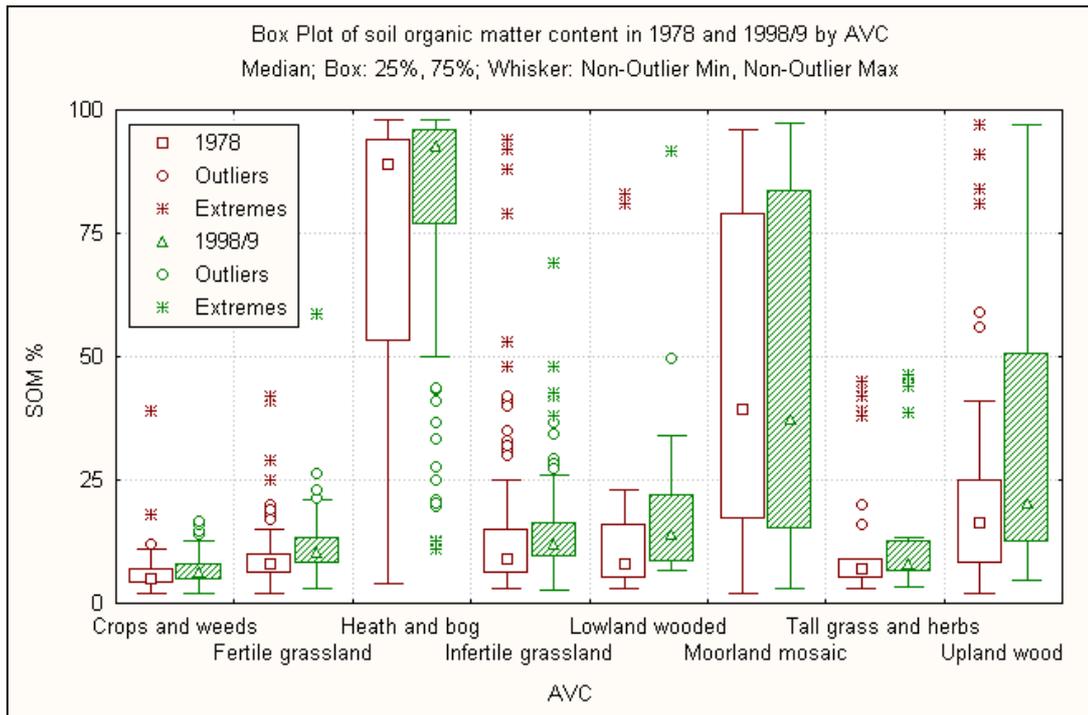
Table 7.11. Summary statistics for soil organic matter content in CS2000 by CVS Aggregate Vegetation Class; all data

AVC	Code	Sample N	mean	median	min	max	st. dev
Crops and weeds	1	185	6.59	6.09	2	17.13	2.67
Tall grass and herb	2	59	10.73	7.19	3.34	50.47	10.85
Fertile grassland	3	192	10.97	9.95	2.92	58.78	5.47
Infertile grassland	4	212	14.31	11.73	2.57	69.08	8.89
Lowland wooded	5	29	18.57	13.84	6.72	91.52	16.88
Upland wooded	6	69	33.59	20.27	4.74	97.05	27.85
Moorland grass mosaics	7	137	45.41	30.9	3.04	97.43	31.7
Heath and bog	8	184	81.47	92.56	11.02	98.02	22.38

Table 7.12. Summary statistics for difference in soil organic matter content between 1978 and CS2000 by CVS Aggregate Vegetation Class for 640 repeat X-plots only

AVC	Code	Sample N	mean	median	min	max	st. dev
Crops and weeds	1	103	1.27	1	-3	8	1.75
Tall grass and herb	2	26	0.77	1	-14	6	4.13
Fertile grassland	3	142	2.67	3	-13	17	3.85
Infertile grassland	4	166	3.01	3	-20	21	5.29
Lowland wooded	5	16	5.50	4.5	0	16	4.55
Upland wooded	6	30	2.07	2	-23	21	9.35
Moorland grass mosaics	7	56	1.05	2	-23	24	13.28
Heath and bog	8	101	1.74	2	-24	21	8.88

Figure 7.6. Box-plots of soil organic matter content from 1978 and CS2000 (1998/9) by Aggregate Vegetation Class.



### 7.3.6 Major Soil Groups

Summary statistics for 1978, CS2000 and difference in soil organic matter between 1978 and CS2000 are presented in Tables 7.13, 7.14 and 7.15 and Figure 7.7. The number of samples in each Group reflects the predominance of the Groups in the British Countryside e.g. Pelosols contain relatively few samples (<24) compared to the other Soil Groups. This group, however, is relatively uncommon in Great Britain.

Both surveys highlight the wide range in soil organic matter values in all Major soil groups for 1978 and CS2000 with outliers in several classes (Figure 7.7). The values require further analyses to determine whether these samples are truly representative of the class they are currently recorded against.

The highest median SOM values were recorded, as would be predicted, in the Peat (organic) soils in both surveys; mean, min and max. values were also higher than in all other soils. There were relatively high SOM contents in lithomorphous and podzolic soils (19 to 24%). These values may reflect the relatively shallow sampling depth (0 - 15 cm). The lowest SOM values were recorded in pelosols and brown earths in both surveys. In general, median SOM values were slightly higher in CS2000 than 1978, especially for lithomorphous and podzolic soils.

This trend concurs with the summary statistics from the difference in SOM between 1978 and CS2000 for repeat sample locations. In all soils, the median results indicate an increase in SOM, ranging from 0.5 to 3%. The highest increases in median SOM values occurred in ground-water gleys, lithomorphous and surface-water gley soils.

Table 7.13. Summary statistics for soil organic matter content from 1978 by Major Soil Group

Major Soil Groups	Code	Sample N	mean	median	min	max	st. dev
Lithomorphic Soils	3	72	36.22	19	4	96	30.92
Pelosols	4	15	7.07	6	4	14	2.66
Brown Soils	5	261	8.82	7	2	59	6.78
Podzolic Soils	6	121	32.97	19	2	95	29.89
Surface-water Gley Soils	7	138	24.67	9	2	95	28.75
Ground-water Gley Soils	8	100	11.91	8	2	77	11.37
Peat (organic) soils	10	105	86.7	93	24	98	17.18

Table 7.14. Summary statistics for soil organic matter content from CS2000 b Major Soil Group

Major Soil Groups	Code	Sample N	mean	Median	Min	max	st. dev
Lithomorphic Soils	3	96	37.50	22.29	2.57	97.43	3.24
Pelosols	4	24	7.34	6.67	4.29	15.51	0.49
Brown Soils	5	347	12.64	8.67	2.38	94.55	0.75
Podzolic Soils	6	153	39.35	24.05	4.74	97.44	2.57
Surface-water Gley Soils	7	191	26.24	12.06	3.83	97.33	2.12
Ground-water Gley Soils	8	126	11.91	9.56	2	94.19	0.98
Peat (organic) soils	10	130	79.75	94.27	6.89	98.02	2.35

Figure 7.7. Box-plots of soil organic matter from 1978 and CS2000 (1998/9) by Major Soil Group.

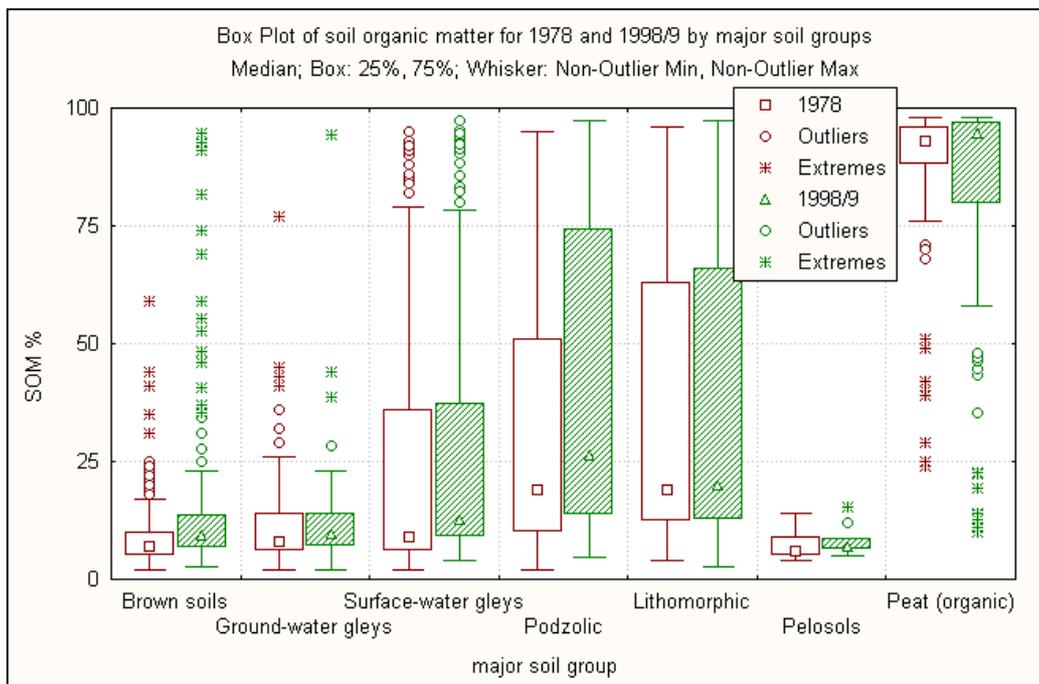


Table 7.15. Summary statistics for difference in soil organic matter contents between 1978 and CS2000 by Major Soil Group for 640 repeat X-plots only

Major Soil Groups	Code	Sample N	mean	median	min	max	st. dev
Lithomorphic Soils	3	54	2.67	3	-23	21	7.83
Pelosols	4	12	0.83	0.5	-3	7	2.72
Brown Soils	5	223	2.75	2	-21	21	4.8
Podzolic Soils	6	75	2.61	3	-24	24	10.42
Surface-water Gley Soils	7	108	3.05	3	-23	21	6.53
Ground-water Gley Soils	8	90	0.94	1	-16	17	4.69
Peat (organic) soils	10	78	0.46	1	-23	23	7.95

### 7.3.7 Analyses of soil organic matter by Major Soil Group and Aggregate Vegetation Class

The median values for 1978, CS2000 and differences in soil organic matter content between 1978 and CS2000 from individual AVCs within each Major Soil Group are presented in Table 7.16. Summary statistics for SOM of the Major Soil Groups recorded in the infertile grassland AVC are shown in Figure 7.8. These results were produced to investigate the potential for analyses within individual Major Soil Groups and AVCs. The numbers of samples in each sub-division highlight characteristic AVC classes within each Major Soil Group (Table 7.16). Several of these sub-divisions have sufficient numbers of soil samples to carry out statistical analyses on SOM data, and other soil properties. The majority, however, have fewer than 20 samples. These sub-divisions could be amalgamated by soil, AVC and/or SOM content to increase N for further analyses. In addition, these should be checked to determine whether the Major Soil Group and/or the AVC were correctly assigned. These results further highlight that the difference in SOM showed a small increase in most sub-divisions between 1978 and CS2000.

Figure 7.8. Box-plot of soil organic matter for 1978 and CS2000 for Aggregate Vegetation Class Infertile grassland by Major Soil Group.

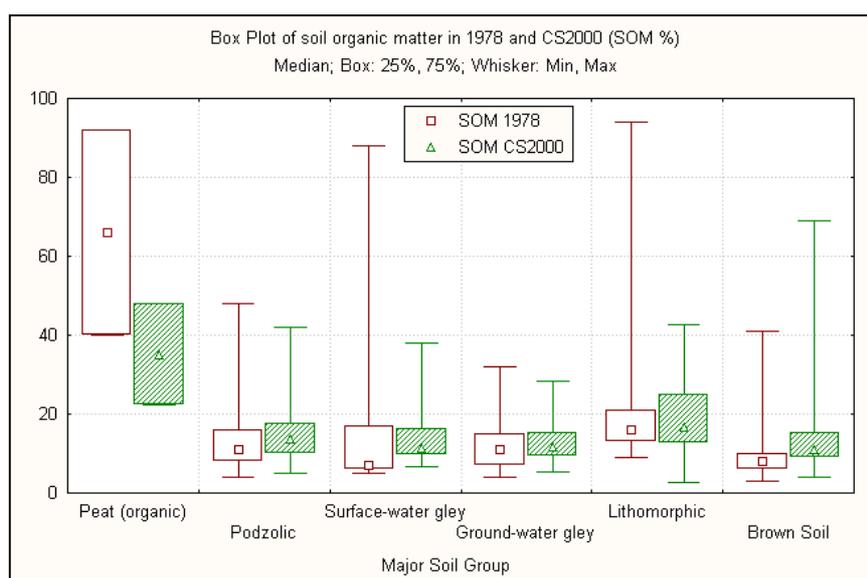


Table 7.16. Median SOM values for 640 repeat X-plots only in 1978, CS2000 and the difference between the two surveys by Major Soil Group and CVS Aggregate Vegetation Class from CS2000.

Major Soil Group	CVS Aggregate Vegetation Class	Sample N	1978	CS2000	difference
Brown soils	Crops and weeds	52	5	5.58	1
	Fertile grassland	57	7	9.29	2
	Heath and Bog	0			
	Infertile grasslands	76	8	10.9	3
	Lowland wooded	7	6	12.7	5
	Moorland Grass Mosaic	7	10	15.1	5
	Tall grass and herbs	13	6	7.19	1
	Upland wooded	11	10	15.7	2
Ground-water gleys	Crops and weeds	22	5	7.03	1
	Fertile grassland	25	8	10.4	2
	Heath and Bog	1			
	Infertile grasslands	29	11	11.9	2
	Lowland wooded	1			
	Moorland Grass Mosaic	4	18	14.9	-3
	Tall grass and herbs	4	25	23.8	0
	Upland wooded	4	11	10.3	-3
Lithomorphic	Crops and weeds	4	6.5	7.94	1.5
	Fertile grassland	9	9	11.4	2
	Heath and Bog	13	80	70.4	3
	Infertile grasslands	18	16	17.1	3
	Lowland wooded	3	19	21.9	4
	Moorland Grass Mosaic	6	82.5	80.6	0.5
	Tall grass and herbs	0			
	Upland wooded	1			
Peat (organic) soils	Crops and weeds	0			
	Fertile grassland	1			
	Heath and Bog	64	94	96	1
	Infertile grasslands	1			
	Lowland wooded	0			
	Moorland Grass Mosaic	8	91	86	3.5
	Tall grass and herbs	2	40.5	45.6	5
	Upland wooded	2	90.5	95	4.5
Pelosols	Crops and weeds	9	6	6.71	1
	Fertile grassland	1			
	Heath and Bog	0			
	Infertile grasslands	0			
	Lowland wooded	1			
	Moorland Grass Mosaic	0			
	Tall grass and herbs	1			
	Upland wooded	0			
Podzolic	Crops and weeds	1			
	Fertile grassland	5	12	17.6	6
	Heath and Bog	13	72	85.4	3
	Infertile grasslands	20	10.5	14	1.5
	Lowland wooded	3	9	14.2	9
	Moorland Grass Mosaic	20	29	25.9	5.5
	Tall grass and herbs	3	7	6.26	-1
	Upland wooded	10	16.5	22.1	1.5
Surface-water gleys	Crops and weeds	15	5	6.68	2
	Fertile grassland	44	7.5	10	3
	Heath and Bog	10	85	89.9	6
	Infertile grasslands	22	7	11.3	4
	Lowland wooded	1			
	Moorland Grass Mosaic	11	65	48	0
	Tall grass and herbs	3	5	10.1	5
	Upland wooded	2	23	17.7	-5
All	All	640	9	12.2	2

The potential for examining soil organic matter contents within individual Aggregate Vegetation Classes is highlighted in Figure 7.8. This box-plot diagram presents summary statistics for soil organic matter content in 1978 and CS2000 for each Major Soil Group in the Infertile Grassland AVC. The number of samples in each Major Soil Group ranged from 2 to 77 (peats to brown soils; no samples in pelosols). The data indicate that median soil organic matter content in this AVC was less than 20% in all Major Soil Groups (except Peats, where sample N was 2). There was, however, a large range in SOM values in both surveys in all Major Soil Groups. There is the potential to explore this variation further to determine whether this, and that seen between the two surveys, corresponds to specific locations or changes in land use.

### **7.3.8 Comparisons with other long-term datasets on soil organic matter or carbon**

The most comparable published data are those from a recent analyses of data from the National Soil Inventory (NSI) and Representative Soil Sampling Scheme (RSSS) (Webb et al., 2001). These results indicate that soil carbon contents declined from 1980 to 1995 in ploughed soils under grassland and lowland peats and peaty soils. These results would correspond with a decline in SOM. The summary result from CS2000 a slight overall increase in SOM (from 1978 to CS2000 although preliminary analyses of AVCs within Major Soil Groups suggests that SOM may have declined in some soils. Further analyses are required to determine whether these correspond to ploughed soils. There are several other possible explanations for the difference between these two studies, from the methodology (sampling to analyses) to the types of land use surveyed. Some of these differences are currently being explored in a comparative analysis of the NSI, RSSS and CS2000 methods and data. Issues over data variability are discussed below.

## **7.4 Conclusion**

Over 1000 soil samples were successfully analysed for loss-on-ignition as a measure of soil organic matter content (SOM) during CS2000. 744 of these soil samples were obtained from the same X-plots where the 1978 soil samples were taken. An ORACLE dataset of SOM data, plus relevant metadata and SOC conversions, has been fully integrated into the Countryside Survey data management system that is maintained at CEH Merlewood. From here the SOM data can be linked to any data from the MASQ project and other Countryside Survey data from the same 1 km square and/or the same X-plot. The SOM datasets are now a valuable resource for investigating the distribution patterns of the soil biological properties and pollutants from CS2000, as well as being able to examine patterns of SOM in soils of the British Countryside. The bi-modal distribution of the SOM data in the British Countryside indicate that future data exploration may benefit from separate analyses of these populations.

Soil organic carbon (SOC) was derived from the SOM data using three equations. These results have been entered into the ORACLE dataset. It was clear from this process, however, that the reliability of these conversions were in doubt. A more reliable method of calculating SOC from SOM would be obtained by deriving an appropriate conversion equation (or equations) based on the relationship between LOI and total C contents of CS2000 soil samples. This would require total C analyses of a sub-set of CS2000 soil samples across the range of SOM values. To establish quantitative amounts of SOM or

SOC in CS2000 soils, the values would also require conversion to a volume basis using bulk density (BD) measurements. It was not practical to take BD measurements during CS2000 since the sampling and measurement of soil samples from BD is currently extremely laborious. If, however, absolute values for carbon contents in soils of Britain are required then consideration must be given to measurement of BD in future sampling, which would require method development.

Summary results produced so far indicate that patterns of SOM from CS2000 correspond with expected patterns. The highest values of SOM were recorded in the uplands, more acidic soils and wetland habitats while the lowest SOM values were recorded in agricultural habitats, especially in the lowlands of England and Wales

The summary statistics from 1978 and CS2000 and the results from differences in SOM between 1978 and CS2000 for known repeat locations all suggest that there has been an overall increase, or at least no change in SOM, over the last thirty years. These results are in agreement with published results from habitat-specific studies (Adamson et al., 1996; Billet et al., 1990). Further analyses are required to examine whether these results are consistent with *in-situ* accumulation of SOM or whether they could be the result of methodology differences and/or small-scale heterogeneity. Part of these analyses must include resolving the variation between the 1978 and CS2000 datasets identified above. Although this variation does not alter the individual datasets from 1978 and CS2000, it does pose questions with respect to expected results from re-sampling the same location. Possible causes of this variation, and potential ways of determining their significance, include:

1. Lack of litter layer in CS2000 soil samples, especially in woodland soils where litter layers tend to build up more than in other habitats. Can be examined by re-visiting 1978 field records to determine whether extreme values had litter removed in 1978.
2. Differences in the LOI methodology (i.e. furnace combustion temperature) between 1978 and CS2000. Can be examined by re-analysing a sub-set of CS2000 soil samples at 1978 furnace temperature to compare with CS2000 methods.
3. Sampling at greater or lower depths during CS2000, resulting in lower or higher relative amounts of SOM compared to mineral material in sample. Can be examined by study of SOM with depth, to determine effects of small differences in sampling depth on SOM.
4. Small-scale spatial heterogeneity in SOM (ca. 2 m between 1978 and CS2000 soil samples from same X-plot). Examine by referring to published data and/or study of *in-situ* heterogeneity.
5. Change in land use over a 30 year period. Can be examined by referring to 1978, 1984, 1990 and CS2000 land use and vegetation data.

The need for further investigation of methodology as well as spatial heterogeneity was also discussed in reference to the unusual differences in 1978 and CS2000 SOM in some samples from the same locations. Differences in the SOM values in the Countryside Survey soil samples between surveys and samples must be explored in greater detail before any significance can be attributed to the trends highlighted in this report.

## **8. HEAVY METALS IN BRITISH SOILS**

### **8.1. Introduction**

The Royal Commission Environmental Pollution report on Sustainable Use of Soil noted that the contents of metals in UK soils have relevance to the broad consideration of soil quality, are related to questions associated with contaminated land and land reclamation and restoration, and are fundamental to the developing considerations on critical loads (RCEP, 1996). Whilst large nation-wide and regional datasets are available on heavy metal contents in UK soils (McGrath and Loveland, 1992; Johnson and Lister, 2001), these are lacking in associated sample site details to support investigation of the relationships between heavy metal contents, other soil properties (in particular soil biology), and the wider environment. Such investigations have been seen as fundamental to the development of sound soil health assessments and soil quality indicators (Doran and Zeiss, 2001).

#### **8.1.1. Specific objectives**

These were:

- Validation of methods prior to analysis, as agreed with the Environment Agency (EA) National Laboratory Service (NLS) Llanelli, Wales.
- Analysis of total concentrations of seven heavy metals in soil samples from Countryside Survey 2000 (CS2000); Cadmium (Cd), Chromium (Cr), Copper (Cu), Nickel (Ni), Lead (Pb), Vanadium (V) and Zinc (Zn).
- Validation of data in collaboration with the EA NLS, Llanelli.
- Compilation of an ORACLE database.
- Production of tables summarising variations in metal contents with Environmental Zone, ITE Land Class and Broad Habitat, Countryside Vegetation System Aggregate Vegetation Class and Major Soil Group.

There are two parts to this section. The first describes the validation exercise carried out in collaboration with the EA NLS laboratories at Llanelli, prior to the full analyses of soil samples collected during CS2000. The second presents the summary results from preliminary analyses of the data. Median metal concentrations in soil were evaluated for three contrasting metals (Cd, Cr and Cu) across four stratification levels of the GB countryside; Environmental Zones (EZ), Broad Habitats (BH), CVS Aggregate Vegetation Classes (AVC) and Major Soil Groups (MSG). Full descriptions of these stratifications are presented in section 2. Data evaluation in terms of current guide values and limits for metals have also been included.

Heavy metal concentrations from soils collected during CS2000 are compared with other regional or national sources of information on heavy metal contents in soils and stream sediments. In the future, there is potential to relate the distribution of these heavy metals in soil to environmental factors such as aerial distribution, historical land use, and soil properties relating to adsorption and mobility.

## **8.2. Method**

The purpose of the study was to produce data of the highest possible quality to ensure that the data would be widely accepted as a baseline indicator of metal contents of soil, against which environmental impact or change may be assessed in the future. It was agreed that the most important aspects of the analysis were to choose a method that was accurate and sufficiently sensitive to reliably quantify the 7 metals in British soil. A target level of 20% error was defined, comprising of 10% bias and 10% precision. A validation exercise was conducted to assess the method performance.

### **8.2.1. Validation of metal analyses in soil samples**

From the outcome of the Scoping Study and subsequent discussions with the Environment Agency, a rigorous validation scheme was established to ensure that the analytical data would be of the best quality possible. At the outset, the targets for quality assurance were defined and a validation process conducted to check the error of the proposed methodology. Following this stage, quality procedures were defined to ensure that there was consistency throughout the period of analysis, and that the data were validated, through the use of certified reference samples and by comparisons in proficiency testing schemes.

During the analysis stage there was continuing assessment of Quality Control (QC) samples and blank samples to check that the method performed within the prescribed limits. In addition, the Merlewood laboratory participated in a proficiency-testing scheme with select sample digests independently analysed at the EA NLS Llanelli to check that there were no major discrepancies. The validation scheme is outlined in more detail in the following sections.

#### **8.2.1.1. Validation analysis**

A validation batch comprised four replicate samples of the following; a certified reference material (CRM calcareous loam soil BCR 141R), an internal reference sample (SR3), a spiked internal reference sample (SR3 and spike), quartz sand blank and a digestion blank. Sample batches were analysed completely independently on 5 different occasions. Analyses were conducted at CEH Merlewood by reflux digestion and Inductively Coupled Plasma-Optical Emission Spectrometers (ICP-OES). The EA NLS, Llanelli performed a parallel study, which included In Addition Microwave Digestion and analyses by Inductively Coupled Plasma-Mass Spectrometry (ICP-MS).

The validation exercise demonstrated that CEH Merlewood could perform trace metal analysis for cadmium, chromium, copper, nickel, lead, vanadium and zinc by reflux digestion, and ICP-OES with a total error of less than 20%. Limits of detection demonstrated that these metals would be quantified at the lowest levels expected in British soils. This methodology provided a valid analytical approach for the analysis of trace metals in soil samples from CS2000.

The EA NLS laboratory demonstrated that microwave digestion contributed to a reduction in the total error in trace metal analysis, but this method of digestion was not yet available at CEH Merlewood and was not, as yet approved by national standards committees. A report on this validation exercise was prepared and the analyses of the

CS2000 soil samples started after the results were discussed between the EA and CEH (see Project Record). CEH Merlewood and the EA NLS Llanelli are compiling the results from this validation for submission in an appropriate refereed journal.

### 8.2.1.2. Definition of quality procedures

The QC protocol was agreed with the EA NLS. A CRM, internal control sample (SR3) and two blanks would be analysed with each batch of 20 digest samples. Quartz sand could not be used as a blank due to its high lead content. QC samples analysed with each batch were recorded on Shewhart charts, and warning limits of 10% ( $2*\sigma$ ) applied. Digest blanks were also monitored for consistency and contamination. CEH agreed to continue participation in a proficiency-testing scheme (International Soil Exchange (ISE) scheme) during the course of project analyses, organised by the University of Wageningen, Netherlands. A sub-set of sample digests was sent to the EA NLS for comparison analysis by ICP-MS.

### 8.2.1.3. Assessment of performance data

Consistent and low blanks were obtained during the analysis of the 60 analytical batches (Table 8.1). A very small proportion of the analytical measurements (0.2%) were below the limit of detection. The majority of these low observations were related to cadmium measurements. The lowest concentration of the metals Cu, Pb, V and Zn were well above the limit of detection. The mean values obtained for the CRM (Table 8.2) indicate only a small bias, up to 5% for lead. This bias represents a significant achievement, and an improvement on the initial validation data. Unfortunately there is no certified value for vanadium to assess bias. Coefficients of variation for CRM (Table 8.2) and SR3 (Table 8.3) were between 3 to 5%, except for cadmium and lead, which are slightly more variable (up to 7%). The overall error of the method (Table 8.2) was between 7% (zinc) and 14% (lead), below the target limit of 20%

Table 8.1 Method performance data relating to blank and minimum concentrations observed in soils.

mg kg <sup>-1</sup>	Cd	Cr	Cu	Ni	Pb	V	Zn
Blank	0.005	0.05	-0.7	0.1	0.7	0.1	0.01
Minimum <sup>1</sup>	<0.02	<0.1	1.2	0.3	1.3	1.8	2.5
Det. Limit <sup>2</sup>	0.02	0.1	0.04	0.2	0.2	0.1	0.2
Less than <sup>3</sup>	11	3	0	0	0	0	0

<sup>1</sup> The lowest content reported; <sup>2</sup> Aqua regia digestion method detection limit; <sup>3</sup> Number of samples with concentrations below the detection limit.

Table 8.2 Summary of certified reference material (CRM141R) concentrations (n=49).

mg kg <sup>-1</sup>	Cd	Cr	Cu	Ni	Pb	V	Zn
mean	13.6	141	47	97.3	48.6	50.3	261
$\sigma$	0.7	5.3	1.7	3.7	3.3	2.1	9.3
CV <sup>4</sup>	5.1	3.8	3.6	3.8	6.9	4.1	3.5
Certified <sup>5</sup>	14	138	46.9	94	51.3	n/a	270
Error <sup>6</sup>	13.1	9.7	11.3	11.0	19.4	n/a	10.4

<sup>4</sup> Coefficient of variation; <sup>5</sup> Certified value based on aqua regia digestion; <sup>6</sup> Error of the method = bias +  $2*\sigma$

Table 8.3 Summary of internal reference (SR3) concentrations (n=71).

mg kg <sup>-1</sup>	Cd	Cr	Cu	Ni	Pb	V	Zn
mean	0.578	62.8	24	39.4	52.8	81	98.4
σ	0.04	1.8	0.8	1.5	3.8	3.6	4.9
CV	7.0	2.8	3.4	3.7	7.2	4.4	5.0

#### 8.2.1.4. Proficiency testing data

Results from the proficiency testing exercises are summarised in Table 8.4. On each occasion (represented by year and distribution e.g. 2000.4) four different samples were analysed for seven specified metals. Values were submitted to the scheme organiser. The bias value represents the mean difference for the 4 samples, from the median value (excluding outliers) of the participating laboratories. The data support the statement that the CEH Merlewood laboratory produces accurate data that are in agreement with those produced by the community performing metal analysis on soil samples. The confirmation is particularly significant for Vanadium, as this is the only means to assess accuracy.

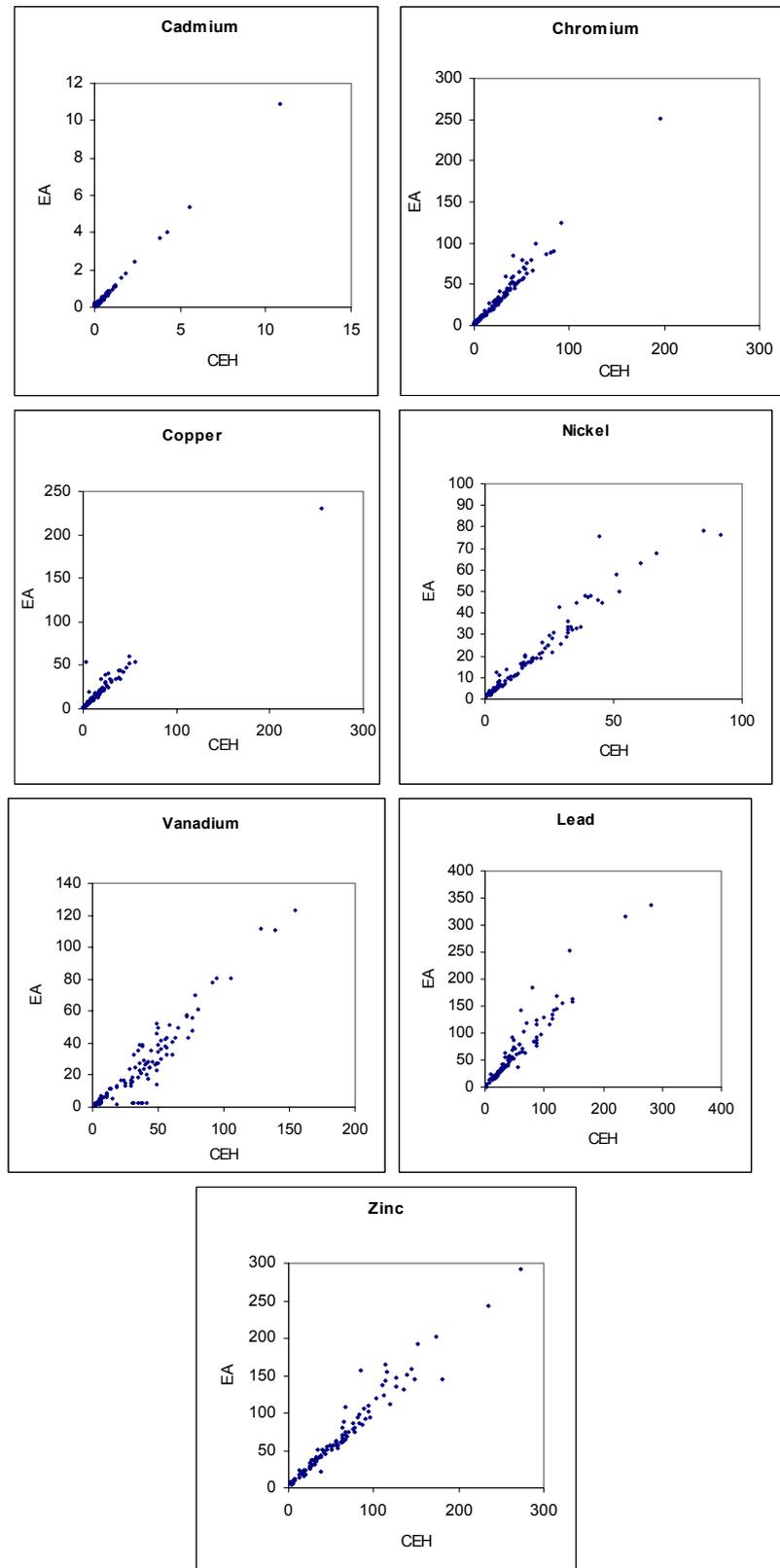
Table 8.4 Summary data of proficiency test data reported to the International Soil Exchange Programme (ISE).

% bias	Cd	Cr	Cu	Ni	Pb	V	Zn
2000.2	-10.0	-3.8	-0.7	12.4	11.7	-6.1	-9.6
2000.3	-12.5	2.8	-3.7	7.6	-6.3	1.0	-6.4
2000.4	-14.1	6.8	2.2	9.2	6.6	9.4	0.2
2001.1	3.5	2.4	4.3	11.5	-6.1	4.6	2.7
mean	-8.3	2.1	0.5	10.2	1.5	2.2	-3.3

Table 8.5 Summary of QC data from CEH Merlewood and EA NLS Llanelli (n=5).

Element	Lab	Mean SR3	Mean CRM141R	Certified CRM141R	Recovery (%)
Cd	CEH	0.560	13.5	14	96.4
Cd	EA	0.535	12.5	14	89.0
Cr	CEH	62.3	142	138	102.6
Cr	EA	67.0	146	138	105.5
Cu	CEH	23.4	46.6	46.9	99.4
Cu	EA	21.8	42.3	46.9	90.3
Ni	CEH	38.8	96.3	94.0	102.5
Ni	EA	34.7	86.1	94.0	91.6
Pb	CEH	52.4	47.6	51.3	92.9
Pb	EA	56.4	56.5	51.3	110.2
V	CEH	82.0	51.2	N/A	N/A
V	EA	65.0	43.1	N/A	N/A
Zn	CEH	97.1	260	270	96.4
Zn	EA	98.6	255	270	94.5

Figure 8.1. A comparison of the results of heavy metal analyses at EA NLS Llanelli and CEH Merlewood



### 8.2.1.5. Comparison of analyses by ICP-MS (EA) and ICP-OES (CEH)

A representative subset of 100 digest solutions was analysed for heavy metals, arsenic and mercury at NLS by ICP-MS analysis. These samples were chosen to provide coverage of all Major Soil Groups across Great Britain. In addition 5\* CRM digests, 5\* SR3 digests and blank solutions were also exchanged. In addition, the NLS laboratory analysed 3 of the digestion solutions in duplicate. Both laboratories reported mean recovery for the certified reference sample of +/-11% (Table 8.5).

There was excellent agreement between laboratory techniques for the metals Cd, Cu, Ni and Zn (Figure 8.1; Table 8.6). The weakest relationships between the laboratories were in the analysis of the metals chromium and vanadium. The CEH laboratory conducted repeat analysis of digests where the greatest discrepancies occurred. The laboratory confirmed their original data.

### 8.2.1.6. Conclusions

CEH Merlewood conformed to quality assurance protocols as agreed with the EA. Tough targets for accuracy and precision were defined and met. Total errors for the metal analysis were between 7 and 14%. Independent checks with the Llanelli laboratory confirms that data produced are fit for the purpose of the project (see Figure 8.1).

Table 8.6 The relationship between the data obtained at the EA NLS Llanelli and CEH Merlewood laboratories ( $EA=a+b*CEH$ )

mg kg <sup>-1</sup>	Cd	Cr	Cu	Ni	Pb	V	Zn <sup>1</sup>
a	0.011	0.597	3.75	0.556	15.1	3.95	4.33
b	0.987	1.24	0.909	1.04	0.960	0.790	1.02
R <sup>2</sup>	0.997	0.967	0.949	0.999	0.963	0.889	0.969

<sup>1</sup> One outlier rejected

### 8.2.2. Analyses of CS2000 soil samples

From the soil samples collected in the field during CS2000, 1080 were analysed for heavy metal concentrations. There was insufficient sample for the remaining for analyses.

#### 8.2.2.1. Method

Samples were air dried and sieved to less than two millimetres (<2mm). A portion of air-dried <2mm sieved soil was systematically sub-sampled (coning and quartering) for ball milling using a non-metallic agate mill. Total metal concentrations were determined following digestion in aqua regia under reflux. 3g of air-dry soil was dissolved in acid and diluted to 100ml (MEWAM, 1986- Method A). Metals concentrations were determined on an ICP-OES JY38plus sequential instrument. The ICP was operated at conditions of high power and low flow (see Table 8.7). The detection limits obtained are listed. Only 12 measurements were observed below the computed limit of detection,

8 for Cd, 3 for Cr and 1 for Ni. The EA NLS laboratory at Llanelli determined values for these samples using ICP-MS. The 12.5% HNO<sub>3</sub> calibration solutions were matched to soil digestion matrix by inclusion of Ca (1071ppm), Mg (300ppm), Fe (750ppm) and Al (500ppm). Samples were organised into analytical batches of 20 samples plus 2 QC samples and 2 blanks. Reflux aqua-regia digestion was performed in groups of 12, i.e. 10 samples, one QC sample and one blank.

Table 8.7 ICP-OES technical details.

Element	2	Wave length	Detection limit (ppb in solution)	Detection limit (mg kg <sup>-1</sup> )
3 Cd		228.802	1	0.02
Cr		357.869	5	0.1
Cu		324.754	2	0.04
Ni		341.476	10	0.2
Pb		220.282	10	0.2
V		292.402	6	0.1
Zn		213.856	12	0.2

### 8.2.2.2. Validation

An initial validation exercise established the capability of the approach and the total error of the methods to be less than 20%. The NLS Llanelli laboratory outlined the QC specification. Shewhart charts were maintained with certified or mean values, and indicative bias limits of plus or minus 10%. Blank values were also monitored on Shewhart charts to monitor background levels and possible contamination.

### 8.2.2.3. Statistical Analyses

Descriptive statistics tables were produced (see Project Record) to illustrate the distribution of metals by Major Soil Group, Countryside Vegetation System Aggregate Vegetation Class (AVC), ITE Land Class and Broad Habitat (BH), within 6 environmental zones (EZ) in England, Wales and Scotland. Summary data representing less than 10 samples are not presented since these groups represent less than 1% of the whole population and conclusions may be unreliable.

## 8.3. Results

The following outlines the summary statistics produced and preliminary analyses of these heavy metal concentrations with other environmental variables. Summary tables were produced for each metal and are presented in the Project Record.

### 8.3.1. Concentrations of heavy metals in soils at the national scale

A summary of the metal concentrations from this survey is presented in Table 8.8 along with concentrations reported for England and Wales by McGrath and Loveland (1992) (Table 8.9). By comparing these data, in general the soils from CS2000 contained lower concentrations of Cd, Cr, Ni, Pb and Zn than those recorded in the 1992 McGrath and Loveland Survey. The soils from CS2000 contained a higher maximum concentration of

Ni and Cu while the mean/median concentrations of Cu were equivalent. The Countryside Surveys are, as named, focussed on the rural environment in Britain with only 1% of locations being classified as within built-up areas or gardens. This may account for the lower overall concentrations of metals in soils since areas excluded from the survey (e.g. conurbations, industrial wasteland) may be expected to have the highest heavy metal concentrations, in particular, cadmium and copper.

Table 8.8 Summary statistics for heavy metal concentrations in soil from CS2000.

mg kg <sup>-1</sup>	Cd	Cr	Cu	Ni	Pb	V	Zn
4 Mean	0.49	28.5	18.1	23.7	88.0	40.2	80.0
Median	0.30	26.5	13.6	16.3	37.4	38.8	61.7
Standard Deviation	0.83	24.0	25.6	79.7	638.8	27.4	119.5
Skewness	7.92	3.0	9.8	19.4	30.8	1.0	11.0
Kurtosis	83.33	20.6	137.1	417.0	987.9	1.8	164.0
Minimum	0.00	0.0	0.3	0.0	1.3	1.8	2.5
Maximum	11.20	267	448	1890	20600	174	2120

Table 8.9. Summary statistics for heavy metal concentrations in the soils of England and Wales from McGrath and Loveland (1992).

mg kg <sup>-1</sup>	Cd	Cr	Cu	Ni	Pb	Zn
<b>Mean</b>	0.80	41	23	25	74	97
Median	0.70	39	18	23	40	82
Skewness	17.6	9.46	21.2	6.95	42.7	13.6
Kurtosis	574.4	205.4	654.6	132.9	2449.6	299.6
Minimum	< 0.2	0.2	1.2	0.8	3.0	5
Maximum	41	840	1510	440	16300	3650

Non-normal distributions of concentrations were observed for all 7 metals (Figure 8.2.). The linear centre portion represents the main population, with the curved portion at each end representing the proportion of those soils that may be classified as either higher or lower than the main grouping. Therefore we observe that 10% of concentrations of Cd, Pb, Cu and Zn measured in soils are low, whilst 30% of Cr, Ni and V may be classified as low. Less than 5% of the metals concentrations appear to be high. McGrath and Loveland (1992) plots show similar patterns, with the notable exception of Cr where their plots is more similar to the other heavy metals.

The distribution patterns show similar trends among metals that are often associated with each other and that are adjacent in the Periodic Table of elements; Cd/Pb, Ni/V/Cr and Cu/Zn. The peak of the population for the metals Cd, Pb, Cu and Zn occurs in the region of the twenty-fifth percentile. In contrast the peak of the population frequency, for Cr, Ni and V is observed within the lowest ten percentile. These close associations are further supported by the significant correlations ( $p < 0.001$ ), derived from  $\log_{10}(x)$  (Table 8.10) and from regressions of heavy metal concentrations (Figure 8.3).

Figure 8.2. Distribution plots for CS2000 soil sample heavy metal analyses

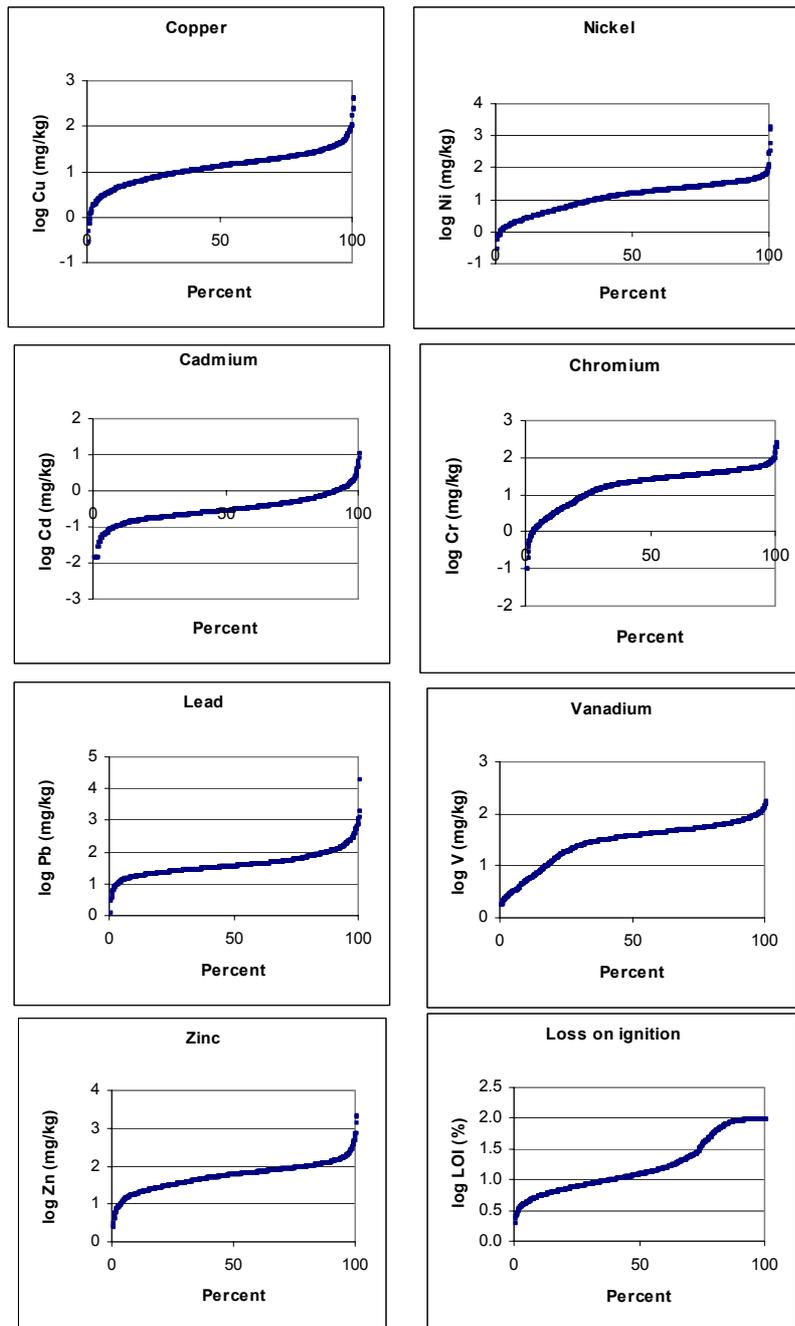
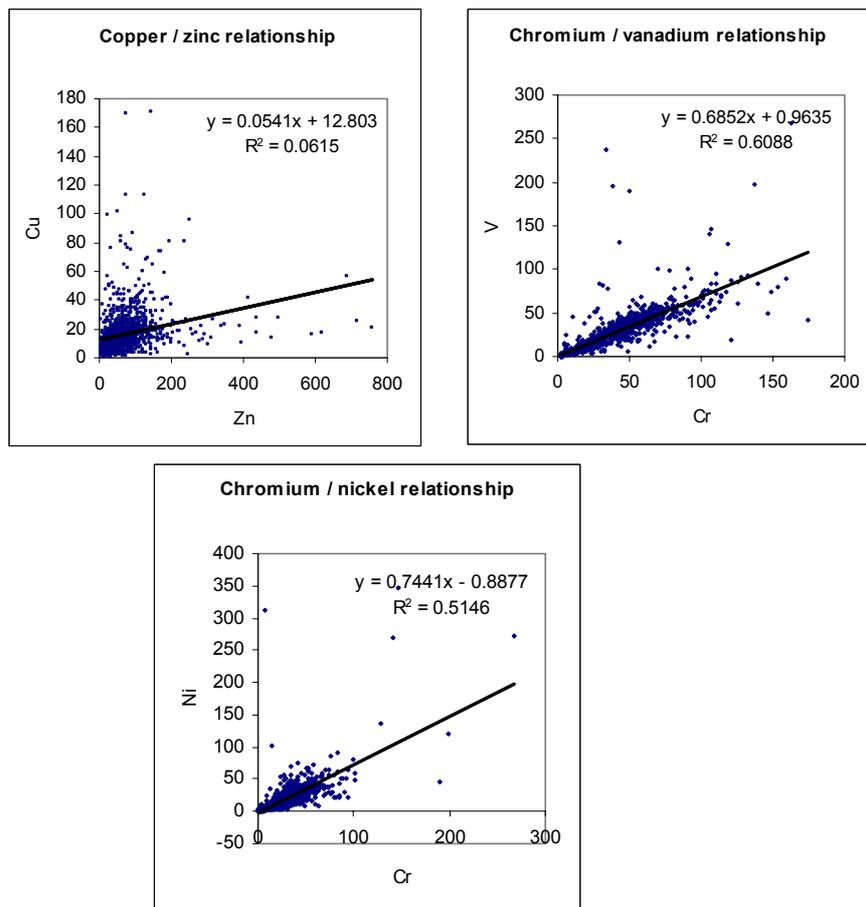


Table 8.10. Correlation matrix of heavy metals, soil pH and soil organic matter content, derived from  $\log_{10}(x)$ .

	Cd	Cr	Cu	Ni	Pb	V	Zn
4.1.1.1 Cd	1	-0.071	<b>0.329</b>	0.076	<b>0.504</b>	-0.081	<b>0.464</b>
Cr	-0.071	1	<b>0.617</b>	<b>0.883</b>	0.072	<b>0.935</b>	<b>0.587</b>
Cu	<b>0.329</b>	<b>0.617</b>	1	<b>0.724</b>	<b>0.469</b>	<b>0.620</b>	<b>0.751</b>
Ni	0.076	<b>0.883</b>	<b>0.724</b>	1	0.151	<b>0.826</b>	<b>0.738</b>
Pb	<b>0.504</b>	0.072	<b>0.469</b>	0.151	1	0.092	<b>0.447</b>
V	-0.081	<b>0.935</b>	<b>0.620</b>	<b>0.826</b>	0.092	1	<b>0.447</b>
Zn	<b>0.464</b>	<b>0.587</b>	<b>0.751</b>	<b>0.738</b>	<b>0.447</b>	<b>0.586</b>	1
soil pH (H <sub>2</sub> O)	0.089	<b>0.542</b>	<b>0.388</b>	<b>0.569</b>	-0.134	<b>0.526</b>	<b>0.473</b>
Soil organic matter content (LOI%)	<b>0.311</b>	<b>-0.674</b>	<b>-0.337</b>	<b>-0.576</b>	0.169	<b>-0.667</b>	<b>-0.363</b>

All metals except Pb, N=1080; Pb, N=1079; values in bold indicate a significant relationship ( $p < 0.05$ )

Figure 8.3. Linear regression relationships between selected heavy metal concentrations in soils from Countryside Survey 2000.



A regional level comparison of metal concentrations was carried out since existing databases separate them in this manner e.g. McGrath and Loveland (1992). We observed that, in general, soils from CS2000 that were sampled in Scotland exhibited lower mean and median heavy metal concentrations than those sampled in England and Wales (Table 8.11) for all except median values for Cadmium. There was remarkable similarity between the median values from England and Wales from McGrath and Loveland (1992) and CS2000 for all metals, although Cd values were lower overall in CS2000 soils.

Table 8.11. Heavy metal concentrations in soils from CS2000 for England/Wales and Scotland, compared with EEC limit values.

mg kg <sup>-1</sup>	Cd	Cr	Cu	Ni	Pb	Zn
England/Wales mean	0.57	33	23.3	24.3	84.7	105
Scotland mean	0.38	22.3	10.9	22.9	47.3	45.4
England/Wales median	0.3	30.9	17.3	21.6	43.3	78.7
Scotland median	0.29	12.9	7.8	6.2	30.6	34.9
Max. limit values for soil <sup>1</sup>	3	600	140	75	300	300
Number of samples above limit	14	0	6	13	29	21

<sup>1</sup> Directive 86/278/EEC (under revision)

From Table 8.11, it is apparent that a relatively small proportion of soils analysed contain heavy metal concentrations that were greater than the maximum soil limits adopted in EEC Directive 86/278/EEC; these limit values are similar to current UK limit values that are also under review. In these samples, Pb was the most frequently above these limit values but this was still in relatively few samples (<2.7% of all samples). The following discusses median values of heavy metal concentrations within stratifications of the British Countryside used within CS2000. The aim was to highlight trends within these stratifications. Appropriate statistical analyses will be required to further substantiate the trends discussed.

### 8.3.2. Environmental Zone

Environmental Zones 1, 2 and 3 occur in England and Wales, and EZs 4, 5 and 6 occur in Scotland. 47% of the heavy metal concentrations in soils from CS2000 were recorded in Environmental Zones 1 and 2 while the least number were recorded from Environmental Zone 3 (11% of analyses). Median values for the concentrations of heavy metals in soils from CS2000 by Environmental Zone are presented in Table 8.12.

Table 8.12. Median heavy metal concentrations in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone.

Environmental Zone	Cd	Cr	Cu	Ni	Pb	V	Zn
1	0.29	32.6	16.2	24.6	33.7	46.1	82.3
2	0.30	31.2	19.4	21.6	47.4	42.8	82.6
3	0.31	22.9	15.7	12.8	79.4	34.1	65.1
4	0.24	31.5	11.5	18.0	31.1	45.0	56.6
5	0.31	6.1	6.0	3.7	24.9	10.9	28.3
6	0.37	6.0	6.2	4.9	44.2	12.8	32.2
All	0.30	26.5	13.6	16.3	37.4	38.8	61.7

These results indicate that similar trends are apparent in the median concentrations of Cr, Cu, Ni, V and Zn across the Environmental Zones. In all these metals, the lowest concentrations in soil were apparent in Zones 5 and 6, while higher median concentrations were found in Zones 1 and 2, and to a lesser extent Zones 3 and 4. Median concentrations of Cd were low and little different across the Environmental Zones, though the median value was lowest in Zone 4 and highest in Zone 6. Median concentrations of Pb in soil followed a different trend, with the highest median value in Zone 3 and lowest in Zone 5.

### 8.3.3. Broad Habitats

Two Broad Habitats (improved grassland; arable and horticulture) contain 49% of the soils analysed for heavy metals. Over 75% of the habitats contain fewer than 10 sampling points. Median values for the concentrations of heavy metals in soils from CS2000 by Broad Habitat are presented in Table 8.13.

Table 8.13. Median heavy metal concentrations in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Broad Habitat.

Broad Habitat <sup>1</sup>	Cd	Cr	Cu	Ni	Pb	V	Zn
Broadleaved, mixed and yew woodland	0.24	22.8	14.1	15.8	48	35.6	68.5
Coniferous woodland	0.30	10.7	7.6	6.3	44.6	16.5	32.8
Arable and horticultural	0.26	36.1	18.8	26.4	31.9	49.9	80.4
Improved grassland	0.26	34.5	15.6	21.8	37.7	49.0	74.4
Neutral grassland	0.29	31.6	13.5	20.7	46.8	41.4	68.1
Acid grassland	0.39	10.4	8.7	6.3	48.4	19.5	29.5
Bracken	0.22	22.6	11.9	16.4	46.2	35.1	52.3
Dwarf shrub heath	0.40	5.9	8	4.2	39.4	11.7	30.4
Fen, marsh and swamp	0.29	27.7	9.3	11.7	37.6	36.7	51.2
Bog	0.42	2.4	5.3	2.7	31.3	5.2	28.9
Built-up areas and gardens	0.31	24.4	27.4	18.2	112	38.8	93.2
All	0.30	26.0	13.5	15.8	37.4	38.3	60.7

<sup>1</sup> Habitats containing less than 10 samples have been excluded.

The range of median values across the Broad Habitats highlights trends across the metals. Relatively higher median values of Cr, Cu, Ni, V and Zn were recorded in soils from arable/horticultural habitats and improved grasslands with lower median values, in general, recorded from bog, dwarf shrub heath, coniferous woodland and/or acid grassland. Concentrations of Cd in soil tended to follow the opposite trend with highest median values in bog, dwarf shrub heath and acid grassland and lowest in bracken and woodland. The highest median values of Zn, Pb and Cu were recorded in built up areas and gardens. However, these values may reflect the relatively few samples taken in this habitat. Relatively higher median values of Pb were also recorded in both woodland habitats and acid grassland.

### 8.3.4. CVS Aggregate Vegetation Class

The majority of soil sample analysed (72%) were taken from four Aggregate Vegetation Classes; crops and weeds, fertile grassland, heath and bog, and infertile grasslands. Grasslands account for 38% of soils analysed; 20% of these grasslands are Infertile

Grassland. Median values for the heavy metal concentrations in soils from CS2000 by CVS Aggregate Vegetation Class are presented in Table 8.14.

Table 8.14. Median concentrations of heavy metal in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Aggregate Vegetation Class.

Aggregate Vegetation Class	Cd	Cr	Cu	Ni	Pb	V	Zn
Crops and weeds	0.25	34.1	18.1	25.3	31.7	48.9	75.2
Tall grass and herb	0.33	41.0	23.1	29.7	45.4	51.6	95.7
Fertile grassland	0.28	33.8	16.2	21.1	39.8	46.8	75.8
Infertile grassland	0.25	33.6	14.7	21.5	38.3	45.4	74.4
Lowland wooded	0.32	22.8	14.1	20.8	62.7	41.3	82.4
Upland wooded	0.20	16.8	10.9	11.1	48.4	32.2	42.4
Moorland grass mosaics	0.30	12.7	8.4	6.4	36.5	21.4	31.8
Heath and bog	0.46	3.4	5.5	3	34.9	5.9	28.2
All	0.30	26.5	13.6	16.3	37.4	38.8	61.7

The values of Cr, Cu, Ni, V and Zn follow similar trends across the AVCs, with highest median values in tall grass and herbs and lowest values in heath and bog. The highest and lowest median values for Pb were recorded in lowland wood and crops/weeds, respectively. The highest and lowest median values for Cd were recorded in heath/bog and upland woodland, respectively.

### 8.3.5. Major Soil Groups

All Major Soil Groups, except terrestrial raw, raw gley and man made soils, are represented by samples analysed for heavy metal concentrations. A large percentage of soils analysed (65%) can be assigned to three Major Soil Groups; brown soils, surface water gleys and podzolic soils; brown soils were the most numerous (33%). Further evaluation of these assignments will be required as there are some discrepancies in soil characteristics i.e. 10% of soils classified as Peat (organic) soils contained low organic matter content (<25%). Median values for the heavy metal concentrations in soils from CS2000 by Major Soil Group are presented in Table 8.15.

Table 8.15. Median concentrations of heavy metals in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Major Soil Group.

Major Soil Group	Cd	Cr	Cu	Ni	Pb	V	Zn
Lithomorphic soils	0.41	21.7	10.3	13.4	37.3	34.1	49.2
Pelosols	0.30	47.9	19.2	36.0	27.3	61.5	91.1
Brown soils	0.28	32.0	16.2	22.7	39.0	44.1	74.3
Podzolic soils	0.25	12.2	9.7	6.3	40.2	22.5	33.3
Surface-water Gley soils	0.30	28.7	16.9	19.1	43.3	42.4	64.8
Ground-water Gley soils	0.20	36.6	14.9	22.9	30.5	50.4	65.9
Peat (organic) soils	0.46	2.6	5.1	2.7	33.2	5.2	30.2
All	0.30	26.5	13.6	16.3	37.4	38.8	61.7

The highest median values for Cr, Cu, Ni, V and Zn were recorded in pelosols. In contrast, median Pb concentrations were lowest in pelosols. There were relatively few samples from this Major Soil Group and this may be reflected in these results. After pelosols, highest median values for Cr, Cu, Ni, V and Zn were recorded in gleys and brown soils. The lowest median values for Cr, Cu, Ni, V and Zn were recorded in podzols and peat soils. These results concur with those recorded by McGrath and Loveland (1992). Peat soils contained the highest concentrations of Cd whilst the lowest concentrations of Cd were recorded in ground water gleys.

### **8.3.6. Contrasting behaviour and origins of heavy metals in soils**

Three groupings corresponding to similarities between metals were proposed earlier; Cd/Pb, Cr/V/Ni and Cu/Zn. The first metal in each of these, Cd, Cr and Cu, are discussed in detail to highlight the contrasting behaviour and origins of the groupings and further illustrate patterns of variation in heavy metal concentrations in soils across Britain.

#### **8.3.6.1. Cadmium**

As discussed previously, the concentration values for this metal were skewed (Figure 8.2) and the median value much lower ( $0.3 \text{ mg kg}^{-1}$ ) than the mean ( $0.49 \text{ mg kg}^{-1}$ ) (Table 8.8). Approx. 10% of the CS2000 soil samples contained low concentrations of Cd ( $< c. 0.12 \text{ mg kg}^{-1}$ ) while c. 5% contained relatively high Cd concentrations ( $>1.4 \text{ mg kg}^{-1}$ ); Figure 8.2.

#### **Broad Habitat**

As stated above, bogs were found to contain the highest median values of Cd. However, when Cd concentrations were categorised by Environmental Zone and Broad Habitat, the highest median values occurred in the arable/horticultural areas and broadleaved woodlands in EZ 1, dwarf shrub heaths in EZ 2, and in acid grasslands in EZ 6 (Table 8.16.). The lowest concentrations were found in coniferous woodland soils in EZs 4 and 5), and in improved grassland and fen/marsh/swamp habitats EZ 5.

#### **Aggregate Vegetation Class**

The highest median values of Cd were associated with heath and bog areas in EZs 3 and 4 and the lowest concentrations with infertile grasslands in EZs 4 and 5 (Table 8.17.).

#### **Major Soil Group**

The lithomorphous soils in EZs 1 and 2, and peat soils in EZ 3 exhibited the highest median concentrations of Cd whilst the lowest median values were recorded in ground water gleys in EZs 4 and 5, brown soils in EZ 5 and podzols in EZ 4 (Table 8.18). McGrath and Loveland (1992) also reported higher concentrations in lithomorphous soils.

Table 8.16. Median concentrations of cadmium in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and Broad Habitat.

Median cadmium	Environmental Zone							
Broad Habitat	Code	1	2	3	4	5	6	Total
Broadleaved, mixed and yew woodland	1	0.36	0.28	0.15	0.13	0.24	0.14	0.24
Coniferous woodland	2	0.42	0.47	0.32	0.05	0.16	0.33	0.3
Boundary and linear features	3	.	0.19	.	.	.	.	0.19
Arable and horticultural	4	0.26	0.29	0.42	0.24	0.11	.	0.26
Improved grassland	5	0.3	0.29	0.25	0.21	0.18	0.19	0.26
Neutral grassland	6	0.37	0.29	0.18	0.3	0.3	0.29	0.29
Calcareous grassland	7	0.94	2.23	.	.	.	.	1.89
Acid grassland	8	0.11	0.34	0.25	0.7	0.37	0.42	0.39
Bracken	9	0.29	0.13	0.31	0.17	.	0.23	0.22
Dwarf shrub heath	10	.	0.34	0.61	0.41	0.33	0.51	0.4
Fen, marsh and swamp	11	.	0.73	0.17	0.27	0.19	0.31	0.29
Bog	12	.	0.41	1.4	0.63	0.43	0.37	0.42
Inland rock	16	0.7	.	.	.	.	.	0.7
Built-up areas and gardens	17	0.24	0.31	2.96	.	.	.	0.31
Supralittoral rock	18	.	0.88	.	.	0.38	.	0.54
Supralittoral sediment	19	.	0.08	.	.	.	.	0.08
Littoral sediment	21	.	0.24	.	.	.	.	0.24
	Total	0.29	0.3	0.31	0.24	0.31	0.36	0.3

Table 8.17. Median concentrations of cadmium in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and CVS Aggregate Vegetation Class (AVC).

Median Cadmium	Environmental Zone							
CVS AVC	Code	1	2	3	4	5	6	Total
Crops and weeds	1	0.26	0.29	0.42	0.23	0.09	.	0.25
Tall grass and herb	2	0.32	0.35	.	0.18	.	.	0.33
Fertile grassland	3	0.31	0.28	0.41	0.26	0.2	.	0.28
Infertile grassland	4	0.4	0.3	0.23	0.17	0.18	0.28	0.25
Lowland wooded	5	0.32	0.31	.	0.39	.	.	0.32
Upland wooded	6	0.35	0.23	0.29	0.12	0.1	0.31	0.2
Moorland grass mosaics	7	.	0.34	0.28	0.29	0.31	0.3	0.3
Heath and bog	8	.	0.41	0.82	0.63	0.4	0.42	0.46
	Total	0.29	0.3	0.31	0.24	0.31	0.37	0.3

Table 8.18. Median concentrations of cadmium in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and Major Soil Group.

Median Cadmium	Environmental Zone							
Major Soil Group	Code	1	2	3	4	5	6	Total
Lithomorphic Soils	3	0.99	1.89	0.33	0.28	0.3	0.37	0.41
Pelosols	4	0.3	.	.	.	.	.	0.3
Brown Soils	5	0.31	0.26	0.25	0.26	0.17	0.27	0.28
Podzolic Soils	6	0.24	0.25	0.24	0.19	0.25	0.31	0.25
Surface-water Gley Soils	7	0.23	0.3	0.48	0.31	0.47	0.33	0.3
Ground-water Gley Soils	8	0.22	0.3	.	0.17	0.17	0.68	0.2
Peat (organic) soils	10	0.27	1.36	0.68	0.58	0.42	0.46	0.46
Total		0.29	0.3	0.31	0.24	0.31	0.37	0.3

### 8.3.6.2. Chromium

As discussed previously, the concentration values for this metal were not normally distributed (Figure 8.2). The median value was only slightly lower ( $27 \text{ mg kg}^{-1}$ ) than the mean ( $29 \text{ mg kg}^{-1}$ ) (Table 8.8). Approx. 30% of the CS2000 soil samples contained low concentrations of Cr ( $< c. 0.15 \text{ mg kg}^{-1}$ ) while c. 5% contained relatively high Cr concentrations ( $>60 \text{ mg kg}^{-1}$ ); Figure 8.2. The patterns of distribution for Cr may also apply to Ni and V.

### Broad Habitats

The highest median concentrations of Cr in soil were recorded in arable and horticultural habitats (Table 8.19). However, within Environmental Zones, the highest median values occurred in improved grasslands in EZs 3 and 4 and in arable/horticultural habitats in EZs 1, 2 and 4. The lowest values were mainly associated with coniferous woodland, acid grassland, dwarf shrub heaths and bogs in EZ 6 in Scotland.

### Aggregate Vegetation Class

Highest Cr concentrations were recorded in tall grass and herbs, and lowest in heath and bog areas in EZ 3 and EZs 4, 5 and 6 (Table 8.20.).

### Major Soil Group

Lithomorphic soils in EZ 2, and pelosols and ground-water gleys in EZ 1 exhibited the highest median concentrations of Cr (Table 8.21.). Lowest levels were measured in peat soils and podzols collected from EZ 5 and 6. McGrath and Loveland (1992) also reported high concentrations in pelosols and ground-water gleys, and low median values for podzolic and peat soils.

Table 8.19. Median concentrations of chromium in soils from CS2000 ( $\text{mg kg}^{-1}$ ) categorised by Environmental Zone and Broad Habitat.

Median chromium	Broad Habitat	Code	Environmental Zone						Total
			1	2	3	4	5	6	
Broadleaved, mixed and yew woodland	1	26.3	20.1	23.6	35.8	2.3	14.2	22.75	
Coniferous woodland	2	44.5	8.45	8.2	21.7	6.35	10.75	10.65	
Boundary and linear features	3	.	20.4	.	.	.	.	20.4	
Arable and horticultural	4	37.5	33.6	64.2	36.1	30.45	.	36.05	
Improved grassland	5	31.9	33.55	37.65	39.5	31.7	33.7	34.5	
Neutral grassland	6	24	31.4	12	35.45	61.75	48.7	31.6	
Calcareous grassland	7	17.6	24.75	.	.	.	.	21.2	
Acid grassland	8	8.6	14.7	19.2	8.45	13.25	7.7	10.4	
Bracken	9	16.65	26.5	17.5	25.8	.	22.6	22.6	
Dwarf shrub heath	10	.	7.6	7.3	5.6	5.4	4.7	5.9	
Fen, marsh and swamp	11	.	43.2	22.9	19.4	27.8	13.5	27.7	
Bog	12	.	5.6	9.2	2.1	1.8	3.95	2.4	
Inland rock	16	21.8	.	.	.	.	.	21.8	
Built-up areas and gardens	17	18.3	37.1	52.7	.	.	.	24.4	
Supralittoral rock	18	.	23.1	.	.	1.35	.	2.2	
Supralittoral sediment	19	.	30.6	.	.	.	.	30.6	
Littoral sediment	21	.	33.75	.	.	.	.	33.75	
	Total	32.55	31	22.5	31.35	6.05	6.2	25.95	

Table 8.20. Median concentrations of chromium in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and CVS Aggregate Vegetation Class.

Median Chromium	CVS AVC	Environmental Zone						Total
		Code	1	2	3	4	5	
Crops and weeds	1	34.5	31.2	64.2	35.95	28.8	.	34.05
Tall grass and herb	2	45.3	37.5	.	40.3	.	.	41
Fertile grassland	3	31.55	32.2	41.4	35.5	34.1	.	33.8
Infertile grassland	4	27.6	33.2	36.15	37.9	31.1	33.7	33.6
Lowland wooded	5	26.3	19.8	.	35.8	.	.	22.8
Upland wooded	6	17.15	10.9	18.4	24.85	11.35	17.8	16.8
Moorland grass mosaics	7	.	20.6	14.7	7.85	11	14.7	12.65
Heath and bog	8	.	6.6	4.5	3.15	2.1	3.9	3.4
	Total	32.55	31.15	22.9	31.5	6.05	6	26.5

Table 8.21. Median concentrations of chromium in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and Major Soil Group.

Median Chromium	Major Soil Group	Environmental Zone						Total	
		Code	1	2	3	4	5		6
Lithomorphic Soils	3		24.9	43.9	26	13.7	8.5	16	21.7
Pelosols	4		47.9	.	.	.	.	.	47.9
Brown Soils	5		30.8	31.4	35.7	38.2	30.5	28.5	32
Podzolic Soils	6		16.5	13.2	22.1	16.9	8.1	8.1	12.2
Surface-water Gley Soils	7		34.7	32.6	18.6	9.8	12.9	14.2	28.65
Ground-water Gley Soils	8		41.6	34.6	.	37.5	36	10.7	36.6
Peat (organic) soils	10		48.7	52.6	4.8	2.2	1.6	3.4	2.6
	Total		32.6	31.2	22.9	31.5	6.05	6	26.5

### 8.3.6.3. Copper

As discussed previously, the concentration values for this metal were not normally distributed (Figure 8.2). The median value was slightly lower (14 mg kg<sup>-1</sup>) than the mean (18 mg kg<sup>-1</sup>) (Table 8.8). Approx. 10% of the CS2000 soil samples contained low concentrations of Cu (< c. 15 mg kg<sup>-1</sup>) while c. 5% contained relatively high Cr concentrations (>60 mg kg<sup>-1</sup>); Figure 8.2. The patterns of distribution for Cu may also apply to Zn.

### Broad Habitat

Soils in broadleaved or mixed woodlands, arable/horticultural and fens/marshes/swamp habitats in EZ 2 of England and Wales, contained the highest median concentrations of Cu while the lowest were mainly associated with coniferous woodland and dwarf shrub heaths located in EZs 4, 5 and 6 in Scotland (Table 8.22.).

### CVS Aggregate Vegetation Class

The highest Cu concentrations were recorded in tall grass and herbs, and lowest in heath/bog and moorland grass mosaic classes (Table 8.23.). In EZs 1 and 2, the highest concentrations were recorded in crops, grasses and woodland soils, and lowest concentrations were recorded in grassland and moorland soils in EZs 5 and 6 in Scotland.

## Major Soil Group

Overall pelosol soils contained the highest median concentrations of Cu and podzols and peat soils contained the lowest Cu median values (Table 8.24.). In EZs 1 and 2, brown and gley soils contained the highest median concentrations of Cu and the lowest median concentrations were measured in peat soils in EZs 5 and 6 in Scotland. McGrath and Loveland (1992) also reported highest concentrations in pelosols, and lowest median values in podzolic and peaty soils.

Table 8.22. Median concentrations of copper in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and Broad Habitat.

Median copper	Environmental Zone							
	Code	1	2	3	4	5	6	Total
Broad Habitat								
Broadleaved, mixed and yew woodland	1	13	24.85	19.9	14.9	5.85	4.9	14.05
Coniferous woodland	2	15.7	11.85	11	6.8	4.95	6.9	7.6
Boundary and linear features	3	.	16.1	.	.	.	.	16.1
Arable and horticultural	4	18	22.65	26.4	20	9.95	.	18.75
Improved grassland	5	15.7	18.2	15.7	11.35	8.9	7.5	15.6
Neutral grassland	6	25.4	15.45	17.35	10.8	10.2	12.2	13.5
Calcareous grassland	7	10	8.65	.	.	.	.	9.9
Acid grassland	8	8.9	15	15.7	11.9	11.35	6.2	8.65
Bracken	9	11.7	10.9	11.9	12.8	.	11.9	11.9
Dwarf shrub heath	10	.	7	17.3	6.6	5	7.8	8
Fen, marsh and swamp	11	.	32.65	27.6	8.7	6.35	4.5	9.25
Bog	12	.	9.9	26.5	7.7	4.5	5.4	5.3
Inland rock	16	34.1	.	.	.	.	.	34.1
Built-up areas and gardens	17	15.3	37.9	27.4	.	.	.	27.4
Supralittoral rock	18	.	170	.	.	7.05	.	8.4
Supralittoral sediment	19	.	12.4	.	.	.	.	12.4
Littoral sediment	21	.	17.9	.	.	.	.	17.9

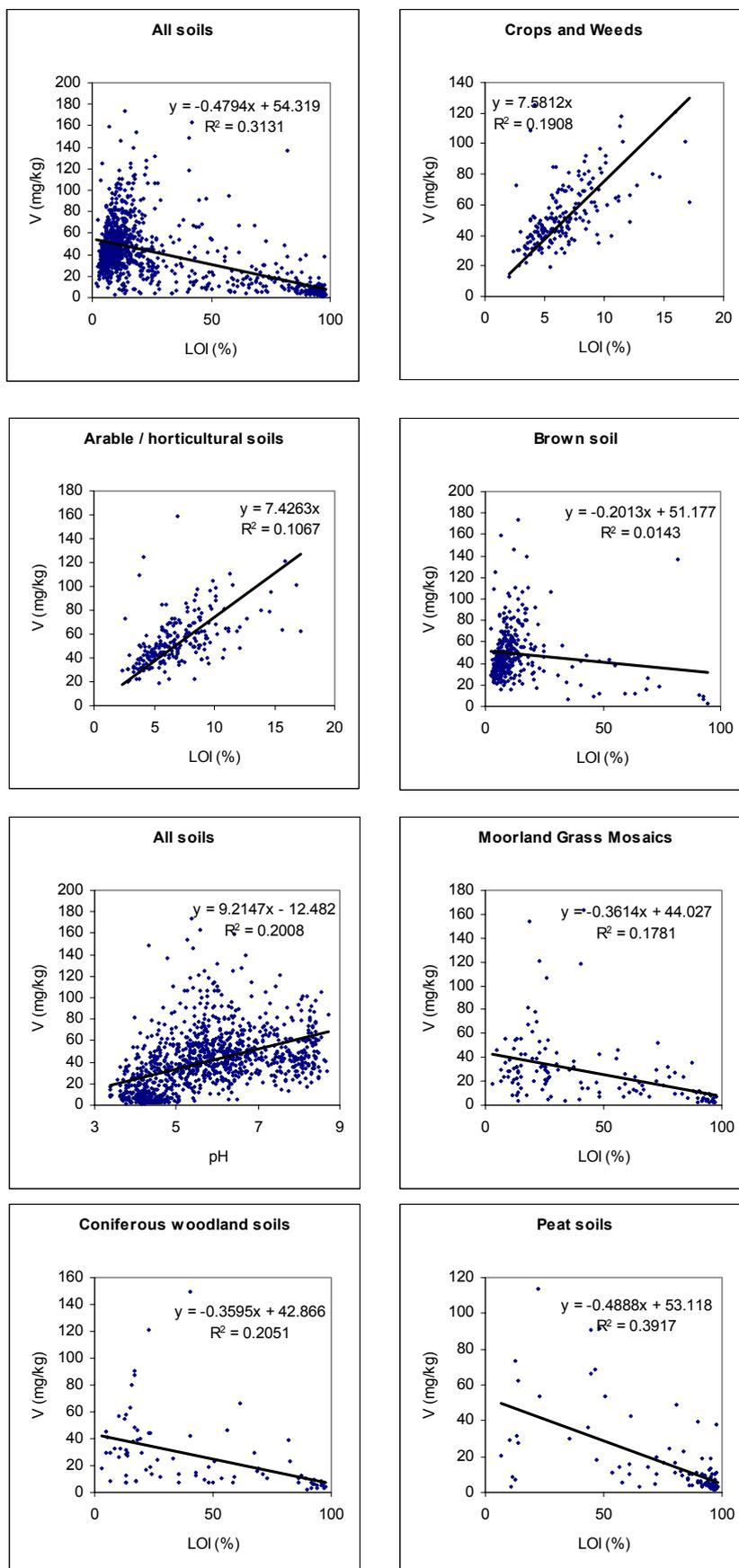
Table 8.23. Median concentrations of copper in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and CVS Aggregate Vegetation Class.

Median copper	Environmental Zone							
	Code	1	2	3	4	5	6	Total
CVS AVC								
Crops and weeds	1	17.2	22.7	26.4	18.9	9.3	.	18.05
Tall grass and herb	2	21.1	29.8	.	22.2	.	.	23.05
Fertile grassland	3	15.9	18.4	17.5	11.5	8.9	.	16.15
Infertile grassland	4	14	16.4	15.5	11.9	8.4	7.5	14.7
Lowland wooded	5	14	20.9	.	16.9	.	.	14.1
Upland wooded	6	14.1	14	14.4	8.3	8.85	9	10.9
Moorland grass mosaics	7	.	40	17	7.75	6.2	8.1	8.35
Heath and bog	8	.	7.5	13.4	8.2	4.4	5.4	5.5

Table 8.24. Median concentrations of copper in soils from CS2000 (mg kg<sup>-1</sup>) categorised by Environmental Zone and Major Soil Group.

Median Copper	Environmental Zone							
	Code	1	2	3	4	5	6	Total
Major Soil Group								
Lithomorphous Soils	3	11.6	22	15.1	7.9	8.1	7.5	10.3
Pelosols	4	19.2	.	.	.	.	.	19.2
Brown Soils	5	15.3	18.35	15.9	17.45	8.2	8.55	16.2
Podzolic Soils	6	11	11.35	14.85	10.9	5.5	6.5	9.7
Surface-water Gley Soils	7	17.4	22.7	15.2	8.7	11	9.1	16.85
Ground-water Gley Soils	8	18.1	19.9	.	14.9	8.4	7.3	14.9
Peat (organic) soils	10	18.6	32.8	22.35	8.9	4.4	5.1	5.1

Figure 8.4. Scatterplots of Vanadium (V) concentrations (mg kg<sup>-1</sup>) plotted against organic matter contents (LOI) for soil from CS2000 in a range of soils and habitats.



### 8.3.6. Integrated analyses

Correlation analyses (Table 8.10; Figure 8.4) indicated that there were a large number of significant correlations between the soil concentrations of the seven heavy metals. Three metals (Cr, Ni and V) were highly correlated with each other, as well as one or two other metals (generally Cu and/or Zn). Zinc and Cu were significantly correlated with each other and also with all other metals. Lead and Cd were significantly correlated with each other and also with Cu, only; Figure 8.3.

Five heavy metals (Ni, Cr, V, Zn and Cu) correlated significantly and positively with soil pH and negatively correlated with soil organic matter content; Table 8.10. Cadmium was the only metal that was positively correlated with soil organic matter but may be influenced by the bi-modal distribution of organic matter in GB soils (Figure 8.2). Soils with high organic content contained low concentrations of Cr and Ni..

This bi-modal distribution may also have influenced other metal relationships with soil organic matter. For example, soil organic matter content in arable/horticultural soils, and those soils under crops and weeds, did not exceed 20%, and were positively related to Vanadium concentrations (Figure 8.4). In contrast, soil organic matter content in moorland grass mosaics, coniferous woodland and peat soils ranged from <5% to 100% and exhibited a negative trend to Vanadium concentrations (Figure 8.4).

## 8.4. Conclusion

A full validation process was carried out between CEH Merlewood and the EA NLS at Llanelli. This has produced excellent results on method comparability and capabilities that will be published in due course. In addition, CEH Merlewood took part in a European Proficiency Testing Scheme with other laboratories that carry out analyses of heavy metals in soil samples. Both exercises established that the analyses of heavy metals at CEH Merlewood would yield data of the highest quality.

During CS2000, 1080 soil samples were successfully analysed for seven heavy metals (Pb, Zn, Cu, Cd, V, Ni). Six of these seven metals analysed in CS2000 were also analysed in the soils of England and Wales by McGrath and Loveland (1992); Cd, Cr, Cu, Ni, Pb, Zn.

Only 12 measurements were observed below the computed limit of detection, 8 for Cd, 3 for Cr and 1 for Ni. The EA NLS laboratory at Llanelli determined values for these samples using ICP-MS. An ORACLE dataset of containing all metal concentrations, with all QC information and relevant metadata, has been fully integrated into the Countryside Survey data management system.

Non-normal distributions of concentrations were observed for all 7 metals. The distribution patterns show similar trends for metals that are normally associated with each other and adjacent in the Periodic Table of elements, i.e., Cd/Pb; V/Cr/Ni and Cu/Zn.

The metal concentrations in the soils of England and Wales were in general lower than those reported by McGrath and Loveland (1992). This may be related to the rural nature

of the CS2000 survey. A relatively small proportion of the soils contained metal concentrations that exceed current maximum limit values.

Mean soil metal concentrations in England and Wales were significantly greater for all metals, except nickel, than those found in Scotland. Median metal concentrations were greatest in pelosols, and least in podzolic and peat soils. Greatest median concentrations were found in soils supporting tall grass and herb species, and least in soils from heath and bog sites. Greatest median concentrations were associated with arable/horticultural and improved grassland soils, and least with dwarf shrub heaths and bog areas.

Future work should now apportion significance to variation in the wider environment; from geology to vegetation characteristics and establish confidence limits to expected concentrations in soils from the GB Countryside. This work should also include more detailed comparisons with published data on heavy metal concentrations in soils and sediments and with data from urban and industrial sites (e.g. EA datasets, URGENT or Environmental diagnostics project sites). Further integrative analyses now need to address the sensitivity of soil biological properties to heavy metal concentrations in relation to other soil and environmental data.

## 9. ORGANIC POLLUTANTS IN SOIL: INITIAL DATA AND ANALYSES

### 9.1. Introduction

A number of studies conducted over the past 40 years have involved the measurement of a range of organic materials in UK soils. Often these studies concentrated on particular situations and chemicals with limited spatial and temporal coverage. For example, a number of studies established the relative persistence of certain pesticides in agricultural or horticultural circumstances. The effort devoted to chemicals of industrial origin has probably been more limited, although surveys of the contamination of certain substances such as PCBs (polychlorinated biphenyls) and dioxins have been done since the 1980s (Cousins *et al*, 1997; Creaser *et al*, 1989; Meharg *et al*, 1998; Zhou *et al* 1998).

Earlier studies of soil contamination, no matter how well designed or conducted, had an important weakness. The sampling regimes were conducted without (it would appear) proper reference to considerations of land use or vegetation type, and with only limited recognition of the potential importance of soil types and their associated biological or physical properties. Moreover, samples were often taken from limited areas of the country, making it difficult to make statements about the national picture or how contamination might vary across the country. Further, within site variation was rarely considered at sites other than historically contaminated ones. In essence, there was no spatially referenced and environmentally characterised dataset to use to determine baseline levels of organic pollutants in soil. In the absence of such information it was difficult to say what constituted "background" or "normal" or "expected" levels of contamination in Great Britain, and, by extension, what levels would be considered outside the normal range.

Accordingly, a major aim of the MASQ project was to examine the levels of organic pollutants in CS2000 soil samples and to determine how the levels of certain POPs (persistent organic pollutants) vary spatially across Great Britain in relation to soil and vegetation type and a number of other broad environmental variables related to geographical location. A select number of the soil samples taken during CS2000 were sub-sampled for the analyses of PCBs, PAHs (polycyclic aromatic hydrocarbons) and a limited number of OCPs (organochlorine pesticides) and their persistent metabolites.

PCBs and PAHs were selected for study because they are both large homologous series of compounds about which sufficient is already known to permit data to be used to test hypotheses about chemical distribution (e.g. about geographical gradients in concentrations, and predictions derived from models of chemical transport and "fate").

The PCBs chosen include the most toxic "dioxin like" congeners and some of the congeners that have been found in birds of prey. The PAHs chosen for analysis were those listed by the US Environmental Protection Agency. The OCPs were chosen for study, because, although prohibitions on their use were put in place some time ago, these compounds are still found in some compartments of the UK environment (e.g. river systems in Yorkshire) and biota (e.g. birds of prey).

### **9.1.1. Specific objectives**

These were to:

- Develop an analytical method for the accurate and efficient analysis of PAHs, PCBs and OCPs in soil.
- Validate the analytical method in collaboration with the Environment Agency (EA) laboratory in Leeds.
- Select soil samples to allow for spatial interpretation of POP levels and to cover the range of environmental circumstances that might have a bearing on the levels of organic contaminants found in soil.
- Analyse of samples for PAHs, PCBs and OCPs.
- Compile an ORACLE database of the results.
- Report results in the form of summary statistics together with brief comments on those statistics and preliminary interpretation of the data so as to indicate what factors may be controlling residue levels.

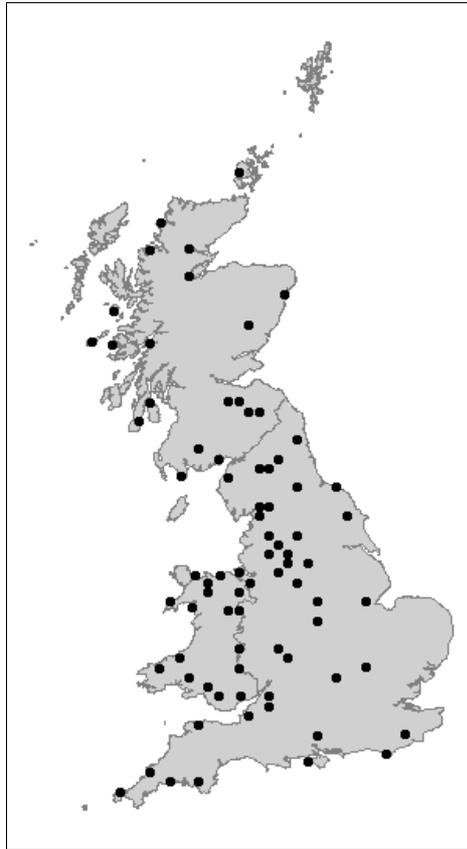
## **9.2. Method**

### **9.2.1. Sample selection**

At the outset of the project, the analysis of organic pollutants was limited to, at most, a quarter (350) of the total number of CS2000 samples taken for MASQ. This limit was due to the availability of funds (estimating costs on the basis on using the, as then, existing Monks Wood analytical methods). Samples were selected to allow for spatial interpretation of POP levels and to cover the range of environmental circumstances that might have a bearing on the levels of organic contaminants found in soil. To obtain the best data for spatial interpretation, the influence of vegetation cover, soil type and recent land use changes was reduced as far as possible. The group of samples that met these criteria most closely were from sites with brown or gley soils, where the vegetation cover (grassland) had been the same on each occasion the Countryside Surveys have been done (of which CS2000 was the latest). The spatial distribution of the samples analysed so far (c.130) is shown in Figure 9.1.

This has been sufficient to provide coverage across Scotland, Wales and England. The gaps, for example, in the south of England and central Scotland will be covered by the next set of samples analysed. The ability to fill all gaps is constrained by the fact that the CS2000 soil samples were themselves only taken from a subset of CS2000 squares. This sub-set of soil samples was selected as it provided a large number of samples from a relatively stable vegetation type. Therefore, the data from these samples will form the main core of the current data package, whatever decisions were made on the numbers and types of further analysis, or the nature of those samples. Further samples will be chosen to include other soil and vegetation cover types e.g. grassland on peat; and to address questions raised by the data collected so far.

Figure 9.1. Locations of the CS2000 1 km squares in Great Britain from which organic pollutant levels in soils were determined.



### 9.2.2. Heterogeneity of organic pollutants in soil

Background levels of contamination are likely to be quite low and experience on industrially contaminated sites shows that residue levels can vary considerably over small distances. Therefore, as this was the first study of its kind in the UK, it was important to establish the within site heterogeneity in the levels of organic pollutants data. A measure of the heterogeneity within a 1 km square can be made using the data from those squares where more than one sample has been taken (20 squares with 2 samples, 8 squares with 3 samples and 4 squares with 4 samples).

In this study the levels in most (83%) of the 1 km squares were represented by analysis of one or two soil samples. Statistical studies on the heterogeneity of data within 1km squares have yet to be done. The heterogeneity on a smaller scale was examined by taking 15 samples from an area of grassland at Monks Wood (approx. 10 ha). This grassland, known as Wilderness II was chosen because its history is reasonably well characterised and it is currently the subject of a long-term succession experiment, which may provide information as to the cause of any heterogeneity. Variation within this field should provide some indication of what might be expected at the sub-1 km scale.

Variation along the vertical soil profile also needs to be investigated at some stage, as this is likely to be significant for organic pollutants. Levels may vary by an order of

magnitude and this could have important implications for interpretation and likely risks for food chains, including human, through uptake by grazing animals and birds of soil, plant and fauna materials.

### 9.2.3. Requirements of the chemical analysis

The work presented a number of analytical challenges, resulting from:

- The small mass or high water content of many of the samples.
- The range of soil and vegetation types involved (lowland and wetland clays to upland peats with a wide range of organic contents from habitats covering grasslands to woodlands).
- The very low levels of POPs likely to be present. (see Table 9.1)
- The need to avoid obtaining a large proportion of "non-detected" values which limit interpretation of the data set.
- The development of sampling and analytical approaches that pass high standard quality assurance procedures, and have quality control procedures that were fit for purpose.

Adopting this approach has been expensive in terms of effort, but worthwhile in terms of the detailed results now being obtained. The project's starting points were recent papers on analytical methods and contaminant levels, together with information already being obtained at CEH Monks Wood on the levels of some of the MASQ suite of chemicals in soils from urban and industrial locations using established Monks Wood methods.

From these sources of information it was concluded that the expected concentration ranges would be between 1 ng/g to 1000 ng/g for PAHs and 0.05 ng/g to 50 ng/g for PCBs and OCPs. To ensure that as few determinands as possible were reported as non-detected the sample based detection limits in a 50 g sample of soil would need to be about 0.2 ng/g (dry weight) for PAHs and about 0.01 ng/g (dry wt) for PCBs and OCPs. Subsequently, it became clear that the analytical method would need to be able to reach these detection limits using only 5 g of soil.

The original design, and costing, of the study agreed between all parties, was based on the use of the methods already available at Monks Wood for analysis of soils (GC-ECD to measure PCBs and OCP pesticides and GC-MS to measure PAHs). Although these methods were well-established at CEH Monks Wood, and could reach the LoDs (limits of detection) for a 50g sample, the emerging EA-led survey running parallel to MASQ, and focusing on urban and industrial locations, led all concerned to invest in the development of a GCMS technique for PCBs and OCPs in the CS2000 soil samples. This change meant that CEH had to (a) improve its sample cleanup methodology to meet the required LoDs for OCPs, PCBs and PAHs, and (b) purchase new GCMS instrumentation capable of large volume injection and negative chemical ionisation (to obtain still lower detection limits).

Table 9.1 List of Analytes

2,4'-Dichlorobiphenyl (PCB 8)	Naphthalene
2,2',5'-Trichlorobiphenyl (PCB 18)	Acenaphthylene
2,4,4'-Trichlorobiphenyl (PCB 28)	Acenaphthene
2,4,5'-Trichlorobiphenyl (PCB 29)	Fluorene
2,4',5'-Trichlorobiphenyl (PCB 31)	Phenanthrene
2,2',5,5'-Tetrachlorobiphenyl (PCB 52)	Anthracene
3,3',4,4'-Tetrachlorobiphenyl (PCB 77)	Fluoranthene
2,2',4,5,5'-Pentachlorobiphenyl (PCB 101)	Pyrene
2,3,3',4,4'-Pentachlorobiphenyl (PCB 105)	Benzo[ <i>a</i> ]anthracene
2,3,4,4',5'-Pentachlorobiphenyl (PCB 114)	Chrysene
2,3',4,4',5'-Pentachlorobiphenyl (PCB 118)	Benzo[ <i>b</i> ]fluoroanthene
2',3,4,4',5'-Pentachlorobiphenyl (PCB 123)	Benzo[ <i>k</i> ]fluoranthene
3,3',4,4',5'-Pentachlorobiphenyl (PCB 126)	Benzo[ <i>a</i> ]pyrene
2,2',3,3',4,4'-Hexachlorobiphenyl (PCB 128)	Ideno[1,2,3- <i>cd</i> ]pyrene
2,2',3,4,4',5'-Hexachlorobiphenyl (PCB 138)	Dibenz[ <i>a,h</i> ]anthracene
2,2',3,4,5,5'-Hexachlorobiphenyl (PCB 141)	Benzo[ <i>g,h,i</i> ]perylene
2,2',3,4',5',6'-Hexachlorobiphenyl (PCB 149)	
2,2',4,4',5,5'-Hexachlorobiphenyl (PCB 153)	
2,3,3',4,4',5'-Hexachlorobiphenyl (PCB 156)	HCB
2,3,3',4,4',5'-Hexachlorobiphenyl (PCB 157)	alpha-HCH
2,3,3',4',5,6'-Hexachlorobiphenyl (PCB 163)	gamma-HCH
2,3',4,4',5,5'-Hexachlorobiphenyl (PCB 167)	HEOD
3,3',4,4',5,5'-Hexachlorobiphenyl (PCB 169)	p,p'-DDE
2,2',3,3',4,4',5'-Heptachlorobiphenyl (PCB 170)	p,p'-DDT
2,2',3,3',4,4',6'-Heptachlorobiphenyl (PCB 171)	p,p'-DDD
2,2',3,4,4',5,5'-Heptachlorobiphenyl (PCB 180)	
2,2',3,4,4',5',6'-Heptachlorobiphenyl (PCB 183)	
2,2',3,4',5,5',6'-Heptachlorobiphenyl (PCB 187)	
2,2',3,3',4,4',5,5'-Octachlorobiphenyl (PCB 194)	
2,2',3,3',4,5,6,6'-Octachlorobiphenyl (PCB 199)	
2,2',3,3',4,5,5',6'-Octachlorobiphenyl (PCB 201)	
2,3,3',4,4',5,5',6'-Octachlorobiphenyl (PCB 205)	
2,2',3,3',4,4',5,5',6'-Nonachlorobiphenyl (PCB 206)	
Decachlorobiphenyl (PCB 209)	

#### 9.2.4. Summary of the method for the determination of persistent organic pollutants in soil

##### Reagents

Anhydrous sodium sulphate (Merck) prepared by heating at 700°C for 4 hours. Aluminium oxide (Merck) prepared by heating at 800°C for 4 hours. Dichloromethane (DCM) HPLC grade (Rathburns), n-hexane 97% grade (Rathburns), and acetone, HPLC grade (Rathburns) were used as supplied.

## **Determination of water content**

A portion of the sample was taken for gravimetric determination of water content. Approx. 1 g of the sample was weighed onto a piece of aluminium foil using a four place electronic balance. The sample was then dried in an oven at 80°C for 48 hours and re-weighed.

## **Extraction**

If there was a delay between sampling and analysis, samples were stored at -20°C. The sample was not dried prior to extraction in order to minimise the loss of the more volatile components and to reduce contamination from the atmosphere. The sample (a wet weight equivalent to approx. 5g dry weight) was weighed into a beaker and <sup>13</sup>C<sub>12</sub> PAH and <sup>13</sup>C<sub>12</sub> PCB surrogate standards were added. The soil was then dried with anhydrous sodium sulphate and transferred to a glass microwave extraction vessel. Any soil residues left in the beaker were transferred to the extraction vessel with 3 x 10ml of DCM. A further 10ml of DCM was added to the vessel. The soil was extracted by open vessel microwave extraction (Prolabo) for 20mins. A drying column containing anhydrous sodium sulphate was prepared. The whole contents of the extraction vessel were filtered through the drying column. The extraction vessel was then rinsed with 3 x 10ml of DCM and the rinsings passed through the column. The column was then rinsed with 2 x 10ml of DCM. The filtered extract was then reduced to approx. 0.5ml under a stream of nitrogen (Electron capture grade) in a TurboVap evaporator at a temperature of 45°C.

A 10mm id glass open chromatography column was slurry packed with 4.2g of activated alumina in DCM and 1cm of anhydrous sodium sulphate. The column was rinsed with 20ml of DCM and the extract transferred from the TurboVap vessel to the top of the column with a glass pasteur pipette. The TurboVap vessel was rinsed with 3 x 0.5ml of DCM and the rinsings added to the top of the column. The analytes were eluted from the column with 15ml of DCM into a Kurdena-Danish flask. The cleaned up extract was then reduced to approx. 1ml in the Kurdena-Danish flask at a temperature of 60°C and reduced to 0.4ml under a stream of nitrogen. The whole extract was transferred to a 1.5 ml chromatography vial using a syringe to give a final volume of 0.5ml.

## **Cleanup by size exclusion chromatography (SEC)**

Further cleanup was carried out by automated high-resolution size exclusion chromatography. The HPLC instrument was an Agilent 1100 HPLC system consisting of vacuum degasser, quaternary pump, autosampler, column oven, UV diode array detector, 6-port 2-position low dead volume switching valve and a Foxy Junior fraction collector. The flow path is shown in Figure 9.2. The fraction collector's plastic tubing was replaced with highly flexible stainless steel and the dropper head was replaced with a stainless steel one. All plumbing in the system was of stainless steel. The SEC column consists of a 300mm x 7.8mm Envirosep ABC column (Phenomenex) protected by a 50mm x 7.8mm Envirosep ABC guard column (Phenomenex). The column was held at

a constant temperature of 30°C and the eluant was 1ml/min of DCM. The total run time was 18mins and the analytes elute between 8.45 to 14.00 mins. The 0.5ml of sample was cleaned up by SEC by running it three times. The first two runs inject 400ul of the sample and the third run injects the last 100ul of the sample and washes any remaining sample in the vial onto the SEC column using DCM from a separate vial.

### **Fractionation of Extract into PAHs and PCBs**

Before cleanup by HPLC the extract solvent was changed from DCM to n-hexane and reduced in volume. The three fractions collected from the SEC were combined in a Kurdena-danish with 15ml of n-hexane and then reduced to approx. 0.5ml at a temperature of 80°C. The reduced extract was adjusted to 0.4ml under a stream of nitrogen. The whole extract was transferred to a 1.5ml-chromatography vial using a syringe to give a final volume of 0.6ml. The cleaned up extract was then separated by HPLC (4.6mm id x 250mm Nucleosil NO<sub>2</sub>) into three fractions containing aliphatic/mono-cyclic aromatic compounds, PAHs and PCBs. OCPs were distributed between the PCB and the PAH fractions. The HPLC was same as that used for the SEC except that the column was plumbed to enable the reversal of the eluant flow (see Figure 9.3). The column was a 4.6mm id x 250mm Nucleosil NO<sub>2</sub> with 1ml/min n-hexane as the mobile phase. With the mobile phase flowing in the forward direction at 1ml/min the PCBs elute between 3.5 and 13 mins. The mobile phase was then reversed and the flow increased to 2ml/min eluting the PAHs between 17.5 and 21.5mins. The flow was switched back to the forward direction and returned to 1ml/min for a further seven minutes. The PCB and PAH fractions were collected by the fraction collector.

### **Analysis by GC-MS**

The PCB and PAH fractions were reduced by Kurdena-danish at 80 °C to approx. 0.5 ml. The PAH and PCB fractions were adjusted to 500 and 240 ul respectively. 120 ul of each fraction were taken for analysis by GCMS. The rest of each fraction was transferred to chromatography vials and refrigerated. The two fractions were analysed on dedicated instruments.

Each instrument comprises of an Agilent 6890 gas chromatograph, 5973 mass selective detector, 6873 autosampler and programmable temperature vaporising inlet. Each detector was run in electron impact SIM mode at the highest mass resolution available. The PAHs fraction was separated on a 30m x 0.25mm id HP5-MS column (Agilent) fitted with a 5m x 0.25mm id Siltek deactivated guard column (Restek).

The PCB fraction was separated on a 50 m x 0.22 mm id HT8 column (SGE) fitted with a 5 m x 0.25 mm id Siltek deactivated guard column (Restek). Where possible, three ions were monitored for each compound. Injection (100 ul) was carried out using the solvent vent technique. Whilst holding the inlet at 20°C, 5 injections of 20ul were made at 10 s intervals. After most of the solvent has been vented the inlet temperature was increased to 350°C at 750°C/min to transfer the analytes to the column.

Figure 9.2. Schematic of the SEC system

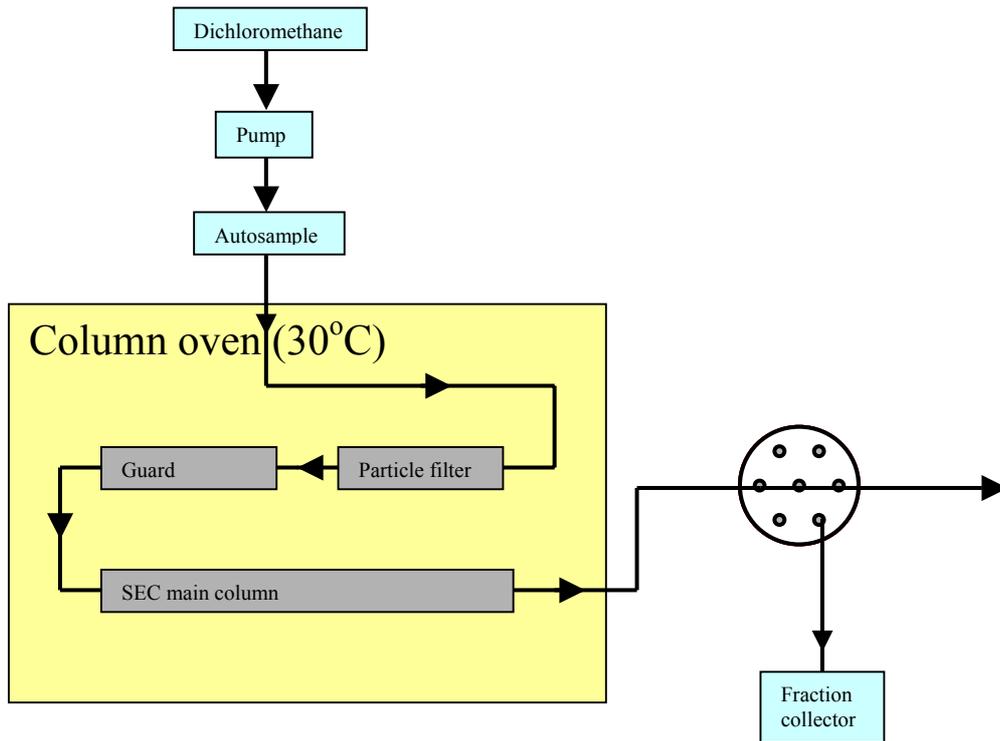
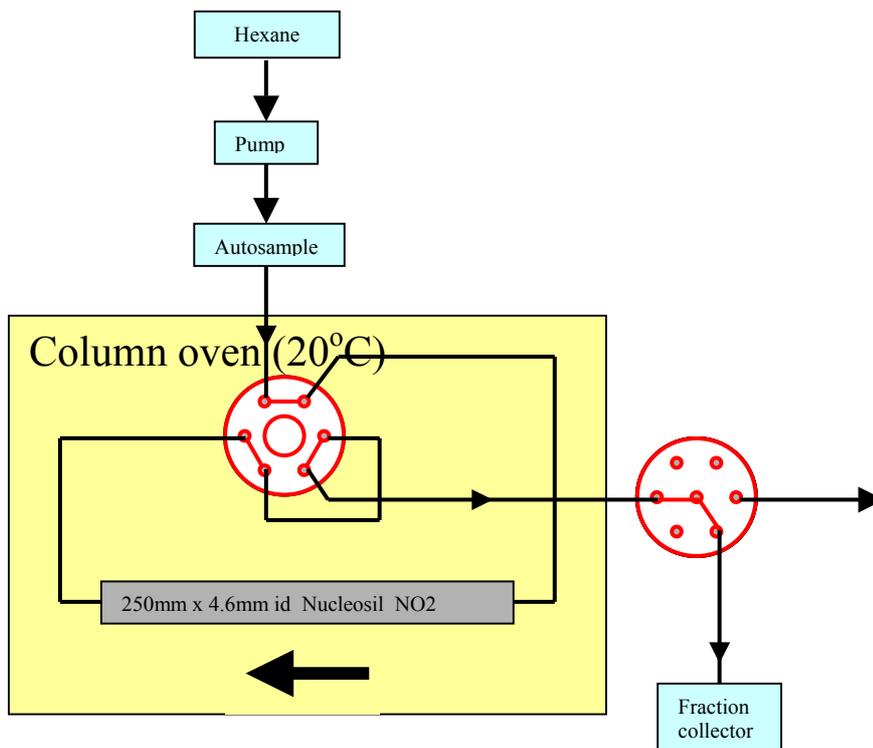


Figure 9.3. Schematic of the HPLC system for the fractionation of PAHs and PCBs



<sup>13</sup>C<sub>12</sub> PAH and PCB standards were used as internal standards and to calculate losses during analysis (isotope dilution). Losses for OCPs were determined using soil spiked with native standards. Each batch of 14 samples included a sample blank, a sample of reference material SETOC 738, a spiked forest soil and a forest soil control. LoDs for the method were determined by the analysis of 7 sample blanks. The LoDs were calculated as the mean of the sample blanks plus 3 standard deviations. The method being used at present was capable of reaching the required LoD and is suitable for all the soil types in MASQ. Despite the extensive clean-up procedures, PCB congeners 167 and 205 could not be quantified due to the presence of interfering substances.

### Method validation

QA was conducted in association with the EA laboratory at Leeds. A suitable Certified Reference Material could not be obtained from commercial sources for the analysis due to import restrictions for soils from outside the EU. The EA Laboratory at Leeds kindly supplied Monks Wood with a Reference Material of estuarine sediment (SETOC 738). Although the reference material was not soil, and the residue concentrations were much higher than those that were expected to occur in the soil samples, it was still a very good test of the methodology and satisfactory for inter-laboratory comparisons.

Six samples of SETOC 738 were analysed for a limited set of PCBs and PAHs. CEH Monks Wood and the EA results of the analysis of SETOC 738 were exchanged and both parties were satisfied with the other's results. Tables 9.2 and 9.3 show the results of reference material analysis from both laboratories along with the reference values. The mean values between the CEH Monks Wood and EA results and the published results for SETOC 738 were comparable. The CEH Monks Wood results for PCBs show lower precision for most of the PCB congeners. The EA labs mean value for benzo[*b*]fluoranthene was higher, probably because this value also includes benzo[*j*]fluoranthene. Arrangements were made with the EA Leeds laboratory for inter-laboratory comparisons using CS2000 soil samples (approx. 30 samples). However the EA laboratory have not so far been able to start this due to the pressure of work. The MASQ samples will remain in storage at CEH Monks Wood until requested by the EA Leeds laboratory.

Table 9.2. Summary of Reference Material data for PCBs. Means expressed as ng/g dry weight.

	SETOC 738			EA Leeds			CEH Monks Wood		
	Mean	% RSD	N	Mean	% RSD	N	Mean	% RSD	N
PCB 28	5.13	23.0	18	4.21	6.6	6	3.23	34.5	6
PCB 31	2.50		1	4.21	6.6	6	4.27	33.8	6
PCB 52	6.00	19.0	22	5.14	14.2	6	4.52	27.8	6
PCB 101	7.08	21.0	29	6.92	9.6	6	5.94	20.2	6
PCB 105	1.80		3	1.07	7.1	6	1.04	25.2	6
PCB 118	4.97	13.0	18	4.61	8.2	6	4.18	24.0	6
PCB 128	1.95		2	1.22	7.5	6	1.33	9.5	6
PCB 138	8.24	31.0	29	7.52	10.7	6	6.46	11.3	6
PCB 149	6.93		3	N/A			7.39	11.8	6
PCB 153	9.05	31.0	31	11.13	11.4	6	9.36	10.2	6
PCB 156	0.93		3	0.80	11.2	60	0.58	10.0	6
PCB 180	4.75	21.0	22	5.03	17.6	6	5.85	23.0	6

Table 9.3. Summary of Reference Material data for PAHs. Means were expressed as ng/g dry weight.

	SETOC 738			EA Leeds			CEH Monks Wood		
	Mean	% RSD	N	Mean	% RSD	N	Mean	% RSD	N
Phenanthrene	1375	18	45	1302	14	5	1371	12	6
Anthracene	287	29	41	253	12	5	218	19	6
Fluoranthene	3506	18	38	4242	11	5	3485	8	6
Pyrene	2641	16	44	3332	12	5	2509	8	6
Benzo[ <i>a</i> ]anthracene	1609	22	48	1516	11	5	1617	8	6
Chrysene	1707	22	49	1533	8	5	1929	8	6
Benzo[ <i>b</i> ]fluoranthene	1897	18	35	2601	7	5	1793	9	6
Benzo[ <i>k</i> ]fluoranthene	955	16	39	969	8	5	1443	8	6
Benzo[ <i>a</i> ]pyrene	1647	19	48	1604	11	5	1878	12	6
Ideno[1,2,3- <i>cd</i> ] pyrene	1352	24	46	1554	8	5	1223	7	6
Dibenz[ <i>a,h</i> ]anthracene	309	28	44	257	8	5	281	6	6
Benzo[ <i>g,h,i</i> ]perylene	1208	21	46	1680	5	5	1197	8	6

### Limits of detection

Table 9.4, shows the method LoDs for 5g of dry soil. Of the LoDs for PCBs, half were at or below the desired LoD (0.01ng/g), the other half were very close to the desired LoD and below the levels expected in the samples. The LoD for PCB 180 (0.055ng/g) was above the minimum concentration expected in the samples, but was low enough to give 85% positive results. Of the total number of determinations for PCBs (excluding the data for PCBs 167 and 205), 76% of the results were positives. The majority of the non-detected values were for PCB congeners 123, 126 and 169. The LODs achieved for PAHs were higher than expected, however of the total number of determinations for PAHs, the proportion of results that were positives (95%) was still high.

Table 9.4. Method LoDs

Analyte	LoD (ng/g)	Analyte	LoD (ng/g)	Analyte	LoD (ng/g)
PCB 8	0.019	PCB 128	0.005	Phenanthrene	17.583
PCB 18	0.023	PCB 167	0.030	Anthracene	12.947
PCB 29	0.002	PCB 171	0.005	Fluoranthene	4.388
PCB 28	0.020	PCB 199	0.003	Pyrene	3.528
PCB 31	0.017	PCB 156	0.003	Benzo[ <i>a</i> ]anthracene	1.114
PCB 52	0.010	PCB 157	0.002	Chrysene	1.423
PCB 101	0.012	PCB 180	0.055	Benzo[ <i>b</i> ]fluoranthene	1.667
PCB 77	0.003	PCB 201	0.018	Benzo[ <i>k</i> ]fluoranthene	1.215
PCB 149	0.018	PCB 170	0.019	Benzo[ <i>a</i> ]pyrene	3.237
PCB 123	0.014	PCB 169	0.008	Ideno[1,2,3- <i>cd</i> ]pyrene	2.826
PCB 118	0.010	PCB 189	0.002	Dibenz[ <i>a,h</i> ]anthracene	1.800
PCB 114	0.004	PCB 194	0.017	Benzo[ <i>g,h,i</i> ]perylene	3.028
PCB 153	0.033	PCB 205	-		
PCB 141	0.010	PCB 206	0.006	gamma-HCCH	0.028
PCB 105	0.006	PCB 209	0.002	HCB	0.294
PCB 163	0.007			alpha-HCCH	0.018
PCB 138	0.015	Naphthalene	1.890	p,p-DDE	0.104
PCB 187	0.027	Acenaphthylene	0.630	HEOD	0.541
PCB 183	0.013	Acenaphthene	0.288	p,p-TDE	0.075
PCB 126	0.003	Fluorene	0.242	p,p-DDT	0.463

### 9.2.5. Analyses of remaining soil samples

MASQ sample analyses are to be completed, and c. 80 samples will constitute the next analytical batch. These samples will be of 3 kinds:

- Peat soils from both the uplands and lowlands
- Grassland soil samples that may help fill gaps in the existing distribution pattern shown in Figure 9.1.
- Samples chosen to test hypotheses generated from the current data.

Thus, analytical work in-hand (in addition to on-going discussions with EA Leeds analysts) includes:

- Re-analysis of the six samples that have unusual analytical characteristics, to quantify analytes.
- Capture of data for two PAHs that were found in a clean-up fraction other than that originally expected
- *Completion of QA for the new ASE procedure.*

For the future, analysis time will be shortened by replacing microwave extraction with Accelerated Solvent Extraction (ASE) and by the use of automated solvent evaporation apparatus. This will result in the loss of naphthalene, but the advantages of the reduction in analysis time will outweigh the disadvantage of the loss of a PAH that is of little importance. The work to assess the suitability of ASE for this analysis is almost complete and the results will be reported separately.

There is insufficient reference material SETOC 738 to analyse along with the remaining CS2000 soil samples. It will be replaced by a certified reference material from the National Institute of Standards and Technology, USA (SRM 1944, New York/New Jersey Waterway Sediment).

To ensure that the project QA is not affected by any of these changes, repeat analysis within the same batch of samples will be made of SETOC 738 and SRM 1944 and recovery of  $^{13}\text{C}$  spikes.

## 9.3. Results

### 9.3.1. Progress to date

The work completed so far is:

- The selection of samples to maximise the scientific output
- The development of a method that meets the analytical requirements
- In partnership with the EA, the establishment of suitable QA procedures to measure the quality of the results and to compare the methods of CEH Monks Wood and EA Leeds laboratory.
- The analysis of a Reference Material to establish compatibility between the EA Leeds and CEH Monks Wood laboratories.
- A study to examine the variability in some of the analytes over a small area, from soil samples collected in part of a 1 km square at Monks Wood, Cambs.

- The analysis of 123 CS2000 soil samples.
- The preliminary interpretation of data and the compilation of a database.

Some 35 PCB congeners and 14 PAHs have been quantified in the CS2000 and Monks Wood soil samples, along with 7 OCP pesticides or their persistent metabolites in the CS2000 soil samples (58 different compounds in total - more than originally envisaged). In addition, PCBs and PAHs have been quantified in a Reference Material. Preliminary studies indicate many more organic compounds were present in both the Reference material and the Monks Wood samples. These compounds include at least one pesticide still in current use. Extracts of soil samples were kept for further studies aimed at identifying currently unknown organic compounds in the CS2000 soil samples.

### 9.3.2. Monks Wood samples from Wilderness II

Within field heterogeneity of PCB 180 and PAH, benzo[*a*]pyrene are illustrated in Figure 9.4. Negative values show results that were below the values that could be determined allowing for the effect of the blank. These samples contain very low concentrations or no PCB 180. The results shown in the two diagrams were typical of the other compounds examined. Within field heterogeneity appears to be greater for PCBs than PAHs. For example, two samples taken about 10m apart appear to differ by more than an order of magnitude. The situation was less variable for PAHs, where differences between neighbouring samples were about three-fold.

### 9.3.3. Concentrations of persistent organic pollutants across Britain

Of the 123 CS2000 soil samples analysed, 6 require partial re-analysis to confirm suspicious or very high results and therefore, for the following data analysis, 117 samples have been used. Table 9.5 shows the distribution of the samples amongst the various stratifications of the GB countryside. Stratifications with less than 7 data points and organics with a high proportion of non-detectables have not been assessed in this report. The non-parametric Kruskal-Wallis test was used to determine significant differences between the classifications;  $P < 0.05$  indicates a significant difference. The descriptive statistics for POPs in CS2000 soil samples across Britain are shown in Tables 9.6 to 9.8. For individual PCB congeners (Table 9.6), median values ( $>0$ ) range from 0.002ng/g (PCB 189) to 0.2ng/g (PCB 153). Based on median values, the dominant congeners were 118, 138, 153, 180 and 187. The list of dominant congeners is based on mean values consists of congeners 8, 18, 28, 31, 138, 153 and 180, shifting the balance to the less chlorinated PCBs. These differences may be due different patterns of congeners across the country. PCB 169 was detected in only one sample. PCBs 126 and 123 were detected in few samples.

For individual PAHs (Table 9.7), median values range from 0.8ng/g (Acenaphthylene) to 45ng/g (Chrysene). The dominant PAHs were those of higher molecular mass, with the exception of Dibenzo[*a,h*]anthracene. Patterns of PAHs produced by mean and median values were similar. Median values for Total PAHs were two orders of magnitude higher than those for Total PCBs. The maximum values of individual PAHs and Total PAHs were roughly three orders of magnitude higher than for PCBs. It is

interesting to note that DDT is still present in UK soils, although it has been illegal to use DDT since the early 1980's.

Table 9.5. Numbers of samples in each classification

Major Soil Group	N	Aggregated vegetation class	N	Broad habitat	N	Environmental zone	N
Brown soils	70	Fertile grassland	46	Acid grassland	4	Zone 1	13
Gleys	47	Infertile grassland	56	Arable and horticultural	2	Zone 2	61
		Moorland grass mosaic	15	Bog	2	Zone 3	24
				Broadleaved, mixed and yew woodland	4	Zone 4	6
				Built-up areas and gardens	1	Zone 5	12
				Dwarf shrub heath	2	Zone 6	3
				Fen, marsh and swamp	7		
				Improved grassland	87		
				Neutral grassland	10		

Table 9.6. Summary data for PCB concentrations (ng/g dry weight) in Britain from CS2000 soil samples

PCB	Mean	Stdev	Median	Min	Max	N
Total PCB	4.140	6.546	2.080	0.000	45.900	119
PCB 8	0.244	0.467	0.073	0.000	3.467	117
PCB 18	0.341	0.735	0.061	0.000	5.040	117
PCB 28	0.389	0.910	0.070	0.000	6.667	117
PCB 31	0.341	0.796	0.054	0.000	5.631	117
PCB 52	0.141	0.271	0.059	0.000	2.110	117
PCB 77	0.024	0.053	0.010	0.000	0.475	117
PCB 101	0.133	0.186	0.076	0.000	1.214	117
PCB 105	0.092	0.224	0.039	0.000	2.271	117
PCB 114	0.007	0.021	0.000	0.000	0.153	117
PCB 118	0.180	0.387	0.076	0.000	3.834	117
PCB 128	0.079	0.202	0.038	0.000	2.035	117
PCB 138	0.308	0.621	0.158	0.000	5.539	117
PCB 141	0.072	0.167	0.033	0.000	1.333	117
PCB 149	0.197	0.434	0.085	0.000	3.513	117
PCB 153	0.376	0.658	0.195	0.020	4.828	117
PCB 156	0.036	0.112	0.015	0.000	1.141	117
PCB 157	0.016	0.047	0.004	0.000	0.344	117
PCB 163	0.109	0.207	0.056	0.000	1.516	117
PCB 170	0.126	0.337	0.049	0.000	2.931	117
PCB 171	0.035	0.092	0.014	0.000	0.818	117
PCB 180	0.308	0.935	0.112	0.000	8.938	117
PCB 183	0.073	0.206	0.028	0.000	1.915	117
PCB 187	0.195	0.490	0.081	0.000	4.395	117
PCB 189	0.008	0.023	0.002	0.000	0.158	117
PCB 194	0.083	0.275	0.027	0.000	2.589	117
PCB 199	0.016	0.048	0.005	0.000	0.416	117
PCB 201	0.124	0.337	0.047	0.000	2.870	117
PCB 206	0.050	0.103	0.023	0.000	0.698	117
PCB 209	0.068	0.120	0.034	0.004	0.857	117

Figure 9.4. Variability in the concentration of organics within the Monks Wood field; figures in the sample grid are ng/g dry wt.

**PCB 180**

<b>Edge</b>		<b>-0.1</b>		<b>-0.9</b>		
<b>Track</b>						
<b>10</b>		<b>-0.4</b>		<b>0.2</b>		<b>-0.7</b>
<b>9</b>						
<b>8</b>				<b>0.7</b>		<b>5.7</b>
<b>7</b>						
<b>6</b>				<b>-0.4</b>		<b>-1.0</b>
<b>5</b>						
<b>4</b>		<b>-0.8</b>		<b>-0.7</b>		<b>2.4</b>
<b>3</b>	<b>0.4</b>					
<b>2</b>				<b>16.8</b>		<b>-0.5</b>
<b>1</b>						
	<b>A</b>	<b>B</b>	<b>C</b>	<b>D</b>	<b>E</b>	<b>F</b>

**Benzo[a]pyrene**

<b>Edge</b>		<b>16.75</b>		<b>17.79</b>		
<b>Track</b>						
<b>10</b>		<b>9.75</b>		<b>12.26</b>		<b>6.32</b>
<b>9</b>						
<b>8</b>				<b>22.82</b>		<b>14.22</b>
<b>7</b>						
<b>6</b>				<b>9.58</b>		<b>9.87</b>
<b>5</b>						
<b>4</b>		<b>15.07</b>		<b>14.38</b>		<b>15.52</b>
<b>3</b>	<b>14.83</b>					
<b>2</b>				<b>12.90</b>		<b>20.64</b>
<b>1</b>						
	<b>A</b>	<b>B</b>	<b>C</b>	<b>D</b>	<b>E</b>	<b>F</b>

Table 9.7. Summary data for PAH concentrations (ng/g dry weight) in Britain from CS2000 soil samples

	Mean	Stdev	Median	Min	Max	N
Total PAH	3507.350	29003.000	438.330	13.380	317019.000	119
Naphthalene	34.459	64.891	13.323	0.000	484.710	117
Acenaphthylene	4.160	10.650	0.842	0.000	87.066	117
Acenaphthene	14.743	30.683	4.144	0.000	232.014	117
Fluorene	13.108	24.982	4.304	0.000	149.534	117
Fluoranthene	95.384	162.355	35.125	0.000	1087.170	117
Pyrene	79.274	117.852	33.308	0.000	751.637	117
Benzo[ <i>a</i> ]anthracene	77.795	128.427	28.315	0.000	634.828	117
Chrysene	107.134	148.289	45.411	0.000	843.936	117
Benzo[ <i>b</i> ]fluoranthene	108.780	170.523	42.188	4.925	953.128	117
Benzo[ <i>k</i> ]fluoranthene	42.618	94.456	15.949	0.000	799.626	117
Benzo[ <i>a</i> ]pyrene	85.216	124.195	35.523	0.000	691.367	117
Ideno[1,2,3- <i>cd</i> ]pyrene	82.335	114.975	38.091	0.000	565.626	116
Dibenz[ <i>a,h</i> ]anthracene	15.982	25.948	6.549	0.000	132.037	116
Benzo[ <i>g,h,i</i> ]perylene	76.888	102.245	36.399	0.000	507.348	116

Table 9.8. Summary data for OCP concentrations (ng/g dry weight) in Britain from CS2000 soil samples

	Mean	Stdev	Median	Min	Max	N
Total OCP	3.560	13.930	0.000	0.000	124.280	119
p,p-DDE	1.914	4.429	0.373	0.000	26.573	117
p,p-DDT	3.457	13.977	0.000	0.000	124.283	117

### 9.3.3.1. England, Wales and Scotland

Tables 9.9 to 9.11 show the data for England and Wales combined and compared to that from Scotland. The median total PCB levels in England and Wales were slightly lower than in Scotland, but the difference was not statistically significant. There was no overall significant difference in concentrations between Scotland and Wales except for four of the more chlorinated PCB congeners (170, 171, 180 and 201). Total PAH median concentrations were approximately twice as high in England and Wales than in Scotland, but this difference was not statistically significant. Most of the individual PAHs were higher in England and Wales than in Scotland and some significantly so. There were no significant differences between OCP concentrations.

Table 9.9. Comparison of PCB concentrations (ng/g dry weight) between England and Wales and Scotland

Country	England & Wales N = 96		Scotland N = 21		P-value (Median)	Significant
	Mean	Median	Mean	Median		
Total PCB	3.776	2.080	5.837	3.060	0.805	No
PCB 8	0.168	0.070	0.588	0.083	0.387	No
PCB 18	0.224	0.061	0.877	0.060	0.217	No
PCB 28	0.244	0.068	1.056	0.071	0.250	No
PCB 31	0.217	0.054	0.910	0.067	0.509	No
PCB 52	0.103	0.060	0.312	0.053	0.424	No
PCB 77	0.018	0.010	0.050	0.011	0.829	No
PCB 101	0.133	0.078	0.129	0.071	0.915	No
PCB 105	0.092	0.039	0.088	0.030	0.624	No
PCB 114	0.005	0.000	0.016	0.000	0.723	No
PCB 118	0.182	0.087	0.168	0.066	0.544	No
PCB 128	0.083	0.039	0.064	0.027	0.288	No
PCB 138	0.323	0.161	0.238	0.108	0.227	No
PCB 141	0.077	0.035	0.052	0.025	0.295	No
PCB 149	0.211	0.089	0.132	0.071	0.450	No
PCB 153	0.396	0.211	0.283	0.138	0.147	No
PCB 156	0.037	0.016	0.032	0.013	0.330	No
PCB 157	0.016	0.005	0.016	0.003	0.643	No
PCB 163	0.113	0.056	0.090	0.042	0.373	No
PCB 170	0.139	0.055	0.068	0.030	0.014	Yes
PCB 171	0.037	0.014	0.026	0.005	0.019	Yes
PCB 180	0.344	0.119	0.140	0.070	0.020	Yes
PCB 183	0.079	0.031	0.042	0.017	0.053	No
PCB 187	0.212	0.089	0.117	0.056	0.069	No
PCB 189	0.007	0.002	0.013	0.000	0.198	No
PCB 194	0.091	0.030	0.042	0.017	0.133	No
PCB 199	0.017	0.006	0.015	0.003	0.094	No
PCB 201	0.137	0.052	0.063	0.020	0.002	Yes
PCB 206	0.052	0.023	0.040	0.016	0.086	No
PCB 209	0.062	0.034	0.093	0.025	0.310	No

Table 9.10. Comparison of PAH concentrations (ng/g dry weight) between England and Wales and Scotland

Country PAH	England and Wales N = 97		Scotland N = 21		P-value (Median)	Significant
	Mean	Median		Median		
Total PAH	4147.910	520.860	518.060	231.880	0.056	No
Naphthalene	35.962	12.704	27.586	25.607	0.289	No
Acenaphthylene	4.523	0.901	2.500	0.664	0.565	No
Acenaphthene	15.683	3.719	10.446	9.659	0.186	No
Fluorene	14.478	4.230	6.845	5.359	0.887	No
Fluoranthene	102.162	33.018	62.510	37.733	0.562	No
Pyrene	84.605	34.609	53.420	31.480	0.879	No
Benzo[ <i>a</i> ]anthracene	88.451	34.953	26.109	13.768	0.048	Yes
Chrysene	118.627	68.828	51.392	26.311	0.063	No
Benzo[ <i>b</i> ]fluoranthene	115.855	54.679	74.463	28.164	0.100	No
Benzo[ <i>k</i> ]fluoranthene	39.221	17.036	59.093	7.701	0.045	Yes
Benzo[ <i>a</i> ]pyrene	96.367	43.739	31.133	17.102	0.021	Yes
Ideno[1,2,3- <i>cd</i> ]pyrene	86.314	44.794	62.018	20.217	0.115	No
Dibenz[ <i>a,h</i> ]anthracene	17.116	7.585	10.190	3.068	0.050	Yes
Benzo[ <i>g,h,i</i> ]perylene	82.323	49.697	49.138	19.852	0.095	No

Table 9.11. Comparison of OCP concentrations (ng/g dry weight) between England and Wales and Scotland

Country OCP	England and Wales N = 96		Scotland N= 21		P-value (Median)	Significant
	Mean	Median	Mean	Median		
Total OCP	3.586	0.000	3.441	0.000	0.845	No
p,p-DDE	1.667	0.330	3.044	0.865	0.123	No
p,p-DDT	3.481	0.000	3.345	0.000	0.882	No

### 9.3.3.2. Environmental Zones

Tables 9.12 to 9.14 show the data for Environmental Zones 1, 2, 3 and 5. There were not enough samples from Environmental Zones 4 and 6 to carry out proper statistical analysis. There was not much overall difference in PCB concentrations between the four Environmental Zones. Environmental Zone three has much higher levels of the lower-chlorinated congeners (8, 18, 28, 32 and 52) than the other three zones (Table 9.12). Class one also has relatively higher levels of lower chlorinated PCBs but not to the extent of Environmental Zone three. For PAHs the four Environmental Zones examined here can be divided into two according to concentration and patterns of individual PAHs. The first pair is Environmental Zones three and five, with similar total PAH concentrations, and patterns of individual compounds. The second pair is Environmental Zones one and two, with higher total concentrations than the other pair and the higher molecular weight PAHs were more dominant.

Table 9.12. Comparison of Median PCB concentrations (ng/g dry weight) between samples categorised by Environmental Zone

EZ PCB	1 N = 13	2 N = 61	3 N = 24	5 N = 12	P	Significant
Total PCB	2.750	1.910	2.280	1.125	0.210	No
PCB 8	0.073	0.053	0.259	0.021	0.362	No
PCB 18	0.068	0.034	0.308	0.018	0.449	No
PCB 28	0.110	0.041	0.293	0.007	0.381	No
PCB 31	0.088	0.035	0.253	0.004	0.327	No
PCB 52	0.074	0.039	0.102	0.020	0.211	No
PCB 77	0.006	0.011	0.008	0.000	0.476	No
PCB 101	0.057	0.080	0.058	0.051	0.687	No
PCB 105	0.031	0.044	0.026	0.024	0.327	No
PCB 114	0.000	0.000	0.000	0.000	0.725	No
PCB 118	0.064	0.093	0.056	0.064	0.231	No
PCB 128	0.021	0.040	0.038	0.024	0.163	No
PCB 138	0.138	0.168	0.154	0.104	0.229	No
PCB 141	0.025	0.036	0.024	0.021	0.182	No
PCB 149	0.060	0.098	0.089	0.053	0.130	No
PCB 153	0.154	0.227	0.176	0.132	0.129	No
PCB 156	0.009	0.017	0.015	0.011	0.238	No
PCB 157	0.001	0.005	0.006	0.002	0.313	No
PCB 163	0.042	0.059	0.049	0.035	0.366	No
PCB 170	0.031	0.061	0.049	0.026	0.002	Yes
PCB 171	0.008	0.016	0.013	0.004	0.011	Yes
PCB 180	0.089	0.148	0.113	0.030	0.003	Yes
PCB 183	0.019	0.033	0.029	0.015	0.017	Yes
PCB 187	0.070	0.093	0.088	0.049	0.049	Yes
PCB 189	0.000	0.002	0.004	0.000	0.020	Yes
PCB 194	0.021	0.035	0.023	0.002	0.045	Yes
PCB 199	0.003	0.006	0.005	0.001	0.043	Yes
PCB 201	0.041	0.056	0.050	0.000	0.000	Yes
PCB 206	0.020	0.024	0.023	0.015	0.118	No
PCB 209	0.029	0.039	0.037	0.024	0.111	No

Table 9.13. Comparison of Median PAH concentrations (ng/g dry weight) between samples categorised by Environmental Zone

EZ PAH	1 N = 13	2 N = 59	3 N = 24	5 N = 12	P	Significant
Total PAH	787.440	586.020	291.560	214.550	0.020	Yes
Naphthalene	24.287	8.840	16.576	15.360	0.418	No
Acenaphthylene	1.853	0.685	0.944	0.372	0.453	No
Acenaphthene	7.863	2.852	5.941	6.878	0.388	No
Fluorene	8.894	3.371	4.606	3.948	0.433	No
Fluoranthene	111.138	31.487	9.288	33.797	0.084	No
Pyrene	52.656	34.318	29.140	26.242	0.432	No
Benzo[a]anthracene	85.256	44.006	12.326	10.490	0.003	Yes
Chrysene	110.526	75.876	24.189	22.592	0.005	Yes
Benzo[b]fluoranthene	79.460	58.314	28.736	23.855	0.042	Yes
Benzo[k]fluoranthene	17.209	18.885	13.693	7.568	0.010	Yes
Benzo[a]pyrene	109.923	52.165	18.844	13.450	0.016	Yes
Indeno[1,2,3-cd]pyrene	80.097	46.585	20.216	19.916	0.032	Yes
Dibenz[a,h]anthracene	15.492	9.595	3.449	1.534	0.024	Yes
Benzo[g,h,i]perylene	83.996	49.837	20.059	19.833	0.033	Yes

Table 9.14. Comparison of Median OCP concentrations (ng/g dry weight) between samples categorised by Environmental Zone

EZ	1 N = 13	2 N = 59	3 N = 24	5 N = 12	P	Significant
Total OCP	0.000	0.000	0.000	0.000	0.462	No
p,p-DDE	0.338	0.249	0.550	0.265	0.397	No
p,p-DDT	0.000	0.000	0.000	0.000	0.552	No

### 9.3.3.3. Broad Habitats

Tables 9.15 to 9.17 show the data for Fen, marsh and swamp, Improved grassland and Neutral grassland. The congener concentrations in all habitats were similar. Improved grassland has slightly lower concentrations than the other two. The Fen, Marsh and swamp class differs from the other two classes in that the median concentrations were zero for congeners 18, 28 and 31. This may be a reflection of different congener patterns as congeners 8 and 77 were also relatively low.

The concentrations of Total and individual PAHs were higher in Neutral grassland than the other two classes. In addition Fluoranthene and Pyrene were more dominant in neutral grassland. Improved grassland has lower relative concentrations of the more volatile PAHs than the Fen, Marsh and swampland class. Neutral grassland has the highest concentrations of OCPs and Fen, marsh and swamp has the lowest concentrations of OCPs.

Table 9.15. Comparison of Median PCB concentrations (ng/g dry weight) between samples categorised by Broad Habitat

Broad Habitat	Fen, marsh and swamp N = 7	Improved grassland N = 87	Neutral grassland N = 10	P-Value	Significant
PCB					
Total PCB	5.750	1.810	2.455	0.169	No
PCB 8	0.042	0.064	0.124	0.881	No
PCB 18	0.000	0.060	0.145	0.614	No
PCB 28	0.000	0.066	0.145	0.660	No
PCB 31	0.000	0.054	0.130	0.453	No
PCB 52	0.053	0.050	0.090	0.587	No
PCB 77	0.000	0.009	0.015	0.623	No
PCB 101	0.140	0.068	0.121	0.010	Yes
PCB 105	0.054	0.031	0.073	0.024	Yes
PCB 114	0.000	0.000	0.000	0.962	No
PCB 118	0.142	0.069	0.144	0.026	Yes
PCB 128	0.054	0.029	0.060	0.012	Yes
PCB 138	0.200	0.140	0.247	0.017	Yes
PCB 141	0.039	0.028	0.047	0.108	No
PCB 149	0.127	0.078	0.122	0.201	No
PCB 153	0.246	0.171	0.283	0.026	Yes
PCB 156	0.020	0.012	0.023	0.020	Yes
PCB 157	0.010	0.004	0.005	0.128	No
PCB 163	0.064	0.051	0.080	0.067	No
PCB 170	0.057	0.046	0.078	0.076	No
PCB 171	0.015	0.012	0.020	0.204	No
PCB 180	0.114	0.108	0.185	0.345	No
PCB 183	0.029	0.024	0.042	0.208	No
PCB 187	0.132	0.073	0.116	0.103	No
PCB 189	0.003	0.002	0.004	0.735	No
PCB 194	0.031	0.027	0.025	0.958	No
PCB 199	0.007	0.004	0.007	0.119	No
PCB 201	0.081	0.044	0.069	0.248	No
PCB 206	0.041	0.016	0.037	0.009	Yes
PCB 209	0.040	0.028	0.058	0.014	Yes

Table 9.16. Comparison of Median PAH concentrations (ng/g dry weight) between samples categorised by Broad Habitat

Broad Habitat	Fen, marsh and swamp N = 7	Improved grassland N = 87	Neutral grassland N = 10	P-Value	Significant
PAH					
Total PAH	379.980	329.760	984.180	0.224	No
Naphthalene	29.524	9.254	21.836	0.067	No
Acenaphthylene	0.000	0.640	1.570	0.276	No
Acenaphthene	17.043	2.947	8.756	0.056	No
Fluorene	10.742	2.975	8.319	0.036	Yes
Fluoranthene	33.317	33.937	153.901	0.329	No
Pyrene	42.182	32.172	123.788	0.182	No
Benzo[a]anthracene	24.444	24.681	78.076	0.789	No
Chrysene	43.483	39.323	112.926	0.725	No
Benzo[b]fluoranthene	94.799	37.413	78.126	0.261	No
Benzo[k]fluoranthene	18.675	14.465	17.482	0.608	No
Benzo[a]pyrene	31.400	32.502	89.111	0.539	No
Ideno[1,2,3-cd]pyrene	39.246	33.282	73.035	0.350	No
Dibenz[a,h]anthracene	7.146	6.411	9.353	0.561	No
Benzo[g,h,i]perylene	35.180	29.473	69.887	0.347	No

Table 9.17. Comparison of Median OCP concentrations between samples categorised by Broad Habitat

Broad Habitat	Fen, marsh and swamp N=7	Improved grassland N=85	Neutral grassland N = 10	P-Value	Significant
Total OCP	0.000	0.000	0.275	0.870	No
p,p-DDE	0.865	0.293	0.361	0.595	No
p,p-DDT	0.000	0.000	0.276	0.860	No

#### 9.3.3.4. Aggregate Vegetation Class

Tables 9.18 to 9.20 show comparisons between fertile grassland, infertile grassland and moorland grass mosaics. The P-values for Total PCB and for most of the congeners show significant differences between the three classes. The median and mean values for Total PCB and individual congeners in fertile and infertile grassland were very close. Total PCB and most congener values were slightly higher in infertile grassland than in fertile grassland. The mean and median concentrations in moorland grass mosaics were much higher than in the other two vegetation classes and this was the most probable source of the significant P-values.

The PAH data shows a similar pattern to the PCB data. Median concentrations were approximately twice as high in infertile grassland as in fertile, but the difference between these classes and moorland grass mosaics was about the same as it was for PCBs. All the P-values except two were significant.

For the OCP data median concentrations for Total OCP and for p,p-DDT were zero for all classes. Median concentrations for p,p-DDE were similar for fertile and infertile grasslands but higher for moorland grass mosaics. The P-value for the p,p-DDE data

was significant and this was probably due to the higher concentrations in moorland grass mosaics.

### 9.3.3.5. Major Soil Groups

Tables 9.21 to 9.22 show mean and median values for POPs in the two major soil groups analysed. Median values for individual PCBs and Total PCB were similar in the two soils. Kruskal-Wallis tests on the median values for Total PCB show no significant difference. For individual PCB congeners there were only a few that show significant differences. There were highly significant differences between total PAH and for individual PAH in brown and gley soils. The median concentrations in gleys were between two to five times higher than in brown soils, with the exception of Acenaphthylene, which was ten times higher. There were no significant differences in OCP concentrations between the two soils.

Table 9.18. Comparison of PCB concentrations (ng/g dry weight) between samples categorised by Aggregated Vegetation Class

AVC PCB	Fertile grassland N = 46		Infertile grassland N = 56		Moorland grass mosaics, N = 15		P-value (Median)	Significant
	Mean	Median	Mean	Median	Mean	Median		
Total PCB	2.799	1.780	3.444	2.080	10.983	9.960	0.000	Yes
PCB 8	0.150	0.045	0.184	0.076	0.754	0.351	0.130	No
PCB 18	0.186	0.037	0.257	0.062	1.129	0.525	0.331	No
PCB 28	0.206	0.038	0.281	0.070	1.357	0.512	0.261	No
PCB 31	0.182	0.031	0.249	0.060	1.176	0.414	0.391	No
PCB 52	0.082	0.057	0.107	0.046	0.446	0.233	0.009	Yes
PCB 77	0.013	0.006	0.018	0.011	0.076	0.039	0.023	Yes
PCB 101	0.090	0.073	0.115	0.068	0.328	0.285	0.000	Yes
PCB 105	0.056	0.037	0.061	0.031	0.315	0.172	0.000	Yes
PCB 114	0.001	0.000	0.005	0.001	0.032	0.013	0.004	Yes
PCB 118	0.110	0.074	0.126	0.063	0.592	0.382	0.000	Yes
PCB 128	0.050	0.031	0.053	0.035	0.267	0.104	0.000	Yes
PCB 138	0.211	0.132	0.240	0.153	0.858	0.421	0.000	Yes
PCB 141	0.057	0.032	0.066	0.030	0.142	0.092	0.002	Yes
PCB 149	0.137	0.074	0.191	0.085	0.401	0.248	0.002	Yes
PCB 153	0.297	0.177	0.328	0.171	0.795	0.455	0.000	Yes
PCB 156	0.019	0.011	0.025	0.016	0.133	0.048	0.000	Yes
PCB 157	0.012	0.002	0.008	0.006	0.061	0.026	0.000	Yes
PCB 163	0.081	0.044	0.089	0.055	0.270	0.188	0.002	Yes
PCB 170	0.102	0.046	0.122	0.050	0.217	0.065	0.108	No
PCB 171	0.025	0.010	0.033	0.014	0.072	0.036	0.015	Yes
PCB 180	0.261	0.110	0.338	0.110	0.337	0.140	0.545	No
PCB 183	0.060	0.026	0.073	0.026	0.107	0.069	0.019	Yes
PCB 187	0.166	0.072	0.188	0.074	0.306	0.233	0.004	Yes
PCB 189	0.004	0.000	0.007	0.003	0.024	0.005	0.014	Yes
PCB 194	0.075	0.024	0.093	0.030	0.069	0.025	0.772	No
PCB 199	0.013	0.004	0.015	0.006	0.030	0.020	0.019	Yes
PCB 201	0.117	0.042	0.117	0.047	0.169	0.127	0.040	Yes
PCB 206	0.037	0.016	0.041	0.023	0.121	0.076	0.001	Yes
PCB 209	0.037	0.026	0.048	0.034	0.235	0.147	0.000	Yes

Table 9.19. Comparison of PAH concentrations (ng/g dry weight) between samples categorised by Aggregated Vegetation Class

AVC PAH	Fertile grassland N = 47		Infertile grassland N = 55		Moorland grass mosaics, N=15		P-value (Median)	Significant
	Mean	Median	Mean	Median	Mean	Median		
Total PAH	1014.530	653.310	6102.980	248.430	1454.830	747.210	0.001	Yes
Naphthalene	27.161	11.803	38.891	8.883	41.077	37.848	0.028	Yes
Acenaphthylene	3.602	1.245	4.760	0.745	3.705	0.000	0.617	No
Acenaphthene	17.526	3.857	8.971	2.729	27.192	13.131	0.007	Yes
Fluorene	13.205	4.505	11.457	3.258	18.856	10.567	0.026	Yes
Fluoranthene	117.570	57.208	50.955	20.537	188.771	50.243	0.031	Yes
Pyrene	89.887	52.656	50.687	26.242	150.842	44.177	0.138	No
Benzo[a]anthracene	103.188	51.742	51.092	12.595	96.138	52.892	0.001	Yes
Chrysene	130.955	81.739	68.616	27.717	173.724	86.895	0.000	Yes
Benzo[b]fluoranthene	117.543	57.682	76.709	29.070	198.913	105.388	0.003	Yes
Benzo[k]fluoranthene	40.345	17.036	29.039	10.207	99.531	34.225	0.005	Yes
Benzo[a]pyrene	113.598	57.805	56.247	17.319	102.506	65.117	0.002	Yes
Ideno[1,2,3-cd]pyrene	96.487	48.376	49.539	18.683	163.660	103.783	0.001	Yes
Dibenz[a,h]anthracene	18.003	11.101	9.626	3.555	34.166	18.489	0.007	Yes
Benzo[g,h,i]perylene	91.786	51.120	45.877	19.659	148.699	90.693	0.001	Yes

Table 9.20. Comparison of OCP concentrations (ng/g dry weight) between samples categorised by Aggregated Vegetation Class

AVC OCP	Fertile grassland N = 46		Infertile grassland N = 56		Moorland grass mosaics N = 15		P-value (Median)	Significant
	Mean	Median	Mean	Median	Mean	Median		
Total OCP	2.199	0.000	3.271	0.000	8.927	0.000	0.517	No
p,p-DDE	1.882	0.307	1.530	0.305	3.450	1.077	0.014	Yes
p,p-DDT	2.140	0.000	3.133	0.000	8.703	0.000	0.316	No

Table 9.21. Comparison of PCB concentrations (ng/g dry weight) for samples categorised by soil type

Major Soil Group	Brown soils N = 70		Gley N = 47		P-value (Median)	Significant
	Mean	Median	Mean	Median		
PCB						
Total PCB	3.925	2.100	4.458	1.850	0.899	No
PCB 8	0.244	0.079	0.243	0.053	0.205	No
PCB 18	0.371	0.068	0.296	0.022	0.044	Yes
PCB 28	0.434	0.081	0.323	0.041	0.066	No
PCB 31	0.376	0.068	0.290	0.020	0.018	Yes
PCB 52	0.145	0.045	0.134	0.061	0.861	No
PCB 77	0.025	0.011	0.021	0.000	0.046	Yes
PCB 101	0.109	0.071	0.168	0.084	0.188	No
PCB 105	0.060	0.032	0.138	0.051	0.043	Yes
PCB 114	0.005	0.002	0.010	0.000	0.001	Yes
PCB 118	0.120	0.068	0.268	0.101	0.037	Yes
PCB 128	0.053	0.036	0.118	0.040	0.338	No
PCB 138	0.232	0.156	0.421	0.160	0.461	No
PCB 141	0.065	0.034	0.083	0.031	0.764	No
PCB 149	0.184	0.097	0.215	0.078	0.414	No
PCB 153	0.320	0.211	0.460	0.194	0.387	No
PCB 156	0.023	0.015	0.056	0.015	0.523	No
PCB 157	0.008	0.005	0.029	0.004	0.926	No
PCB 163	0.088	0.057	0.139	0.052	0.675	No
PCB 170	0.118	0.051	0.138	0.047	0.775	No
PCB 171	0.031	0.015	0.041	0.013	0.274	No
PCB 180	0.333	0.116	0.270	0.110	0.857	No
PCB 183	0.073	0.029	0.072	0.028	0.539	No
PCB 187	0.198	0.088	0.190	0.075	0.336	No
PCB 189	0.008	0.003	0.009	0.000	0.009	Yes
PCB 194	0.098	0.033	0.060	0.019	0.013	Yes
PCB 199	0.016	0.006	0.016	0.005	0.949	No
PCB 201	0.131	0.051	0.113	0.044	0.522	No
PCB 206	0.044	0.023	0.060	0.020	0.574	No
PCB 209	0.042	0.033	0.106	0.040	0.055	No

Table 9.22. Comparison of PAH concentrations (ng/g dry weight) for samples categorised by soil type

Major Soil Group	Brown soils N = 70		Gley N = 47		P-value (Median)	Significant
	Mean	Median	Mean	Median		
PAH						
Total PAH	534.110	241.180	7905.270	787.635	0.000	Yes
Naphthalene	25.464	6.558	47.856	33.464	0.000	Yes
Acenaphthylene	1.979	0.222	7.408	2.251	0.001	Yes
Acenaphthene	7.239	2.151	25.920	11.090	0.000	Yes
Fluorene	6.662	2.413	22.708	8.929	0.000	Yes
Fluoranthene	61.003	21.861	146.589	67.765	0.007	Yes
Pyrene	52.097	26.820	119.751	80.618	0.033	Yes
Benzo[a]anthracene	44.313	13.238	127.661	64.356	0.000	Yes
Chrysene	65.204	27.017	169.583	95.363	0.000	Yes
Benzo[b]fluoranthene	69.327	26.808	167.539	78.126	0.000	Yes
Benzo[k]fluoranthene	30.023	8.656	61.377	20.299	0.000	Yes
Benzo[a]pyrene	53.739	17.324	132.096	81.338	0.000	Yes
Ideno[1,2,3-cd]pyrene	51.471	18.002	129.300	71.799	0.000	Yes
Dibenz[a,h]anthracene	9.433	3.255	25.947	12.910	0.000	Yes
Benzo[g,h,i]perylene	47.267	19.419	121.963	75.726	0.000	Yes

Table 9.23. Comparison of OCP concentrations (ng/g dry weight) for samples categorised by soil type

Major Soil Group	Brown soils; N = 70		Gley; N = 47		P-value (Median)	Significant
OP	Mean	Median	Mean	Median		
Total OCP	3.780	0.000	3.235	0.000	0.097	No
p,p-DDE	2.268	0.382	1.388	0.338	0.714	No
p,p-DDT	3.644	0.000	3.177	0.000	0.098	No

### 9.3.4. Preliminary interrogation of data

Preliminary data analyses were carried out to examine the distribution patterns of the POPs, correlations between compounds within and between batches and relationships with environmental properties, by multi-variate techniques. The aim was to identify appropriate suitable methods to describe and analyse larger datasets. These procedures might also be a useful way of initially identifying underlying processes that might be regulating the compound persistence.

This preliminary work has revealed:

- The distribution of data for many compounds was strongly non-normal. Mean values are not a proper method of expressing the results, and standard errors an inappropriate form of expressing variation for PCBs and OCP pesticides.
- Some samples contain much higher levels of organic compounds than do others. It is not clear yet why this should be the case, or whether this variation could simply reflect variability within the sampling site.
- Some samples contain high levels of more than one compound; sometimes this association may prove easy to explain as one compound is the metabolite of another or was present as an impurity when another substance was applied.
- Preliminary multivariate analysis suggests that the pattern of residues across all three classes of compounds may be understood by reference to the physico-chemical properties of the chemicals or their more fundamental chemical properties. It is too early to draw any conclusions on this however, as correlations between the data points may arise simply from the skewed nature of the data distribution - this may be biasing the outcome of the data analysis. Preliminary mapping of the data suggest that there were no consistent geographical gradients (clines) across GB as a whole. There is some evidence that different classes of compounds have different geographical distributions.

## 9.4. Conclusion

The method meets the requirements of the project and in addition has made it possible to effectively investigate the presence of unknown organic contaminants. It is likely that there were is still a number of unknown contaminants in the CS2000 soil samples that have not been identified.

So far the data has been described by summary statistics such as median, mean etc and has been divided according to a few classifications. This has revealed significant differences in POP distributions (e.g. amongst soil type) and has suggested that there may be other patterns in the data. However this kind of straightforward analysis can only lead to limited understanding of the processes that affect POP distribution and concentrations.

The proportions of compounds in a samples should be assessed to determine whether there is variation in the patterns of compounds between samples. Studies would be needed on each group of compounds separately and in combination. This would be achieved using PCA (principle component analysis) and pattern recognition algorithms. If there are patterns common to a number of samples, then this group of samples may be dealt with separately. Suspected differences in patterns should be subjected to statistical testing

The distribution of POPs is not homogenous in British soils. The data and preliminary statistical work suggests that there were differences that may be related to soil type or altitude but this should be confirmed by more detailed study and statistical analysis. The spatial distribution patterns may be related to other factors not so far investigated, such as atmospheric deposition processes, biological processes which differ qualitatively and quantitatively between different soil types, latitude and longitude, and proximity to urban areas or industrial sources. The geographical characteristics of POP distributions need to be investigated using GIS. This would provide a means of visualising obvious spatial patterns. Spatial statistical techniques could then be used look for correlations with other spatial data (e.g. positions of potential sources).

Given the results obtained from statistical studies done to date, further interpretation of particular aspects of the data is required. Most importantly we need to:

- Obtain further insights into the moorland-lowland differences that were apparent in the data.
- Explain why gley and brown-soils contain very different PAH concentrations, initially by examining gley soil sub-types more closely.
- Compare MASQ organics data with other national pollution databases.

In particular, more multi-variate analysis needs to be done to determine whether all compounds were distributed as might be predicted from their physico-chemical properties or whether environmental factors have led to a distribution determined by physical and biological processes. This study would need to involve computational chemistry and the use of investigative approaches such as those that are deployed when developing quantitative structure activity relationships (QSARs).

### **Exploring concentrations, distribution and patterns of POPs in the environment**

An attempt should be made to explain the concentrations, distribution and patterns of compounds by examining correlations with other data from the same sampling locations e.g. physical, chemical and biological soil properties and other CS2000 data such as vegetation and land use, and with available information on the size and location of previous and current sources of pollutants. The range of these data sets is very large and some selection is required depending on the questions being examined. For example we need to test:

#### **Spatial correlation of PAH data with the size and location of sources**

Spatial correlation of PAH data with the size and location of sources, such as urban areas, roads, industrial complexes (e.g. Humberside, Teeside, Merseyside) and large combustion plants (eg Didcot and other baseload power stations). The MASQ data here

could be compared to the NERC URGENT Thematic Programme datasets, and to those obtained from a range of other situations (e.g. from accidents, FMD pyres etc).

#### **Correlation of OC pesticide data with specific agricultural land uses such as cereals and sheep farming.**

Analysis would need to be by individual compound. It would be important to note presence and absence of certain compounds from certain areas as well as differences in levels. Some compounds such as DDT are no longer in use and therefore data on land use prior to the start of the countryside survey would have to be sought.

#### **Correlation of chemical concentrations with a variety of CS2000 and MASQ physical and demographic variables.**

Since these chemicals will bind to soil organic matter in some way then we need to compare levels with physical soil characteristics gathered in MASQ. A different exercise will be required in relation to land class and land use information. It is a moot point whether we will be able to clearly separate the physicochemical effects from those due to land demographics. This must be a general challenge within MASQ and CS2000. But, for policy and regulatory purposes, we must make the effort to do so.

#### **The quantitative inputs, outputs and intermediate life-cycle of POPs in the environment**

CS2000 provides a snapshot in time of POP levels in soil. Theoretically this could be related to how much is being or has been put into the environment, and how much is being lost through volatilisation or metabolism by soil organisms. However, we are some way short of obtaining this level of dynamic understanding, partly because we know too little about half-lives of POPs in soil. The best we could do is to examine this data in the context of information being obtained in the NERC Environmental Diagnostics programme; but as yet the relevant projects are not yet complete.

#### **The use of the data to explain the levels and distribution in biota**

The MASQ data for OCPs and PCBs should be compared with that for birds of prey to determine whether there are any correlations between concentrations, compound patterns and geographical distribution. This is most appropriate for PCBs where bird of prey data from recent years is dominated by a relatively few congeners. Such a study could provide data on the transfer of POPs through food chains. Findings could inform studies of human as well as wildlife food chain.

#### **Global fractionation**

The snap shot data obtained in MASQ could be compared to predictions derived from the theory that the semi-volatile compounds analysed in MASQ move round the globe as a result of their physicochemical properties. This would involve relating levels to altitudinal and latitudinal gradients. This sort of study may be constrained by other associations in the data (e.g. if there are large local sources or a strong correlation with soil organic matter).

### **Investigation of what other compounds may be present**

A search should be made for unknown compounds in the samples. This sort of study would be extremely expensive if the aim were to identify all potentially toxic compounds present in the samples. However using the methods employed at Monks Wood for the MASQ samples, it should be possible to identify many compounds for little extra cost. Even for those compounds for which no positive identification could be made, frequency of occurrence can be recorded and an estimate made of their relative concentration.

### **Studies of heterogeneity**

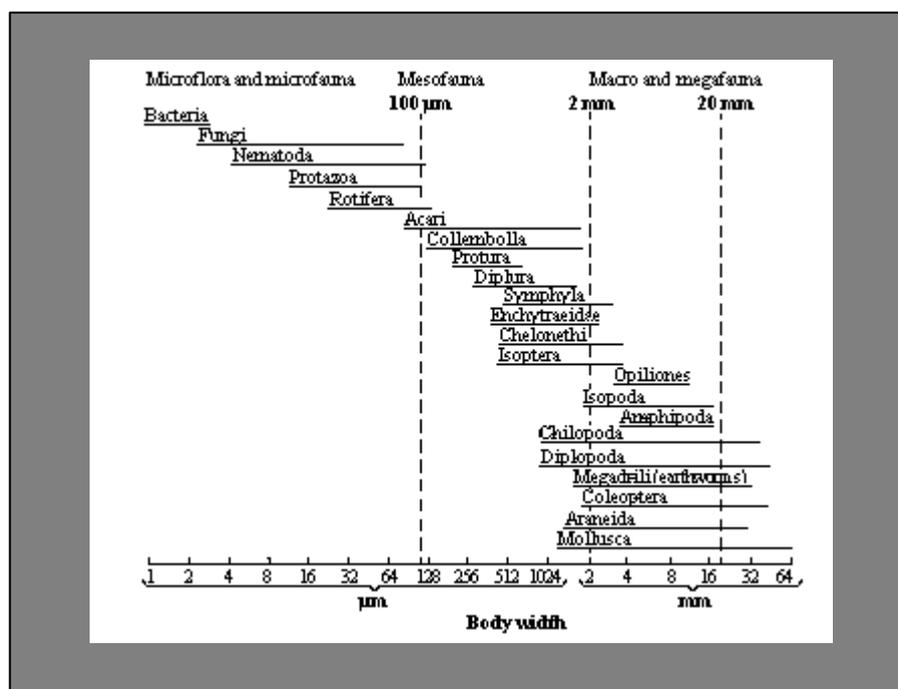
At an appropriate local scale, the POP levels found in soil should be compared in a GIS framework with a 3D model of GB land surface overlain with the latest version of the Land Cover Map. This exercise might greatly improve interpretative power as the relationship with sources and wind direction could be examined with much reduced error and uncertainty. To achieve this effectively it may be necessary to select an area of country for intensive chemical analysis, or compare and contrast heterogeneity of data in two contrasting areas of Britain (e.g. East Anglia and Cumbria).

## 10. SOIL INVERTEBRATES FROM COUNTRYSIDE SURVEY 2000

### 10.1. Introduction

Soil invertebrates are an important but relatively neglected component of the soil ecosystem (Brussaard et al, 1999). The relevance, however, of knowing what invertebrates are present where, and what they do in the soil, has renewed significance as environmental scientists attempt to predict how soils will respond to environmental change, in particular climate change, pollution and land use change, and the increasing awareness that soils are the greatest reservoir of terrestrial biodiversity that warrant some form of protection (UNEP, 1992). Soil invertebrates can be split into two main groups; those that perform all of their life cycle in the soil, and often play an integral role in mediating soil processes, and those that only carry out a part of their life cycle in the soil, and, in general have a lesser role in nutrient cycling but often a significant role in above/below ground food webs; many of the world's terrestrial insect species are soil dwellers for at least some stage of their life-cycle (Bater, 1996). Soil invertebrates of the former group include the soil micro, meso and macro-fauna and cover a large body size range (Wallwork, 1970). A simple stratification of the soil biota is often used to classify these groups, along with the microflora (Swift et al., 1979). The soil invertebrates, as illustrated in Figure 10.1, start at the microfauna with the nematodes and end with the megafauna of the larger earthworms, millipedes and slugs.

Figure 10.1. Body size-based classification of the soil biota. Adapted from Swift et al. 1979.

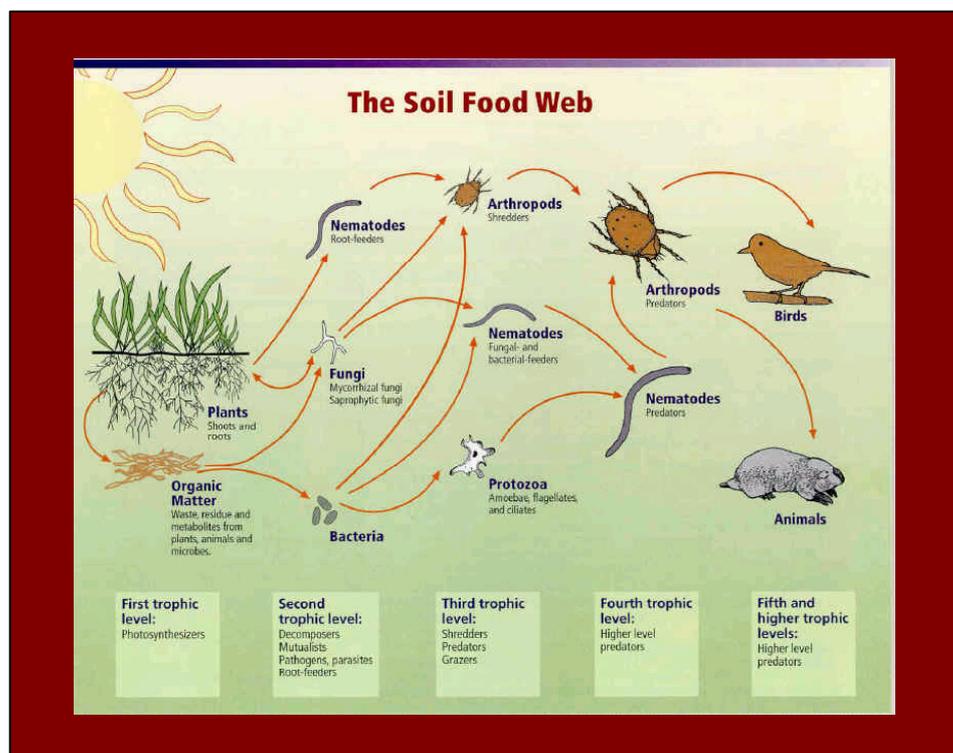


We know from detailed single site studies a typical temperate grassland soil may contain up to 24 different taxonomic groups of soil invertebrates, from 5 Phyla and

more than 20 Orders (Curry, 1994) and that the predominant group, the Acari mites may include > 40 species from 4 sub-orders (Peterson and Luxton, 1986). In a temperate beech forest there may be as many as a 1000 species in 1 m<sup>2</sup> of beech forest (Schaefer and Schauer mann, 1990) However we are still unable to accurately predict what or how many species are associated with specific habitats (Weeks et al., 1996). Future considerations of the role soil fauna in maintaining the health of the soil and their role in the development of appropriate soil protection practices require such data, as highlight by the Royal Commission on Environmental Protection (RCEP, 1996).

There is a basic understanding of the ecology of a few of the more common soil invertebrates, especially where they are important for nutrient cycling in agricultural systems (Edwards and Bohlen, 1996) or crop pest species (Phillips et al., 1998). With recent advances in our understanding of the behaviour of soil communities, it is apparent that this diverse group of soil organisms perform several fundamental functions that regulate soil processes, in particular carbon and nitrogen cycling and water transfer (Cadisch and Giller, 1997; De Ruiter et al, 1998). Invertebrates contribute to soil processes via various routes. Through direct feeding on plant material and soil, organic matter is made more readily available to soil microbes, the primary level in the soil food web (Figure 10.2). The invertebrates are, themselves, a significant biomass component at different levels of the soil food web where they contribute directly to food chains both above and below ground. Invertebrates also mediate soil processes indirectly as they stimulate microbial activity via grazing (hence altering the soil's metabolism) and alter soil chemical and properties by soil selection, ingestion and production of faeces. Earthworms are well known examples of indirect actions effecting soil physical properties (Edwards and Bohlen, 1996).

Figure 10.2. A diagrammatic representation of a soil food web, typical of a temperate region soil. Adapted from Tugel & Lewandowski (2001).



Within the UK context, The Royal Commission on Environmental Pollution Report on Soil Sustainability identified the development of indices of soil biological activity and diversity as a key research priority (RCEP, 1996). As outlined above, a major difficulty in developing such indices is the requirement for baseline data from which a set of standards can be developed. A recent review identified that existing data on soil invertebrate ecology are inadequate to assess the potential of their use as bio-indicators. The available data were either poorly structured, inconsistent in methodology/objectives or lacking in sufficient detail on the environment in which they were recorded (Weeks et al, 1996). A nationwide survey was proposed to establish a framework for comprehensive sampling of soil invertebrates that would provide the necessary site information, as well as methodology consistency, to investigate the potential of bio-indicators further. It is within this context that an assessment of soil biodiversity was deemed timely within the Countryside Surveys of Great Britain. Countryside Survey 2000 (CS2000) provided a cost-effective framework for integrating a soil biological survey with existing and subsequent soil and land use data. The programme of sampling was targeted to enable the CS2000 field surveyors to re-sample X-plots used for soil sampling in the 1978 survey, linking in site characteristics from the 3 previous surveys prior to CS2000 (1978, 1984 & 1990).

As highlighted above, the soil invertebrate community is diverse in both taxonomic and functional groups and a wide range of techniques would be required to sample and identify all invertebrate diversity within any soil. Such an All Taxa Biodiversity Inventory (Hawksworth, 1997) would be a mammoth and highly expensive task to complete, as shown by recent research programmes (e.g. NERC TIGER in tropical soils and The UK Soil Biodiversity Programme at Sourhope). This approach was not deemed appropriate for MASQ as the primary objective was to produce a large, baseline dataset across all major soil groups and habitats of Great Britain that could be used to examine the potential for using soil invertebrates in soil quality assessment. Therefore, our strategy was to capture soil invertebrates that would be abundant and relatively cost-effective to sample and identify. The category of soil invertebrates that currently, best suits these criteria is the soil meso-fauna.

We have taken a combined approach to soil biodiversity assessment by looking at the discriminating power of functional and taxonomic groups of soil biodiversity. Our efforts have been focused on groups that could be sampled, extracted and characterised with relative ease and within a limited budget. The detailed protocols developed under the project, for sampling, analyses and data-management, would be available for future monitoring or national scale sampling programmes. The datasets themselves now form a valuable resource that can be used as baseline data for future soil and other environmental monitoring programmes and a means to place specific site, region and country-scale issues within context e.g. regional, national, European and the wider international environment. Summary results from invertebrate taxa richness and occurrence are presented to illustrate the nature of the data collected and the range in values within the British countryside.

### 10.1.1 Specific Objectives

There were to :

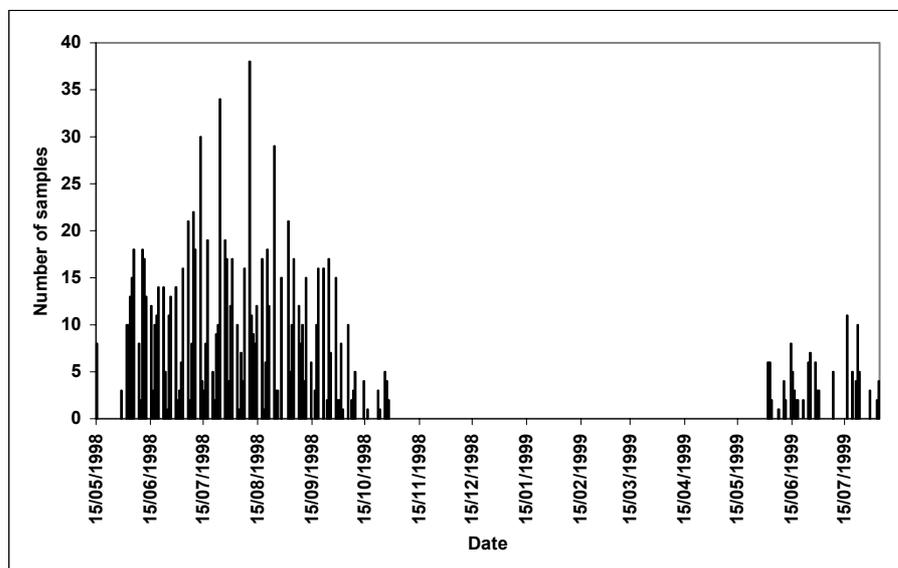
1. Develop and populate ORACLE and GIS spatially referenced datasets on soil invertebrate properties of CS2000 soil samples.
2. To provide a national overview of soil faunal (invertebrate) properties and a baseline against which specific sites can be compared.
3. Quantification of invertebrate diversity (taxa richness and diversity indices) from CS2000 soil samples. Definition of patterns with respect to geographical area, habitat, major soil type and vegetation type.

### 10.2. Methods

#### Collection of soil samples

Full details on sample collection and initial processing are presented in proceeding sections. In summary, a single soil sample (8 cm depth x 4 cm diameter core taken using an inert plastic pipe) was taken for extraction of soil invertebrates from each X-plot during the CS2000 field survey seasons in 1998 and 1999. A total of 1052 samples were obtained from CS2000. The rate of sample arrival and processing is illustrated in Figure 10.3.

Figure 10.3. Number of MASQ soil samples arriving at CEH Merlewood for fauna extraction by date



#### Extraction of soil invertebrates

Each sample was logged in a record book on arrival at CEH Merlewood and its progress through the extraction process logged at appropriate times. Soil invertebrates were extracted using a dry extraction technique using Tullgren Funnel apparatus. The soil was placed under a 25 W (or equivalent) light source for 5 days and the invertebrates are collected below in 70% Ethanol preservative in a pre-labelled 50 ml plastic tube.

Full protocols are provided in the Project Record: Posted Cores Zoology Extraction Protocol. The 50 ml tubes were sealed and stored in numerical order in plastic trays in a cool room until identification.

### **Identification**

The identification levels achieved for each major taxonomic group are given in Table 10.1. A detailed protocol was prepared for the identification and enumeration of the soil invertebrates (see Project Record: Fauna Identification Protocol). This involved a first order sort, identification, count and re-storage of each major invertebrate taxa in specific colour coded storage tubes, which facilitated the next phase of identification. Larvae were counted separately where relevant. The major taxa were identified using the AIDGAP key (Tilling, 1987). Three trained staff carried out the first order identifications with more detailed identifications obtained using specialist keys and/or knowledge i.e. by sending samples of individual taxa to expert taxonomists for key groups (where possible). The project has compiled a detailed list of invertebrate identification experts within UK. Some of these experts were contacted and may be willing to do further identifications, if required. The following experts were involved in the species identification carried out to date.

- Mr F Monson (Liverpool) = Oribatid mite species
- Mrs. P Self (CEH Merlewood) = Collembola species using a test version of AIDGAP Key to British Collembola by Dr. S Hopkin (on-going, key released in 2000).
- Dr G Legg (BRC) = Pseudoscorpion species
- Mr E Rispin (CEH Dorset) = Coleoptera (beetle) adults, Coleoptera larvae, Diplopoda (millipedes) and Chilopoda (Centipedes), Hemiptera (true bugs) and Isopoda (woodlice) species.

### **Quality control**

After identification to Taxonomic level 1, every one in ten of the first 500 samples were re-counted and identified by another member of staff. This second identification and count was compared with the original. Differences in identifications were resolved at this stage. Any mislabelling was corrected at this stage. Any changes were notes on the record sheets. The process was repeated at a reduced rate as the identifications proceeded (5 percent for the next 300 down to 2 percent for the final 252). T-test analysis indicated that there were no significant differences between the original fauna numbers and the validated data. This process, however, highlighted that a small percentage of the smallest invertebrates could be lost in transfer from the original extraction sample tube to the colour-coded tubes. A note was made to ensure that future re-assessment of identifications or counts would take this factor into account.

### **Further levels of identification**

Specialist identifiers were sent sorted samples with a sample list on hard copy and/or Excel spreadsheet. Paper records were transferred onto Excel spreadsheets by CEH staff and then passed to the data manager for further processing in ORACLE.

## Sample storage

All samples are preserved in 70% Ethanol at CEH Merlewood in long-term storage facilities. X-plots can have a series of sub-samples depending on the level of identification obtained. These are kept together in a sealed plastic bag and all sub-samples and bags are labelled accordingly. A reference collection of the Oribatid mite species collected under this project is also maintained in 70% ethanol.

Table 10.1. Levels of identification achieved, to date, for each invertebrate taxonomic group, listed by common name.

Common name	Taxonomic Level 1	Taxonomic Level 2	Taxonomic Level 3	Taxonomic Level 4
Mites	Order = Acari	sub-order = Oribatida	family/genus = Oribatid	species = Oribatid spp.
Spiders	Order = Araneae			
False scorpions	Order = Pseudoscorpions	super-family = Neobisioidae	genus = Acanthrocreagris	species = Ronconcreagris cambridgei
Harvestmen	Order = Opiliones			
Centipedes	Class = Chilopoda	Order = Geophilomorpha Order = Lithobiomorpha	family = Geophilidae family = Lithobiidae	species species
Beetle larvae Beetle adults	Order = Coleoptera	adults          larvae	family = Carabidae family = Hydrophilidae family = Ptiliidae family = Staphylinidae family = Pselaphidae family = Scarabaeidae family = Cantharidae family = Nitulidae family = Cryptophagidae family = Lathridiidae family = Chrysomelidae family = Curculionidae	species species species species species species species species species species species
Springtails	Class = Collembola Order = Arthropleona  Order = Neelipleona Order = Symphypleona	super-family = Entomobryoidea, Poduroidea  family = Neelidae family = Sminthuridae	family, sub-family, genus (on-going)  genus (on-going) subfamily, genus (on-going)	species (on-going)  species (on-going) species (on-going)
Copepods	Class = Copeopoda			
Millipedes	Class = Diplopoda	suborder/superfamily = Polydesmoidea	family = Polydesmidae family = Iulidae	species species
2 pronged bristletails	Order = Diplura			
True fly larvae True flies	Order = Diptera	larvae adults		
Slugs/snails	Class = Gastropoda			
Bugs	Order = Hemiptera			
Ants/wasps	Order = Hymenoptera			
Woodlice	Order = Isopoda Series Ligienne	superfamily = Trichoniscoidae superfamily = Pseudotracheata	family = Trichoniscidae family = Armadillidae	species species
Butterflies/moths	Order = Lepidoptera	Adults/larvae		
Nematodes	Phylum = Nematoda			
Parasitic worms	Phylum = Nematomorpha			
Worms	Class = Oligochaeta			
Pauropods	Class = Pauropoda			
Protura	Order = Protura			
Booklice	Order = Psocoptera			
symphyla	Class = Symphyla			
Thrips	Order = Thysanoptera			

## Data Entry

Identification level 1: Each identifier recorded their data on Excel spreadsheets (see Table 10.2 for an example). These sheets were then sent electronically to the data manager who checked the data for anomalies, replaced written notes with quality control codes and saved all files as “.csv” files (comma delimited) ready for storage in ORACLE data storage package (see Table 10.3).

Table 10.2. An example of an Excel invertebrate recording sheet.

SQUARE_NUM	PLOT_TYPE	REP_NUM	SOIL_TYPE	QC_CODE	ACTO	ARAN	CHTO	CHGE	SIG
6	X	1	6		65	0	1	0	AR
6	X	2	6		278	0	0	0	PS
6	X	3	6		52	0	0	0	PS
6	X	4	6		5	0	0	0	AR
6	X	5	6		34	0	0	0	AR
15	X	1	5		52	0	0	0	PS
15	X	2	5		5	0	0	0	AR
15	X	3	5		0	0	0	0	AR
15	X	4	5		51	0	0	0	PS
15	X	5	5		5	0	0	0	AR
18	X	1	5		3	0	0	0	AR
18	X	2	5		6	1	1	5	DB
18	X	3	6		1	0	3	1	DB

Table 10.3. MASQ fauna\_view: a small section of the data as it looks in ORACLE

SQUARE_NUM	PLOT_TYPE	REP_NUM	SOIL	EZ	AVC	LC	COUNTRY_CODE	QC_CODE	ACTO
6	X	1	Podzolic Soils	2	Heath and bog	6e	ENG		65
6	X	2	Podzolic Soils	2	Upland wooded	6e	ENG		278
6	X	3	Podzolic Soils	2	Moorland grass mosaics	6e	ENG		52
6	X	4	Podzolic Soils	2	Infertile grassland	6e	ENG		5
6	X	5	Podzolic Soils	2	Infertile grassland	6e	ENG		34
15	X	1	Brown Soils	2	Infertile grassland	6e	ENG		13
15	X	2	Brown Soils	2	Infertile grassland	6e	ENG		26
15	X	3	Brown Soils	2	Fertile grassland	6e	ENG		4
15	X	4	Brown Soils	2	Infertile grassland	6e	ENG		9
15	X	5	Brown Soils	2	Infertile grassland	6e	ENG		21
18	X	1	Brown Soils	2	Fertile grassland	6e	ENG		2
18	X	2	Brown Soils	2	Infertile grassland	6e	ENG		1

## Data storage

The MASQ fauna data were inserted into ORACLE and connected to CS2000 data (Broad habitat (BH), Environmental Zone (EZ), Major Soil Group (SOIL), Aggregate Vegetation Class (AVC) and 1998 Land Class (LC) by the data manager. Connected ‘views’ were used to access the data in ORACLE.

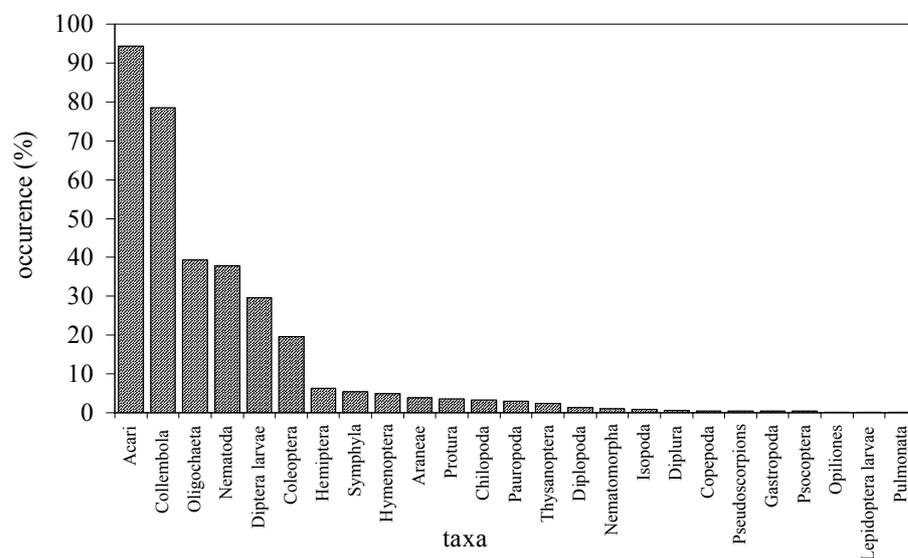
## Statistical analyses

A SAS Library was created for the MASQ fauna views. Summary statistics were produced for the whole dataset in SAS Analyst. A record of zero indicated that no taxa were recorded in that square although as soil sample had been extracted. Samples, missing, not extracted or anomalous entries, were identified by a quality control field (QC\_CODE). Summary statistics were calculated (see Project Record: MASQ\_Data\_Analysis\_Protocol) for the number of taxa (“all taxa”) that occurred and the total number of Collembola and mites. Statistics presented are; number of samples, mean, median, maximum, minimum and standard error. Summary tables were created in Excel and normal distributions were produced using Minitab statistical package. Graphs of box-plots were produced in Statistica that show median, maximum and minimum and the inter-quartile range. Diversity indices were calculated for Oribatid mite species using BIODIVPRO software (Natural History Museum free download). All summary tables produced by Environmental Zone sub-divided by Broad Habitat, CVS Aggregate Vegetation Class and major soil group are available in the project record.

## 10.3 Results

Soil invertebrates were extracted from over 98% of all soil samples (Table 10.4.) with 25 major taxa identified from all soil samples (Table 10.5; Figure 10.4). Acari were the most frequently recorded soil invertebrate group (94% of all samples received) with the Collembola (springtails) the next most frequent (78% of all samples). The remaining taxa were less common and found in less than 50% of the samples received (Figure 10.4.). Of these, the two most frequent groups were Oligochaeta and Nematoda (ca. 40% of all samples). These occurrences are a little unusual as these groups are usually captured using a different extraction technique. However, both groups are common in soil and may have been collected in higher numbers due to the extremely wet summer during 1998.

Figure 10.4. Relative occurrence of invertebrate taxa in soil samples collected from CS2000 X-plots. Each taxa shown as a percentage of the total number of soil samples extracted for invertebrates.



A significant number of Diptera and Lepidoptera adults were present in the extracted samples. This was an artefact of the extraction technique since the adult flies and moths were attracted to the lights above the soil samples during the extraction process and for there fell into the preservation tube. These groups were not included in calculation of the total taxa number presented in the summary tables and graphs.

Table 10.4. Number of soil samples (i) extracted and (ii) containing invertebrates within each Environmental Zone.

Environmental Zone	Soil samples extracted	Soil samples with invertebrates
1	224	219
2	257	253
3	116	116
4	138	131
5	184	182
6	133	132
Total	1052	1033

Table 10.5. Number of records for each invertebrate taxonomic group (Taxa) from CS2000 extracted soil samples (“All) within each Environmental Zone.

TAXA	Environmental Zone						
	1	2	3	4	5	6	All
Diptera - adults	205	233	113	129	155	113	948
Acari	208	239	114	124	178	129	992
Collembola – total	191	217	84	100	138	95	825
Oligochaeta	72	117	61	50	63	50	413
Nematoda	107	115	60	48	34	34	398
Diptera – larvae	71	78	42	40	44	37	312
Coleoptera – total	45	56	28	35	21	21	206
Coleoptera – larvae	30	41	22	26	14	14	147
Coleoptera – adults	22	19	9	12	8	8	78
Hemiptera	19	17	8	5	6	11	66
Lepidoptera – adults	10	14	1	11	20	9	65
Symphyla	25	22	3	5	1	0	56
Hymenoptera	8	15	6	3	9	11	52
Araneae	10	10	4	6	5	6	41
Protura	16	15	1	3	2	0	37
Chilopoda – total	10	12	4	2	4	3	35
Paupoda	12	14	1	5	0	0	32
Thysanoptera	7	11	2	2	3	0	25
Diplopoda	8	2	0	3	1	0	14
Nematomorpha	2	4	2	1	1	1	11
Isopoda	6	2	0	1	0	0	9
Diplura	5	1	1	0	0	0	7
Copepoda	1	3	1	0	0	0	5
Pseudoscorpions	1	2	1	1	0	0	5
Gastropoda	3	1	0	0	0	0	4
Psocoptera	1	0	1	1	1	0	4
Opiliones	0	1	0	0	1	0	2
Lepidoptera – larvae	0	0	1	0	0	0	1
Pulmonata	0	0	0	0	1	0	1
Taxa – total	219	253	116	131	182	132	1033
Number of samples extracted	224	257	116	138	184	133	1052

### Distribution of datasets

Results from Normality Tests indicate that the numbers of Taxa, Collembola and Oribatid per samples were not non-normally distributed (Figures 10.5, 10.6 and 10.7 respectively). These results indicate that appropriate data transformations would be required for parametric statistical analyses. Hence, although results are presented for mean numbers of Taxa, Collembola and Acari in all summary tables, the following section discusses median values only, where relevant.

Figure 10.5. Results from Normality test for number of taxa per sample

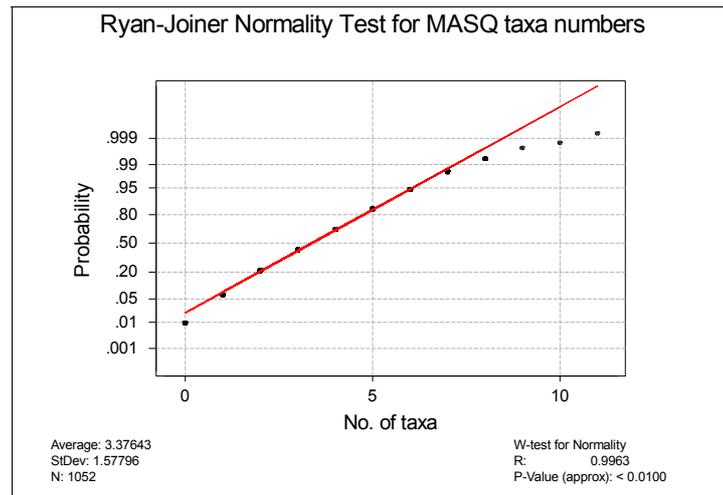


Figure 10.6. Results from Normality test for number of Collembola per sample

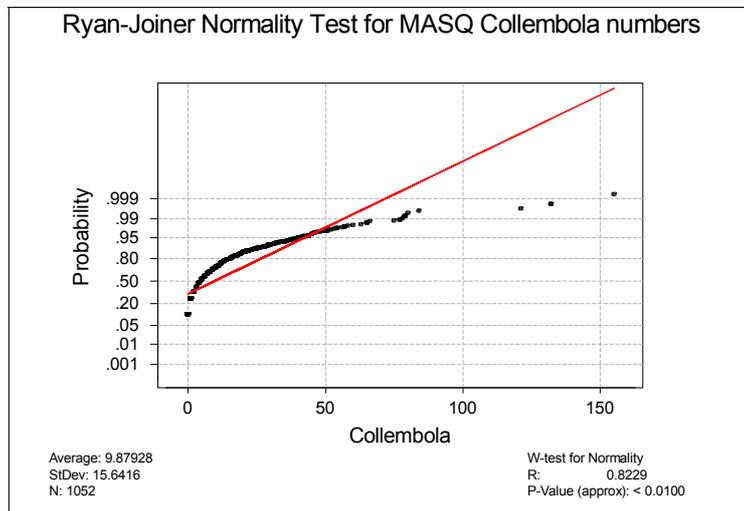
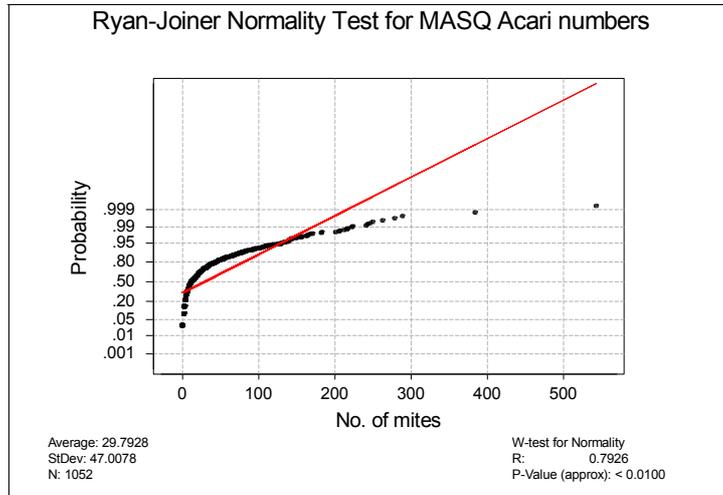


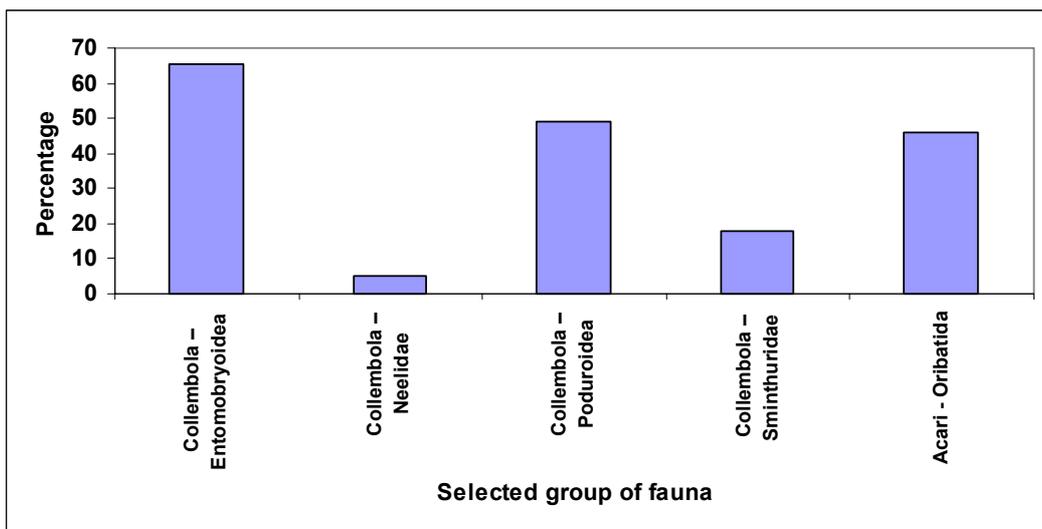
Figure 10.7. Results from Normality test for number of Acari per sample



### Collembola super-families and Oribatid mites

Results, as shown in Figure 10.8., indicate that the larger Collembola, Entomobryoidae and Poduroidea, were recorded in over 50% of all soil samples extracted, Neelidae and Sminthuridae, smaller Collembola, in less than 20% and Oribatid mites from 46% of all soil samples extracted). A total of 130 Oribatid mites species were identified from all samples (see Table 10.10). Within this group, there were 9 species and 2 genera that were new records to Great Britain. A further 6 species are as yet unidentified and may be new species to science while there may also be new genus (Monson, pers. comm.).

Figure 10.8. Relative occurrence of Collembola super-families and Oribatid mites in CS2000 soil samples; percentage of the total N of samples extracted for invertebrates.



In the following sections, summary statistics are presented for the number of Taxa, Collembola and Acari recorded for soil samples from each Environmental Zone, Broad habitat, ITE 1998 Land Class, CVS Aggregate Vegetation Class and Major Soil Group.

Unless otherwise stated, summary statistics were calculated from all samples extracted and include samples where no taxa were recorded. For individual taxa, this has resulted, in some instances, in low median and median values.

### 10.3.1 Environmental Zones

#### Number of Taxa

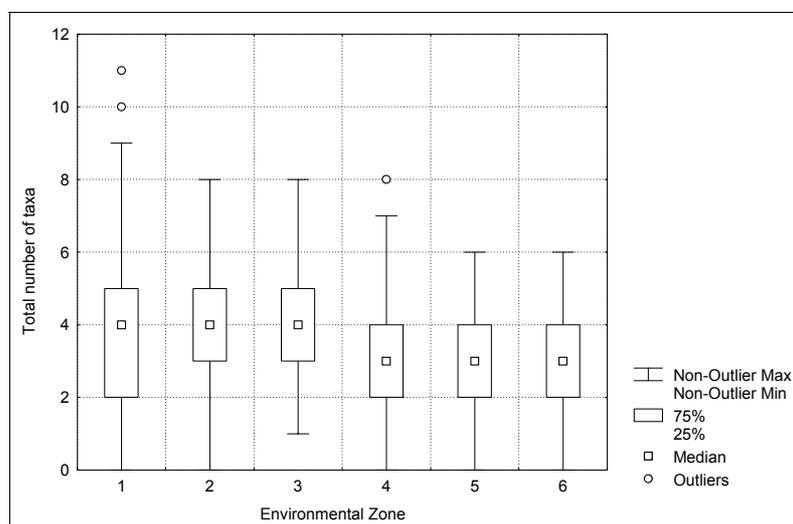
Table 10.6 presents the summary statistics for Taxa recorded from soil samples in each Environmental Zone (EZ). Details for individual taxa are presented in Table 10.5. The median number of taxa per sample varied little (3 or 4 per sample) across all Environmental Zones with the maximum number ranging from 6 to 11 per sample. It is worthy of note that, from 1052 samples, only 19 samples contained no taxa (Table 10.4). A Box-plot of number of Taxa in each sample (Figure 10.9) indicates that there are few outliers in the datasets and that the number of taxa was between 2 and 5 for 50% of all samples in each Environmental Zone.

The results show (Figure 10.9) that there was, in general, a higher number of Taxa per sample from Environmental Zones in England and Wales (median 4; maximum 11) than samples from Environmental Zones in Scotland (median 3; maximum 8). Further analyses are required to determine the significance of this result in relation to sample size. Results may be biased by the differences in sample N among the Environmental Zones.

Table 10.6. Summary statistics for Taxa numbers per soil sample by Environmental Zone.

Environmental Zone	Number of soil samples extracted	Mean number of Taxa per sample	Median number of Taxa per sample	Maximum number of Taxa per sample	Minimum number of Taxa per sample
1	224	3.7	4	11	0
2	257	3.71	4	8	0
3	116	3.66	4	8	1
4	138	3.15	3	8	0
5	184	2.78	3	6	0
6	133	2.99	3	6	0
Total	1052	3.38	3	11	0

Figure 10.9. Box-whisker plot of the number of taxa per sample by Environmental Zone



## Collembola

As illustrated in Table 10.5, Collembola were recorded in 84 to 217 soil samples from the Environmental Zones; 71 to 85% of all soil samples extracted in each Environmental Zones. Collembola were recorded at greater frequency from soil samples collected in EZ 1 and 2 than all other zones (an additional 10% of samples). The median number of Collembola ranged from 2 to 6.5, with a maximum of 155 (Table 10.7). The highest median number of Collembola per sample was recorded from EZ3. There were higher numbers of Collembola in EZ1, 2 and 3 compared with EZ4, 5 and 6, i.e. more per sample in England and Wales compared to Scotland. Figure 10.10 illustrates the wide range in Collembolan numbers per soil sample that may reflect environmental differences as well as sample numbers. The number of soil samples with no Collembola ranged from 14.7% (EZ1) to 28.6% (EZ6). Figure 10.10 shows that there were many outliers and extremes in the untransformed dataset that require closer examination. Within the Collembola (Table 10.8), Entomobryoidae were the mostly frequently recorded Collembola; 57 to 78% of all samples. This group were most frequent in EZ 1 followed by EZ2. Poduroidae were the next most frequently recorded Collembola, ranging from in 44 to 57% (most in EZ3 and with least in EZ1). Sminthuridae were recorded in 11.5 to 25.2% of all samples with most in EZ2. Neelidae were recorded in 2.2 to 7.5% of soil samples, most in EZ6.

Table 10.7. Summary statistics for number of Collembola extracted from soil samples by Environmental Zone.

Environmental Zone	Number of soil samples extracted	Mean number of Collembola per sample	Median number of Collembola per sample	Maximum number of Collembola per sample	Minimum number of Collembola per sample	Standard deviation
1	224	10.3	6	65	0	12.63
2	257	11.2	5	132	0	17.14
3	116	14.84	6.5	155	0	24.8
4	140	7.26	3	54	0	11.12
5	184	6.04	2	63	0	9.09
6	133	10.21	4	121	0	16.64
Total	1054	9.86	4	155	0	15.63

Figure 10.10. Box plot showing number of Collembola per soil sample within each Environmental Zone

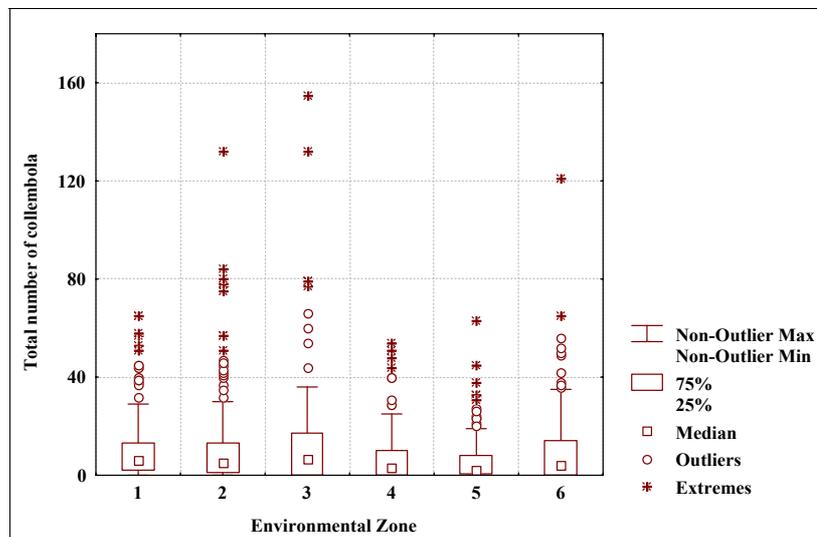


Table 10.8. Relative frequency of Collembola and Oribatid mites in soils samples from each Environmental Zone.

Relative frequency of Collembola and Oribatid taxa in soil samples (% of samples extracted)	Environmental Zone						Total
	1	2	3	4	5	6	
Collembola – Entomobryoidea	78	68	63	59	57	61	65
Collembola – Neelidae	6	7	4	2	3	8	5
Collembola – Poduroidea	44	50	57	52	49	46	49
Collembola – Sminthuridae	18	25	16	12	16	14	18
Acari – Oribatida	39	43	64	32	50	57	46
Number of samples extracted	224	257	116	138	184	133	1052

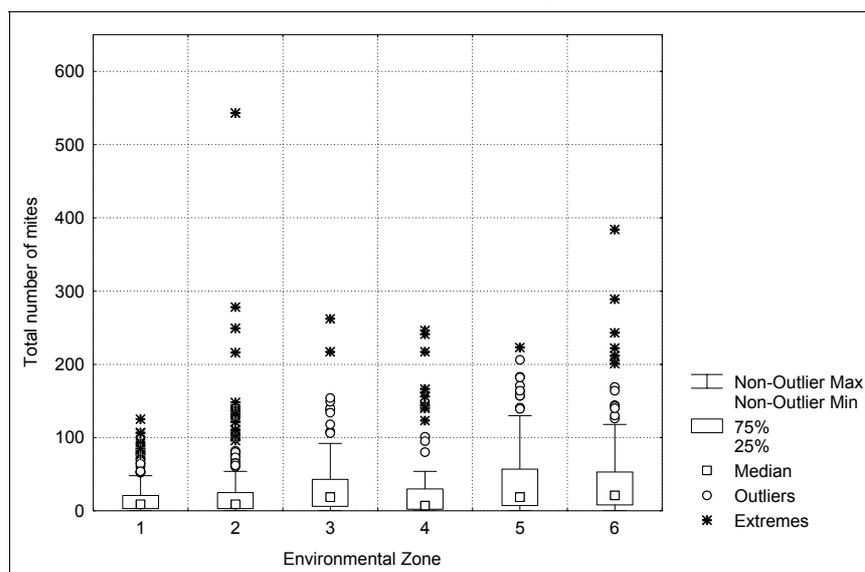
### Acari

Acari were recorded in 90 to 98% of all samples across the Environmental Zones (Table 10.5). The median number of Acari per sample ranged from 7 to 21, with a maximum of 543 in EZ2 (Table 10.9). Acari were most numerous in samples from EZ's 3, 5 and 6. Figure 10.11 illustrates that there are many outliers and extremes in the dataset. Further data analyses are required to determine what data transformations will reduce the variability for statistical analyses.

Table 10.9. Summary statistics for Acari numbers in soil samples by Environmental Zone

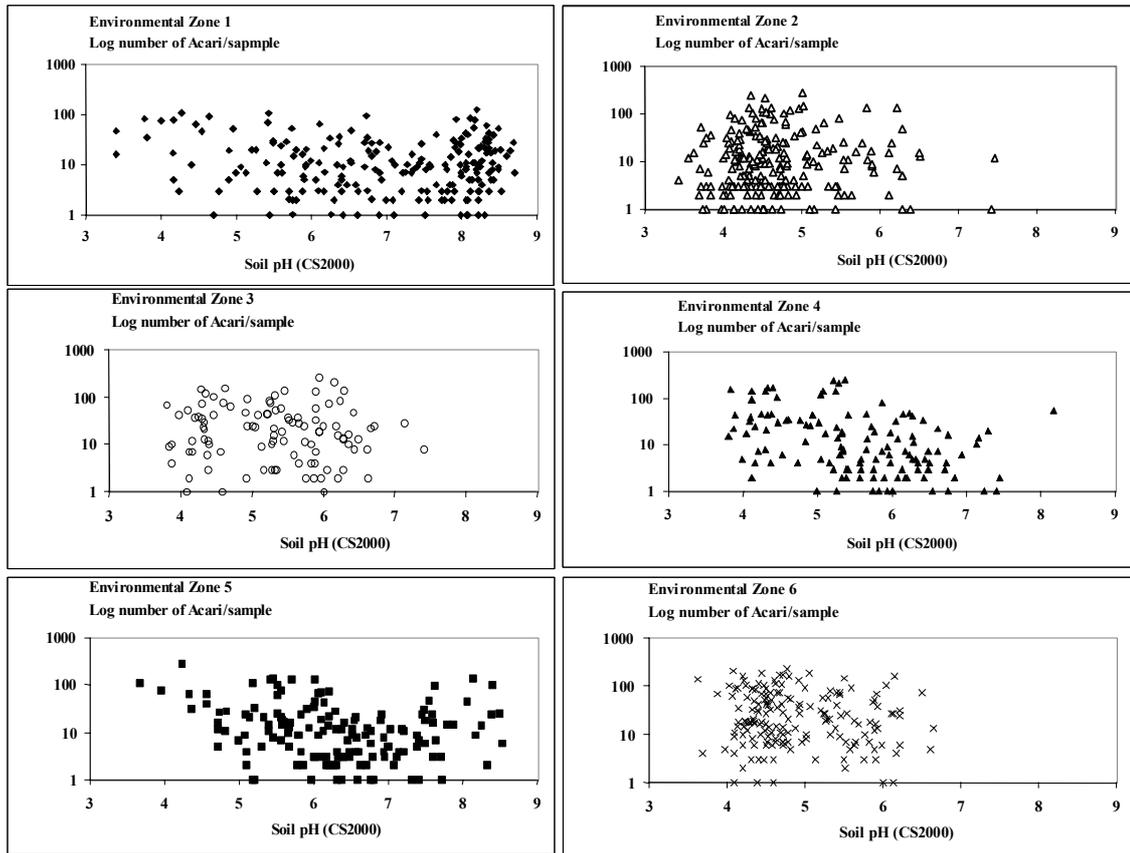
Environmental Zone	number of samples	mean	median	maximum	minimum	standard deviation
1	224	16.96	9	125	0	22.15
2	257	25.79	9	543	0	51.38
3	116	34.3	19	262	0	44.8
4	138	26.78	7	246	0	47.63
5	184	39.64	19	223	0	45.83
6	133	44.73	21	384	0	62.71
Total	1052	29.79	12	543	0	47.01

Figure 10.11. Box plot of number of Acari per sample across the Environmental Zones



The numbers of Acari per sample were plotted against soil pH from the corresponding chemistry sample within each Environmental Zone (Figure 10.12). These results indicate that there was, at this level, no clear relationship between soil pH and Acari in any Zone.

Figure 10.12. Scatterplots for each Environmental Zone showing Acari mite numbers per soil sample plotted against soil pH for soil collected from the same X-plot during CS2000.



### *Oribatid mites*

Further identification indicates that the Oribatid mites were recorded from 46% of all soil samples extracted (Table 10.8). A total of 130 species were identified from all samples (Table 10.10), which was much greater than the total number of species identified from individual Environmental Zones, where the lowest total number of species was recorded from Environmental Zones 3, 4 and 5. The median number of Oribatid mite species per sample in the Environmental Zones ranged from 0 to 17 per sample with numerous outliers and extreme values (Figure 10.13). The maximum number of species ranged from 12 to 17 per sample across the Environmental Zones with the highest number, and greatest range in number, of species per sample in the upland Environmental Zones 3, 5 and 6.

Figure 10.13. Box-plot of Oribatid mites species per soil sample within each Environmental Zone; medians, 25-75% quartiles, min-max., outliers and extremes.

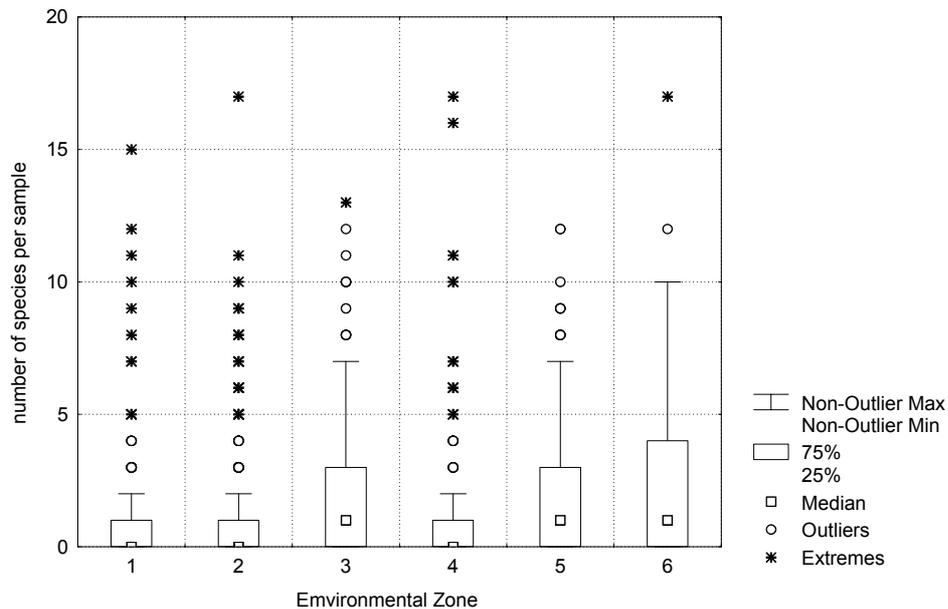


Table 10.10. Number of Oribatid mites species within each Environmental Zone, Major Soil Groups and CVS Aggregate Vegetation Class.

Environmental Zone	Spp. N.	Major Soil Group	Spp. N.	Aggregate Vegetation Class	Spp. N.
1	73	Brown soils	76	Crops and Weeds	18
2	84	Ground-water gleys	67	Tall grass and herbs	16
3	63	Surface-water gleys	45	Fertile grassland	27
4	57	Lithomorphics	73	Infertile grassland	64
5	64	Peat (organic soils)	52	Lowland wood	63
6	70	Podzolic soils	82	Upland wood	65
		Pelosols	5	Heath and bog	64
Total number of species recorded		130		Moorland grass mosaic	80

### 10.3.2. Broad Habitats

The following section presents summary statistics for Taxa, Collembola and Acari numbers by Broad Habitat. Descriptions of the Broad Habitats are presented in the Countryside Survey section. There are currently 22 Broad Habitats. There is a large range in the number of soil samples associated with each of these (the CS2000 sampling stratification is based on ITE Land Classes). Of these twenty two Broad Habitats, only nine have more than 30 soil samples (Table 10.11), seven contain less than 30 samples and the remaining four have no soil samples. Most samples were collected from improved grassland followed by arable/horticultural land. Those with no samples are not listed in the summary tables. Unless otherwise stated, this section only discusses results from Broad Habitats where more than 30 samples were extracted. Future analyses should consider the value of amalgamating Broad Habitats to increase sample numbers to a statistically valid and ecologically realistic size. It should be noted that the total number of soil samples per Broad Habitat listed in various tables in this section is higher than that actually extracted. This is due to an artefact of assigning Broad Habitat

type to each X-plot. An X-plot may be recorded against more than one Broad Habitat. However it was considered more realistic to include these double figures in the analyses since the corresponding X-plot could not be assigned to a single Broad Habitat.

### Numbers of Taxa

From the nine Broad Habitats with over 30 samples, the median number of Taxa per sample ranged from 3 to 6 with a maximum of 11 (Table 10.11). The Broad Habitats have been placed into three groups according to the number of Taxa recorded (Table 10.11). The first group has the highest number of Taxa per sample; broadleaved woodland (min. 5 & max. 11). The second group is intermediary and includes the three grassland habitats and coniferous woodland (min. 4 to max. 9). The third group has the lowest number of Taxa per sample (min. 2 to max. 7) and includes two wetland habitats, heath and arable/horticultural land. There is an overlap between arable/horticultural and neutral grassland and hence they have been assigned to groups 2 & 3.

**Table 10.11. Summary statistics for number of Taxa recorded from soil samples by Broad Habitat.**

Broad Habitat	Code	number of samples	mean	median	maximum	minimum	standard deviation	Taxa group
Broadleaved, mixed and yew woodland	1	61	4.75	5	10	1	1.71	1
Coniferous woodland	2	82	3.46	4	8	0	1.52	2
Boundary and linear features	3	1	4	4	4	4	.	
Arable and horticultural	4	217	3.15	3	7	0	1.53	2/3
Improved grassland	5	304	3.57	4	9	0	1.51	2
Neutral grassland	6	41	3.51	4	6	0	1.55	2/3
Calcareous grassland	7	6	6.5	6	11	4	2.74	
Acid grassland	8	65	3.68	4	8	1	1.44	2
Bracken	9	17	3.59	3	7	1	1.8	
Dwarf shrub heath	10	95	3.21	3	6	1	1.32	3
Fen, marsh and swamp	11	37	2.92	3	5	1	1.19	3
Bog	12	134	2.34	2	6	0	1.21	3
Inland rock	16	1	4	4	4	4	.	
Built-up areas and gardens	17	10	5.1	5	7	3	1.2	
Supralittoral rock	18	3	4	5	5	2	1.73	
Littoral sediment	21	5	2.8	3	3	2	0.45	
Total		1079	3.37	3	11	0	1.58	

### Collembola

The median number of Collembola per sample ranged from 3 to 6, with a maximum of 11, from the Broad Habitats with >30 samples, Table 10.12. The Broad Habitats have been placed into three groups according to the number of Collembola (Table 10.12). The first group, with the highest number per sample, includes broadleaved woodland and coniferous woodland. The second, intermediary, group includes the three grassland habitats, arable/horticultural land and dwarf shrub heath. The third group, with the lowest number of Collembola per sample, includes both wetland habitats (fen and bog). There is a degree of overlap between dwarf shrub heath and the two woodland habitats; hence this habitat has been assigned to groups 1 and 2.

Table 10.12. Summary statistics for number of Collembola per sample by Broad Habitat

Broad Habitat	Code	number of samples	mean	median	maximum	minimum	standard deviation	Taxa Group
Broadleaved, mixed and yew woodland	1	61	18.28	12	132	0	21.33	1
Coniferous woodland	2	83	17.84	11	121	0	21.42	1
Boundary and linear features	3	1	45	45	45	45	.	
Arable and horticultural	4	217	8.84	5	78	0	11.85	2
Improved grassland	5	305	7.8	4	79	0	11.99	2
Neutral grassland	6	41	11.68	4	77	0	16.65	2
Calcareous grassland	7	6	24.33	20	57	0	23.4	
Acid grassland	8	65	9.31	4	80	0	14.79	2
Bracken	9	17	22.24	9	132	0	35.86	
Dwarf shrub heath	10	95	13.98	4	155	0	23.64	½
Fen, marsh and swamp	11	37	5.22	3	33	0	7.11	3
Bog	12	134	3.88	1	49	0	6.97	3
Inland rock	16	1	6	6	6	6	.	
Built-up areas and gardens	17	10	15.9	12.5	41	2	14.02	
Supralittoral rock	18	3	7.33	6	12	4	4.16	
Littoral sediment	21	5	4.8	1	17	1	6.94	
Total		1081	9.99	4	155	0	15.93	

### Acari

From the nine Broad Habitats with over 30 samples, the median number of Acari per sample ranged from 5 to 36 with a maximum of 543 (Table 10.13). The Broad Habitats have been placed into two groups according to the number of Acari recorded (Table 10.13). The first group has the highest number of Acari per sample and includes all broad habitats except arable/horticultural. There is a large range in the median and maximum number of Acari recorded per sample, however this groups is distinct from Arable/horticultural land, since the later had far fewer Acari per sample than the other habitats.

Table 10.13. Summary statistics for number of Acari per sample by Broad Habitat

Broad habitat	Code	number of samples	mean	median	maximum	minimum	standard deviation	Group
Broadleaved, mixed and yew woodland	1	61	50.87	27	543	0	78.24	1
Coniferous woodland	2	82	63.85	36	289	0	67.95	1
Boundary and linear features	3	1	15	15	15	15	.	
Arable and horticultural	4	217	10.78	5	80	0	14.55	2
Improved grassland	5	304	16.37	8	246	0	27.67	1
Calcareous grassland	7	6	63.17	46	134	4	53.97	
Acid grassland	8	65	42.62	20	278	0	55.97	1
Bracken	9	17	63.41	42	217	1	69.09	
Dwarf shrub heath	10	95	42.67	22	278	0	54.2	1
Fen, marsh and swamp	11	37	35.19	12	183	1	44.76	1
Bog	12	134	38.4	21	384	0	49.94	1
Inland rock	16	1	17	17	17	17	.	
Built-up areas and gardens	17	10	38.1	17	92	3	35.57	
Supralittoral rock	18	3	65	88	96	11	46.94	
Littoral sediment	20	5	4.6	3	15	0	6.02	
Neutral grassland	21	41	38.22	16	262	0	57.27	1
Totals		1079	30.2	12	543	0	47.84	

## ITE Land Class

As discussed in the Countryside Survey section, there are now 40 ITE Land Classes, eight more than in 1978, when the 1 km x 1 km squares were identified. As a consequence, there are no samples from Land Class 17w2, 21 Land Classes contained fewer than 30 samples and only 18 Land Classes contain more than 30 samples. Further analyses could determine the validity of statistical analyses in the Land Classes with fewer than 30 samples, and the amalgamation of Land Classes to increase sample size.

### Number of Taxa

Of the 18 Land Classes (LC) with over 30 samples, 8 were from Scotland and 10 from England (Table 10.14). In these Land Classes, the median number of Taxa ranged from 2 to 5, with maxima of 4 to 11 (Table 10.14). These Land Classes have been placed into three groups according to the number of Taxa recorded (Table 10.14). The first group has the highest number of Taxa per sample (median 5 and max. 11) and only includes Land Class 2e. The second group contains 16 Land Classes with a range of values from lowest median of 3 to highest maximum of 9. The third group contains two Land Classes where the lowest numbers of Taxa were recorded (lowest median 2 and highest maximum 5). Two Land Classes overlap groups 2 and 3 (LC 21s and 29s).

Table 10.14. Summary statistics for number of Taxa per soil sample by ITE Land Class

ITE Land Class	Code	number of samples	mean	median	maximum	minimum	standard deviation	Group
1e	1	28	4.32	4	8	2	1.72	
2e	2	45	4.82	5	11	1	2.14	1
3e	3	46	3.41	3	7	0	1.63	2
4e	4	14	3.43	3.5	5	2	1.02	
5e	5	11	3.91	4	5	2	0.94	
6e	6	36	3.94	4	7	1	1.6	2
7e	7	34	3.74	4	6	1	1.36	2
7s	8	21	3.43	3	6	1	1.4	
8e	9	30	3.33	4	6	0	1.63	2
9e	10	43	3.7	4	7	0	1.66	2
10e	11	52	3.65	4	7	1	1.28	2
11e	12	48	3.1	3	9	1	1.65	2
12e	13	19	3	3	10	0	2.33	
13e	14	26	3.5	3	6	0	1.48	
13s	15	15	3.13	3	5	1	1.06	
15e	16	16	3.63	4	5	1	1.09	
16e	17	24	3.42	3	6	1	1.47	
17e	18	20	3.8	4	5	1	1.24	
17w1	19	9	4.11	4	6	2	1.17	
17w2	20	.	.	.	.	.	.	
17w3	21	18	3.22	3	7	1	1.83	
18e	22	25	3.96	4	7	1	1.57	
18s	23	4	3	2.5	6	1	2.16	
19e	24	15	3.93	4	8	1	1.62	
19s	25	5	3.4	3	5	2	1.52	
21s	26	37	2.59	2	6	1	1.36	2/3
22e	27	14	3.36	3	6	1	1.5	
22s	28	32	3.34	3	6	1	1.1	2
23e	29	15	3.27	3	7	1	1.62	
23s	30	27	3.3	3	6	1	1.35	
24s	31	37	2.86	3	6	0	1.57	2
25e	32	9	4.56	5	6	3	0.88	
25s	33	38	3.39	4	7	0	1.78	2
26s	34	30	2.3	2	5	0	1.29	3
27s	35	34	3.47	3.5	8	1	1.46	2
28s	36	36	3.19	3	6	0	1.37	2
29s	37	43	2.95	3	5	1	1.19	2/3
30s	38	35	1.77	2	4	1	0.81	3
31s	39	32	2.75	3	6	1	1.19	2
32s	40	29	3.14	3	5	0	1.16	

## Collembola

Collembola were recorded in all ITE Land Classes where samples were taken (Table 10.15). Summary results presented in Table 10.16 show that the median values for these 18 Land Classes ranged from 1 to 16 with maxima from 23 to 132 individuals per sample, with the highest number from LC 10e. In the Land Classes with more than 30 samples, the median number of Collembola per sample ranged from 1 to 9, with a maximum of 132. These Land Classes were ordered into three groups according to the number of Collembola per sample (Table 10.15). The first group, with the highest number of Collembola per sample, includes LC 10e and 22s (median 4.5 and max. >100). The second group contains 16 Land Classes with a range of values from lowest median of 3 to highest maximum of 9. The third group contains two Land Classes where the lowest numbers of Taxa were recorded (lowest median 2 and highest maximum 5). Two Land Classes overlap groups 2 and 3 (LC 21s and 29s).

Table 10.15. Summary statistics for number of Collembola numbers in soil samples by ITE Land Class.

ITE Land Class	Code	number of samples	mean	median	maximum	minimum	standard deviation	Group
1e	1	28	14.43	10	46	0	15.29	
2e	2	45	12.84	9	57	0	13.48	2
3e	3	46	10.26	8.5	65	0	11.7	2
4e	4	14	8.64	6.5	23	0	6.61	
5e	5	11	4.36	4	10	1	3.01	
6e	6	36	14.83	6	84	0	20.69	2
7e	7	34	11.44	7.5	46	0	12.05	2
7s	8	21	5.48	3	19	0	6.19	
8e	9	30	7.23	3	41	0	9.38	3
9e	10	43	12.47	6	65	0	16.65	2
10e	11	52	12.85	4.5	132	0	22.92	1
11e	12	48	7.85	4	44	0	10.83	3
12e	13	19	7.63	2	38	0	10.85	
13e	14	26	13.42	5	78	0	19	
13s	15	15	9.6	5	40	0	11.64	
15e	16	16	3.06	0.5	11	0	3.96	
16e	17	24	9.17	4	77	0	17.77	
17e	18	20	5.8	2.5	34	0	8.16	
17w1	19	9	5.89	7	9	0	3.26	
17w2	20	0	.	.	.	.	.	
17w3	21	18	8.89	0	79	0	18.79	
18e	22	25	20.64	16	77	0	20.47	
18s	23	4	3.25	3.5	6	0	3.2	
19e	24	15	11.4	5	79	0	20.07	
19s	25	5	4.4	4	7	3	1.67	
21s	26	37	10.41	4	56	0	14.18	3
22e	27	14	26.29	12.5	155	0	42.38	
22s	28	32	14.38	4.5	121	0	24.81	1
23e	29	15	22.47	11	132	0	35.45	
23s	30	27	7.63	3	36	0	10.41	
24s	31	37	8.3	3	50	0	13.46	3
25e	32	9	8.67	8	28	0	8.5	
25s	33	39	9.72	4	51	0	14	3
26s	34	31	5.19	1	40	0	10.48	3
27s	35	34	6.41	4	54	0	9.89	3
28s	36	36	8.39	5	45	0	10.81	2
29s	37	43	4.91	1	33	0	7.8	3
30s	38	35	2.29	1	23	0	4.4	3
31s	39	32	6.16	4	38	0	8.18	3
32s	40	29	9.9	6	63	0	12.65	
Total		1054	9.86	4	155	0	15.63	

## Acari

Acari were recorded in all ITE Land Classes where samples were obtained. Summary results presented in Table 10.16 show that the median values for these 18 Land Classes ranged from 2.5 to 28, with maxima from 52 to 543 individuals per sample. The lowest counts were recorded in 26s and the highest were obtained from 10e, the same LC with the highest counts of Collembola per sample. The large range in median and maximum values do not allow groupings of Land Classes by numbers of Acari per sample.

Table 10.16. Summary statistics for Acari numbers in MASQ soil samples by ITE Land Class

ITE Land Class	number of samples	mean	median	maximum	minimum	standard deviation	Group
17w2	0	.	.	.	.	.	
18s	4	61.75	69	101	8	43.54	
19s	5	10.4	7	21	6	6.23	
17w1	9	15.78	13	48	1	15.75	
25e	9	13.89	3	48	0	16.4	
5e	11	10.45	8	34	0	10.42	
4e	14	10	8.5	33	2	7.76	
22e	14	37.07	17.5	149	0	48.58	
13s	15	63	40	241	4	70.1	
19e	15	25.07	19	72	4	19.55	
23e	15	40.2	24	134	2	42.4	
15e	16	11.25	3.5	62	0	18.22	
17w3	18	20.22	9	84	0	25.6	
12e	19	9.37	2	94	0	21.34	
17e	20	22.4	9	92	1	27.72	
7s	21	41.43	17	246	2	63.91	
16e	24	15.21	3.5	73	0	22.82	
18e	25	61.08	37	262	2	68.48	
13e	26	26.31	16	216	0	42.71	
23s	27	23.33	17	76	0	22.38	
1e	28	27.43	9.5	132	0	40.04	
32s	29	42.52	19	223	0	49.06	
26s	30	7.17	2.5	80	0	15.53	
8e	30	18.97	9	137	0	32.25	
31s	32	30.78	19	183	0	37.12	
22s	32	69.06	27	212	0	70.8	
27s	34	18.59	7	162	0	32.78	
7e	34	34.56	15	139	0	42.52	
30s	35	31.77	17	206	0	44.46	
6e	36	42.92	12.5	278	0	65.89	
28s	36	44.19	27	182	0	49.14	
21s	37	38.73	19	289	0	62.66	
24s	37	45.3	26	384	0	70.04	
25s	38	27.18	7	217	0	46.45	
9e	43	18.65	10	92	0	21.47	
29s	43	48.21	28	171	0	49.19	
2e	45	30.09	20	125	0	32.63	
3e	46	14.85	8.5	85	0	17.31	
11e	48	10.77	6.5	52	0	12.04	
10e	52	23.58	7	543	0	77.16	
Total	1052	29.79	12	543	0	47.01	

### 10.3.4. CVS Aggregate Vegetation Class

The number of soil samples collected within each CVS Aggregate Vegetation Class (AVC) ranged from 29 to 207 (Table 10.17). Results are discussed for all AVC's, irrespective of sample size. Further analyses will be required to assess the influence of sample size on numbers of taxonomic groups, and individuals per taxonomic group, per sample.

#### Taxa

Results presented in Table 10.17 show that the median number of Taxa per sample ranged from 2 to 6 per sample with maxima of 6 to 11. The lowest number of Taxa was recorded in heath and bog with the highest number of Taxa recorded in infertile grassland. In general, the aggregate vegetation classes can be assigned to two groups. In the first, a higher numbers of Taxa were recorded and include the upland and lowland woodlands and the fertile and infertile grasslands. Lower Taxa numbers were recorded in the second group; crops/weeds, tall grass/herbs and moorland grass mosaics.

Table 10.17. Summary statistics for number of Taxa per soil sample by CVS Aggregate Vegetation Class

CVS AVC	Code	number of samples	mean	median	maximum	minimum	standard deviation	Group
Crops and weeds	1	176	3.25	3	7	0	1.44	2
Tall grass and herb	2	58	3.1	3	6	0	1.67	2
Fertile grassland	3	185	3.54	4	9	0	1.52	1
Infertile grassland	4	207	3.7	4	11	0	1.67	1
Lowland wooded	5	29	5.55	6	10	2	1.8	1
Upland wooded	6	66	4.08	4	8	1	1.55	1
Moorland grass mosaics	7	137	3.34	3	6	0	1.26	2
Heath and bog	8	194	2.55	2	6	0	1.23	2
Total		1052	3.38	3	11	0	1.58	

#### Collembola

Results presented in Table 10.18 show that the median counts of Collembola per sample ranged from 2 to 16 per sample with maxima of 57 to 155. In general, Collembola numbers can be used to assign the aggregate vegetation classes to two groups. The first group, the upland and lowland woodlands, is where the highest numbers of Collembola were recorded. Far lower Collembola numbers were recorded in the second group, all other aggregate vegetation classes, except heath and bog. Although the lowest number of Collembola was recorded in this AVC, it also contains the highest number of Collembola per sample, and thus is assigned to both groups.

Table 10.18. Summary statistics for numbers of Collembola in soil samples by CVS Aggregate Vegetation Class

CVS AVC	Code	number of samples	mean	median	maximum	minimum	standard deviation	Group
Crops and weeds	1	176	9.61	5	78	0	12.73	2
Tall grass and herb	2	58	9.02	6	57	0	11.55	2
Fertile grassland	3	185	8.39	4	79	0	12.2	2
Infertile grassland	4	207	8.05	3	77	0	12.53	2
Lowland wooded	5	29	25.86	16	132	0	26.25	1
Upland wooded	6	66	21.91	15	132	0	25.91	1
Moorland grass mosaics	7	137	9.4	4	79	0	14.18	2
Heath and bog	8	194	7.45	2	155	0	16.27	1/2

## Acari

Results presented in Table 10.19 show that the median counts of Acari per sample ranged from 5 to 46 per sample with maxima of 80 to 543. In general, the aggregate vegetation classes can be assigned to three groups on the basis of Acari numbers. The first, the upland and lowland woodlands, is where the highest numbers of Acari were recorded. The second is where intermediate numbers of Acari were recorded (moorland grass mosaic and heath/bog). The third group is where the lowest number of Acari were recorded (crops/weeds, tall grass/herbs, fertile grassland and infertile grassland). Infertile grassland and upland wood were intermediary between groups 2 & 3 and 1 & 2, respectively, and thus is assigned to both groups.

Table 10.19. Summary statistics for numbers of Acari per soil sample by CVS Aggregate Vegetation Class

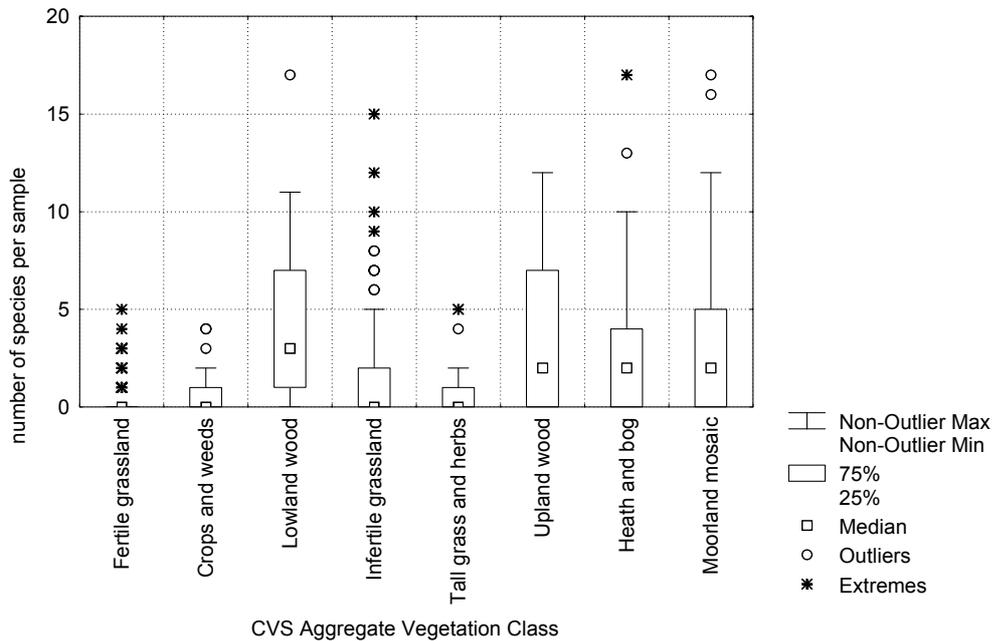
CVS AVC	Code	number of samples	mean	median	maximum	minimum	standard deviation	Group
Crops and weeds	1	176	10.41	5	80	0	13.91	3
Tall grass and herb	2	58	12.72	5.5	148	0	22.75	3
Fertile grassland	3	185	13.84	7	134	0	20.68	3
Infertile grassland	4	207	23.43	10	246	0	35.25	3/2
Lowland wooded	5	29	67.45	46	543	0	100.9	1
Upland wooded	6	66	67.74	41.5	289	1	74.29	1/2
Moorland grass mosaics	7	137	53.45	24	384	0	65.57	2
Heath and bog	8	194	39.23	25.5	206	0	41.18	2
total		1052	29.79	12	543	0	47.01	

## *Oribatid mites*

Figure 10.14 illustrates the range in numbers of Oribatid species per soil sample within individual Aggregate Vegetation Classes. The Aggregate Vegetation Classes can be assigned to two groups on the basis on number of species per sample. The first, with the greatest number of species per sample, contains upland woodland, lowland woodland, moorland grass mosaic and heath/bog; median numbers of species per sample range from 2 to 4. The second, with fewest species per sample, contains infertile and grasslands, tall grass/herbs and crops/weeds; the median number of species per sample was less than 2.

Table 10.10 details the total number of Oribatid species recorded within each Aggregate Vegetation Class and highlights that far more species were recorded from all samples than from individual samples, ranging from minimum total of 16 species to a maximum total of 80 species. However, the process of assigning Aggregate Vegetation Classes by species numbers resulted in similar groupings, with only two exceptions. Far more species were recorded from Moorland Grass Mosaics (80 spp.) than in any other aggregate vegetation class. Infertile grassland was assigned with lowland and upland woodland and heath/bog rather than tall grass/herbs, fertile grassland and crops/weeds, as assigned for species per sample. The total number of species recorded ranged from 16 to 27 in the later group and from 63 to 65 in the former group.

Figure 10.14. Number of Oribatid mite species per soil sample by CVS Aggregate Vegetation Class



### 10.3.5. Major Soil Group

Soil samples were collected from seven of the ten Major Soil Groups found in Great Britain; no samples for raw gleys, man-made and terrestrial raw soils. Soil samples in each group range from 24 to 340, with most from Brown soils (Table 10.20). There are relatively few soil samples from the Pelosol group and further analyses would be required to determine the effect of low sample size on taxonomic numbers within this Major Soil Group compared to the other Major Soil Groups.

#### Taxa

Summary statistics are presented in Table 10.20. The median number of Taxa per sample ranged from 2 to 4 with maxima 6 to 11. The highest number of Taxa per sample was recorded in lithomorphous and brown soils. The lowest number of Taxa per sample was recorded in peat (organic) soils. There were no obvious groupings amongst the Major Soil Groups on the basis of number of Taxa per sample.

Table 10.20. Summary statistics for sum of taxa in MASQ soil samples by Major Soil Group.

Major Soil Group	Code	number of samples	mean	median	maximum	minimum	standard deviation
Lithomorphous	3	97	3.77	4	11	1	1.83
Pelosols	4	24	3.25	3	9	1	1.78
Brown	5	340	3.78	4	10	0	1.6
Podzolic	6	152	3.62	4	7	1	1.39
Surface-water Gley	7	186	3.03	3	8	0	1.44
Ground-water Gley	8	117	3.16	3	8	0	1.46
Peat (organic)	10	136	2.49	2	6	0	1.26
Total		1052	3.38	3	11	0	1.58

## Collembola

Summary statistics are presented in Table 10.21. The median number of Collembola per sample ranged from 2 to 6, with maxima ranging from 44 to 155. The highest number of Collembola per sample was recorded in the surface water gleys and lowest in pelosols. As with number of Taxa per sample, there were no obvious groupings amongst the Major Soil Groups on the basis of number of Collembola since there was no obvious link between median and maximum values.

Table 10.21. Summary statistics for numbers of Collembola per soil sample by Major Soil Group

Major Soil Group	Code	number of samples	mean	median	maximum	minimum	standard deviation
Lithomorphic	3	97	9.02	4	57	0	12.29
Pelosols	4	24	7.58	4.5	44	0	10.25
Brown	5	340	10.28	6	79	0	13.19
Podzolic	6	153	15.29	5	132	0	23.92
Surface-water Gley	7	186	9.55	3	155	0	17.82
Ground-water Gley	8	118	7.39	4	58	0	10.69
Peat (organic)	10	136	6.27	2	79	0	10.57
Total		1054	9.86	4	155	0	15.63

## Acari

Summary statistics are presented in Table 10.22. The median number of Acari per sample ranged from 5 to 34.5, with maxima from 28 to 543. The highest number of Acari per sample was recorded in the Podzolic soils. The lowest number of Acari per sample was recorded in pelosols, as with Collembola. Unlike Collembola and Taxa, there is some pattern in the numbers of Acari per sample. The first grouping is podzolic soils, with far more Acari per sample than all other major soil groups. The second group contains peats, lithomorphic and brown soils, since they contained intermediate numbers of Acari per sample. The third group contains pelosols and both the gley soil with, in general, fewer Acari per sample than the other Major Soil Groups.

Table 10.22. Summary statistics for acari numbers in MASQ soil samples by Major Soil Group

Major Soil Group	Code	number of samples	mean	median	maximum	minimum	standard deviation
Lithomorphic	3	97	30.61	19	171	0	34.66
Pelosols	4	24	7.92	5	28	0	8.02
Brown	5	340	23.59	11	384	0	39.53
Podzolic	6	152	61.59	34.5	543	0	75.95
Surface-water Gley	7	186	20.73	7	149	0	32.25
Ground-water Gley	8	117	18.79	7	164	0	30.84
Peat (organic)	10	136	34.91	18	289	0	46.41
Total		1052	29.79	12	543	0	47.01

## *Oribatid mites*

Figure 10.15 illustrates the range in numbers of Oribatid species per soil sample within individual Major Soil Groups. The Major Soil Groups can be assigned to two groups on the basis on number of species per sample. The first, with the greatest number of species per sample, contains podzolic, peat and lithomorphic soils; median numbers of species

per sample range from 1 to 5. The second, with fewest species per sample, contains brown soils, ground water and surface water gleys and; the median number of species per sample was 0. However, there was a wide range in the numbers of Oribatid species per sample in all Major Soil Groups, with more than 10 species per sample not uncommon.

Figure 10.15. Number of Oribatid mite species per soil sample by Major Soil Group

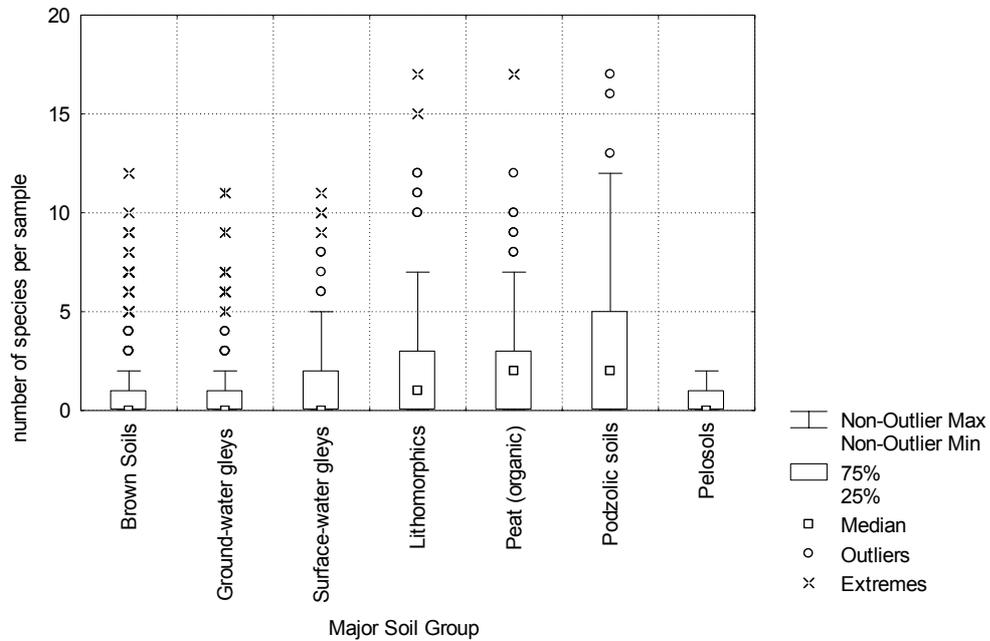
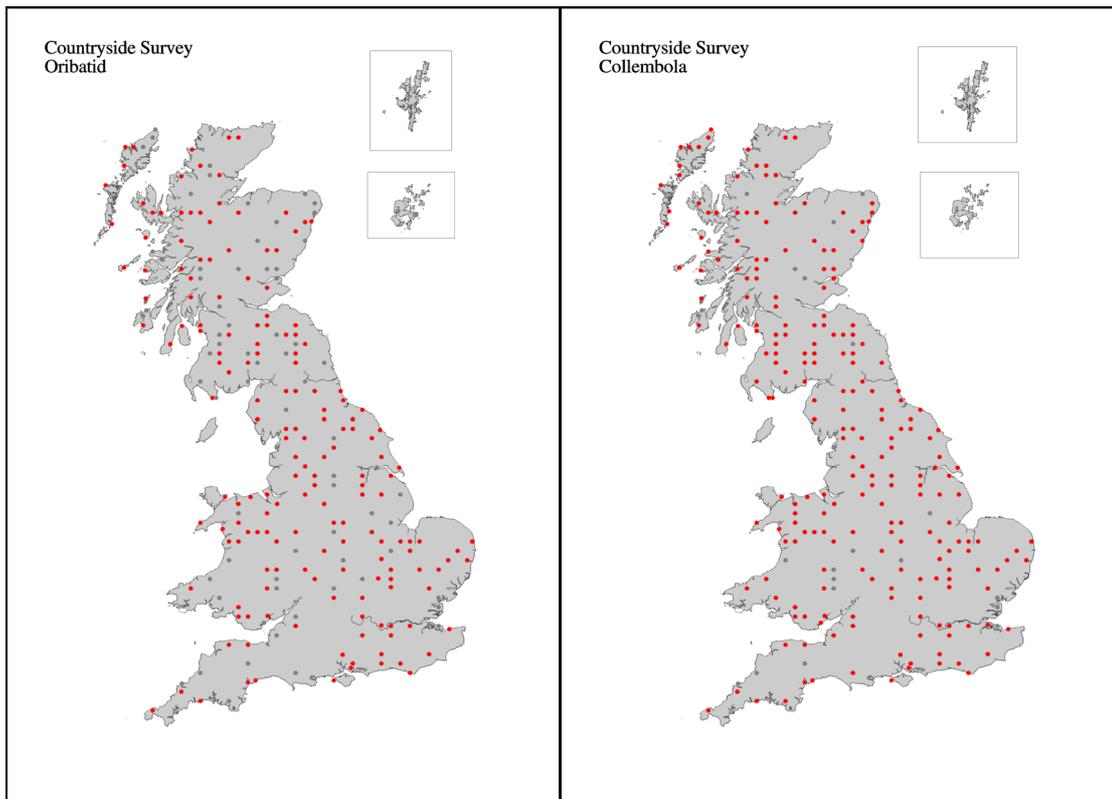


Table 10.10 details the total number of Oribatid species recorded within each Major Soil Group and highlights that far more species were recorded from all samples than from individual samples, ranging from minimum total of 5 species to a maximum total of 82 species. The process of assigning Major Soil Group by species numbers resulted in different groupings to those above, principally due to differences in total species numbers compared to species per sample in peats and ground-water gleys. The group with the highest total number of Oribatid species includes podzolic, brown and lithomorphous soils and ground-water gleys. The second group, with fewer species, includes surface-water gleys and peat soils. The last group consists of the pelosols, where low sample numbers may be responsible for the relatively low number of Oribatid species.

### 10.3.6. Mapping soil invertebrate diversity across Great Britain

Figure 10.18 illustrates, using Collembola and Oribatids as examples, the potential for mapping the distribution and occurrence of soil invertebrates, or any other MASQ soil parameter, across the British Countryside. The exact location of each 1 km square is confidential but mapping is possible by direct linkage with the CIDS database. This supports production of such maps via ARC-Info without revealing sample locations.

Figure 10.16. Map showing the locations of (a) Oribatid mite species and (b) Collembola in dark dots on the CS2000 1 km squares across Great Britain.



#### 10.4. Conclusion

During CS2000, soil invertebrates were extracted and identified from 1052 soil samples. These data, along with all associated metadata, have been fully integrated into the Countryside Survey data management system that is maintained at CEH Merlewood. From here, microbial data can be linked to any other Countryside Survey data from the same 1 km square and/or the same X-plot.

The number of major taxa recorded is typical of the soil invertebrate community, although there are no large-scale geographical studies to compare with these results. Collembola and Acari are amongst the most common soil dwelling fauna, especially in semi-natural environments, while the sampling and extraction methods used in this study would favour the collection of two groups (Collembola and Acari). Therefore it is not surprising that the results confirm their pre-dominance at the national scale. It is reassuring, that by using relatively simple sampling and extraction systems, two groups of soil invertebrates were recorded in sufficient numbers to further examine distribution patterns and relationships with other soil properties and the wide environment.

Variation and trends in invertebrate properties at the Environmental Zone, CVS aggregate vegetation class and Major Soil Group provide an opportunity to determine whether the outlier and extreme values at these higher levels correspond to other, more site-specific, environmental factors or higher-level spatial or temporal factors. For example, Oribatid mite species and microbial community responses may correspond to

soil organic matter content and/or soil pH (van Straalen and Verhoef, 1997; Bending *et al.*, 2000), land use (Brussaard *et al.*, 1990), dominant vegetation (Myers *et al.*, 2001; Grayston *et al.*, 1998; Migge *et al.*, 1998) or pollutant levels (Ellis *et al.*, 2001; van Wensem *et al.*, 1997). What are the characteristics of the soils where no Taxa were extracted?

The prevalence of both Oligochaeta and Nematoda is somewhat surprising since these groups are usually extracted from soil using different techniques. However, both groups are common in soil and may have been collected in higher numbers due to the extremely wet summer in 1998. The number of Oribatid mites is within ranges published from regional and site specific studies. Overall, the results indicate that there were more Oribatid species in upland sites, with soils with higher organic matter and least disturbance.

Appropriate statistical analyses are required to determine the significance of the trends highlighted by these results. This should include examination of the relationship between sample number and taxa occurrence.

The project has compiled a comprehensive list of potential sources of taxonomic identification of soil invertebrates; most people on this list are retired academics and further outline the decline in expertise that may limit future biodiversity assessments.

Reference collections of all invertebrates collected during CS2000 are held at CEH Merlewood. These can be used in future studies for quality control and/or further identification of remaining groups.

Identification of the Oribatid mites has produced several new records for Britain and potentially new species and a new genus to science. Collembola species identification was initiated after the release of a new British Key early in 2000; this was too late to complete all CS2000 samples. This process should be completed to provide a fully comprehensive dataset. Collembola is one group of soil invertebrates for which a great deal of information is available on distribution and occurrence at a site-specific level, often in relation to soil contamination.

## 11. SOIL MICROBIAL DIVERSITY

### 11.1. Introduction

It is acknowledged that the scale of effort and taxonomic expertise imposes serious limitations on our ability to fully assess soil biodiversity (Lawton *et al.*, 1998). Some of these limitations are being tackled by the development and application of novel genetic and functional analytical techniques for the characterization of soil communities (e.g. Grayston *et al.*, 1998; Widmer *et al.*, 2001). One simple approach to evaluating the functional diversity of a microbial community is based on the ability of members of the community to utilize different substrates. Garland and Mills (1991) first introduced the use of community-level carbon source utilization patterns for comparison of microbial communities from different habitats. These authors used the Biolog<sup>TM</sup> microtitre plate system (BIOLOG Inc., Haywood, CA. USA) that is relatively inexpensive and does not require technically skilled staff to perform the assay. Statistical analysis of the results can provide information on the number of substrates utilized by different samples (diversity of metabolic potential) and cluster samples that have similar values (high or low activity) for certain substrates or certain groups or guilds of substrates (Zak *et al.* 1994).

As yet there is little evidence to suggest that the numerous soil processes mediated by microorganisms do not persist across the range of undisturbed to heavily cultivated soils. However, more knowledge of the changes in soil functional diversity in response to environmental stress is needed. Previous studies show that populations of some soil microorganisms (e.g. *Rhizobium*) and of certain functional groups of soil microorganisms (e.g. cellulolytic microorganisms, nitrifiers and mycorrhizal fungi) change in abundance in response to changes in management practices (see Brussaard *et al.*, 1990; Dick, 1992; Hirsch *et al.*, 1993).

Such management-induced changes in microbial diversity can be seen as a consequence of changes in vegetation and substrate availability through alterations in the quality and quantity of compounds which enter the soil system through rhizodeposition and plant (litter and root) senescence (Grayston *et al.*, 1998; Myers *et al.*, 2001).

#### 11.1.1. Specific objectives

These were;

1. To develop and populate ORACLE and GIS spatially referenced datasets on soil microbial properties of CS2000 soil samples.
2. To provide a national overview of soil microbial properties and a baseline against which specific sites could be compared.
3. To quantify soil microbial diversity from CS2000 soil samples with definition of patterns with respect to geographical area, habitat, major soil type and vegetation type.

## 11.2. Methods

The following presents a summary of methods. Detailed protocols for the sampling strategy and microbiological analyses are available in the Project Record. Prior to microbial analyses, the intact soil cores from the CS2000 were stored at -20°C. Samples were processed in chronological order starting with those received in May/June 1998. The first samples were processed for microbiological analyses in March 1999 and processed in batches of 40-120 at any one time. Gloves were worn when handling samples to avoid contamination. The weight of the whole core (soil plus core) was recorded, samples were thawed overnight at 20°C and the soil from each core was then wet sieved (2 mm stainless steel mesh) to remove root material. The moisture content of each sample was determined by drying at 105°C for 2 days. Sieved samples were stored at 4°C until use (no longer than 8 weeks).

### 11.2.1. Extraction and enumeration of heterotrophic bacteria

Bacteria were separated from soil particles using a cation-exchange resin method (MacDonald, 1986). This was chosen as it represented the best compromise between time taken for extraction and efficiency of recovery. No extraction method is 100% efficient for the recovery of bacterial cells from soils. Tests based on acridine orange direct counting done at INRA, Dijon showed that the chosen method recovered between 50-70% of bacterial cells from different soil samples (Soulas and Lagacherie, unpublished data). As with all extraction media (water, phosphate buffer etc.), the efficiency of extraction differed according to soil type with less bacterial cells being extracted from highly clay soils.

Sieved soil (5 g dry wt. equivalent) was aseptically added to a sterile 30 ml centrifuge tube containing 4 g Na<sup>+</sup> amberlite, 5 g glass beads (3 mm diam.) and 20 ml aqueous solution of polyethylene glycol 6000 (2.5%) and Tween 80 (1%). The tubes were shaken on an end-over-end shaker at 4°C for 1 hr. For very moist samples, 40 ml of the Tween/PEG solution was added to 5 g dry wt. of soil in a 100 ml Duran bottle, these samples were shaken on a side-to-side shaker at 4°C for 1 hr. Samples were then decanted for 10 min followed by centrifugation at 1000 g for 2 min to remove particulates. The soil extract supernatant was then serially diluted in ¼ strength Ringer's solution. Dilutions were spread plated onto nutrient agar (NA, 1/10<sup>th</sup> strength; Difco) containing 50µgml<sup>-1</sup> cycloheximide to prevent fungal growth. Plates were incubated at 20°C and numbers of total viable bacteria in terms of colony forming units (cfu) were counted after 7 and 14 days.

### 11.2.2. Characterization of the metabolic activity and functional diversity of heterotrophic microbial communities

The functional diversity of the soil microbial community within each samples was investigated using BIOLOG microtitre plate technology (Garland and Mills, 1991; Zak *et al.*, 1994). The commercially available BIOLOG GN microplate system (BIOLOG, 1993) contains 95 separate carbon sources and a control well without a substrate, each well also contains a redox dye (tetrazolium violet). Upon respiration of a substrate the dye is reduced and the insoluble, colored compound is incorporated into the respiring cell (Bochner and Savageau, 1977). The carbon sources, dye and nutrients were present in a dried form in each well and were reconstituted upon addition of a liquid sample.

Table 11.1 shows details of the carbon sources in BIOLOG GN (Biolog Inc, Hayward, CA, USA) plates as used in this study, compounds are listed by substrate guilds.

Table 11.1. Carbon sources in BIOLOG GN microplates categorized by substrate guilds.

Substrate Guild	Number of substrates
Carbohydrates	28
Carboxylic acids	27
Amino acids	20
Amides and Amines	5
Polymers	6
Miscellaneous compounds	9

It is important to standardize the inoculum size so that the results were more representative of the metabolic potential of the microbial community rather than being due to differences in inoculum size (Kerstens et al., 1997). However, due to the number of samples being analysed in this survey it was impossible to standardize the density of microbial cells in the inoculant suspension. Instead we used a standard soil dilution to inoculate the BIOLOG GN microplates so that the results could be related directly to the population density in the sample. For each sample, bacteria were dispersed from soil particles using the cation-exchange resin method as above.

Soil suspensions were diluted in ¼ strength Ringer's solution containing 50µg/ml cycloheximide to inhibit fungal growth. The original extract was serially diluted and the lowest dilution with negligible background colour, either the 10<sup>-1</sup> or 10<sup>-2</sup> dilution, was used to inoculate BIOLOG GN plates. Each well of the BIOLOG GN plate was inoculated with 150µl of the appropriate dilution using an eight-channel multipipette. To ensure homogenous dispersion of cells, dilutions were mixed by vortexing and also by pipette before inoculation. Soil dilutions were also spread plated onto 1/10<sup>th</sup> NA plates, as above, to confirm the inoculum density. BIOLOG plates were incubated at 20°C and colour production read as optical density at 590nm (OD<sub>590</sub>) using a spectrophotometer microplate reader (Spectracount, Hewlett Packard) after incubation for 0, 4 and 7 days of incubation. Agar plates were also incubated at 20°C and colonies were counted after 4 and 7 days.

For each BIOLOG plate, the optical density values for each substrate well was adjusted for background colour by subtraction of both the value for the control (water) well and the value at time 0 for the same well. These control and time 0 adjusted values were used in all further analyses. The responses to the 95 BIOLOG GN substrates were analysed as a whole for the purpose of this report. Responses for each set of carbon sources will be analysed separately in the future so that the discriminating power of different substrate groups can be evaluated.

### 11.2.3. Density of bacterial cells used to inoculate the BIOLOG GN microplates

The mean inoculum density of the suspensions used to inoculate the BIOLOG GN plates varied from 2.88 x 10<sup>1</sup> to 4.57 x 10<sup>7</sup> and is detailed in Table 11.3. As the 10<sup>-1</sup> or 10<sup>-2</sup> soil extract dilutions were used to inoculate BIOLOG plates, there was a very good

correlation between the numbers of viable bacterial cells per g dry wt. of soil and the density of cells in the BIOLOG inoculant suspension (Figure 11.1). It is important to determine whether the density of bacteria in the inoculant suspension is related to the final activity seen in response to all 95 BIOLOG GN substrates. The correlation between inoculum density and global activity (towards all 95 BIOLOG GN substrates) was calculated and a weak correlation ( $r = 0.395$ ) was seen (Figure 11.2).

Figure 11.1. Relationship between numbers of viable bacterial cells per gram dry weight of soil with density of BIOLOG inoculant suspension

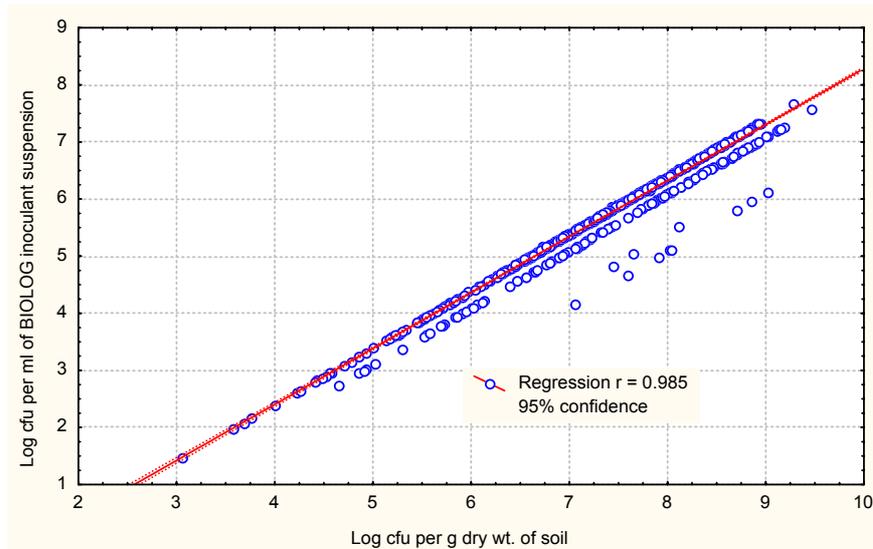
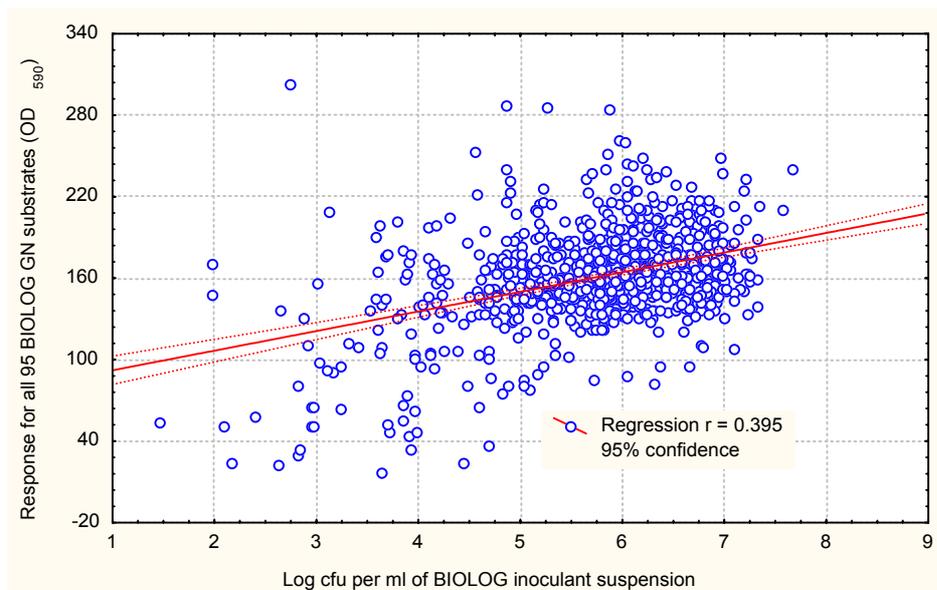


Figure 11.2. Relationship between density of BIOLOG inoculant suspension and response to all 95 BIOLOG GN substrates



The correlation was especially relevant for samples where a low inoculant density was used (below  $1 \times 10^3$  cells/ ml of inoculant suspension). In order to separate out effects of inoculum density versus the activities and functional diversities on the observed substrate utilization patterns, further analyses of the BIOLOG data will have to account for the correlation between global activity at low inoculum densities.

#### **11.2.4. Quality control**

All microbiological samples were processed in an identical manner (time and conditions) from being thawed to being refrozen for subsequent chemical analyses. Project log books were used to detail all samples processed as well as information regarding any problems or future actions to be taken with respect to a particular sample. A logbook was also kept for all BIOLOG plates that were read with notes of any problems regarding the plate-reader machine.

All equipment used for microbiological analyses was regularly calibrated and checked before the processing of each sample. Laboratory pipettes were calibrated and the plate-reader programs and filters were checked. Any microbiological samples, for which a problem was identified, e.g. plate reader error, were re-analysed in the following batch.

#### **11.2.5. Data management and analysis**

All data for both microbial counts and substrate utilization assays were entered in to Excel spreadsheets and then transferred in to ORACLE for long-term storage and linkage to other related information in the CS2000 database. Statistical analyses of all data were done using SAS and STATISTICA software packages. Data for bacterial counts presented in the text are presented in terms of CFU per g dry wt. soil or per ml of BIOLOG inoculant suspension; these numbers were not log transformed. The standard deviation and standard error of the mean are also indicated.

The current report is based on an initial analysis of the BIOLOG dataset and provides an overview of results, the data for each substrate guild, and also of each substrate, will be analysed in more detail in the next phase of this project. Several different methods have been put forward in the literature for the analysis of BIOLOG data sets. However, there is no available literature on the analysis of such a large data set as the CS2000 dataset. A simple method was chosen for the initial data analysis to provide an overview of the data. Future examination of the data will require careful assessment of analytical tools through review and discussion.

### **11.3. Results**

The tables in this section present summary statistics for the numbers of samples analysed with mean, median, minimum, maximum values for the three major microbiological parameters; log transformed numbers of colony forming units (CFU) per g dry wt. of soil, log transformed density of viable cells in the BIOLOG inoculant suspension, and the summed response of extracted soil microbial communities to all 95 BIOLOG GN substrates (in terms of OD at 590 nm). Summary results are presented by a national over-view followed by individual stratifications of the British countryside; Environmental Zone, Broad Habitat, ITE Land Class, Aggregate Vegetation Class and Major Soil Group, respectively.

### 11.3.1. National over-view

A total of 943 samples were analysed for bacterial counts and 944 for the substrate utilization assays using the BIOLOG GN system. The number of culturable heterotrophic bacteria extracted from the soil samples ranged from  $1.15 \times 10^3$  to  $2.95 \times 10^9$  cfu per g dry wt. soil, after seven days of incubation on 10% nutrient agar plates at 20°C (Table 11.2). The mean number of viable cells was  $2.51 \times 10^7$  cfu per g. dry wt. soil. Variation around this mean was very low as indicated by a standard deviation of 9 cfu per g. dry wt. soil. The distribution of numbers of viable cells per g dry wt. of soil is shown in Figure 11.3. The Shapiro-Wilk's test of normality shows that although the data fitted well with the curve for expected normality, it was not normally distributed ( $p < 0.0000$ ). The data were skewed towards those with low numbers of viable cells per g dry wt. of soil. This type of non-normal distribution is not unusual for a large and varied data set.

Initial analysis of the BIOLOG results was done using the response of extracted microbial communities to all 95 BIOLOG GN substrates. This provides an overall picture of the global metabolic activity of the heterotrophic soil microbial community within samples. The results are presented as the summed response to all substrates in terms of control and time zero adjusted optical density (OD) at 590nm after 7 days incubation of BIOLOG plates at 20°C. Average activities over all soil samples ranged from 17.1 to 302.9 ( $OD_{590}$ ). The mean activity was 160.4 but the variation around this mean was large as indicated by a standard deviation of 35. This variation was more important than that for the number of viable cells in the samples. The distribution of global activity is shown in Figure 11.4. The Shapiro-Wilk's test of normality shows that the data were not normally distributed ( $p < 0.0000$ ). As for the abundance of viable cells, the data were skewed towards those samples with low activities.

Figure 11.3. Distribution of mean number of viable bacterial cells per gram dry weight of soil after 7 days incubation at 20°C on 10% nutrient agar.

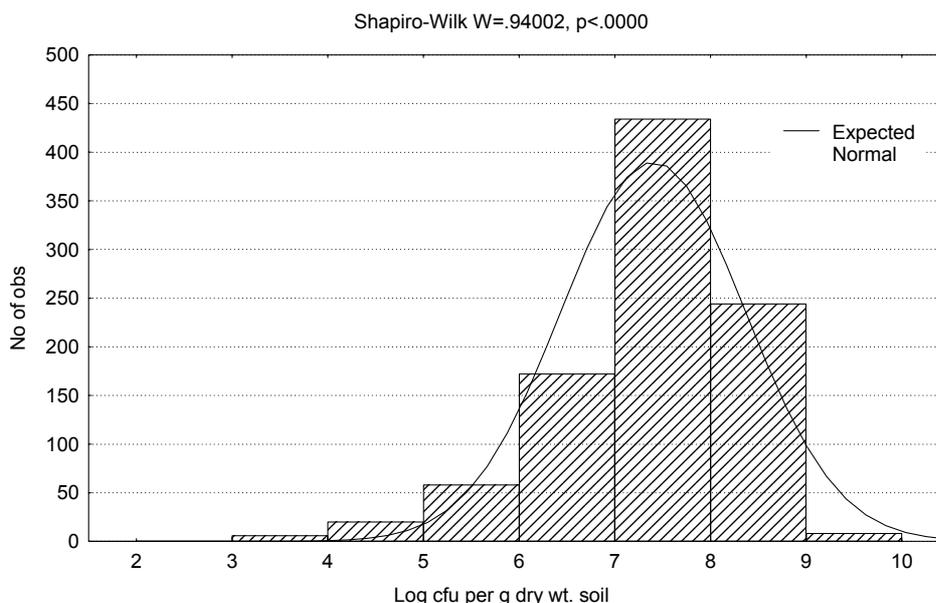
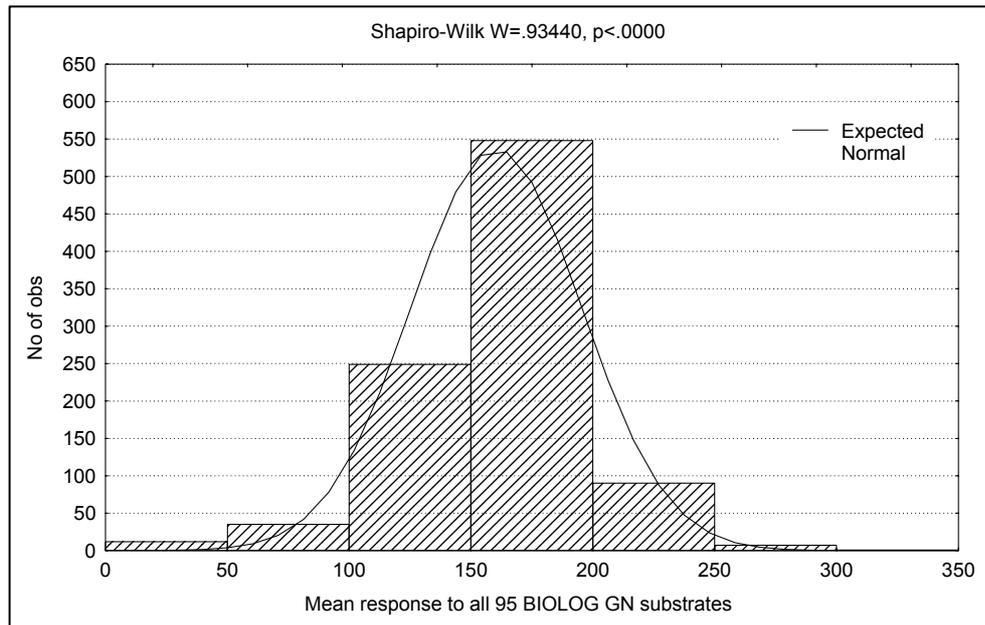


Figure 11.4. Distribution of global activity towards all 95 BIOLOG GN substrates after 7 days incubation at 20°C.



### 11.3.2. Environmental Zones

Summary data by Environmental Zone for mean number of viable cells, density of heterotrophic soil bacteria and response of extracted soil microbial communities to all 95 BIOLOG GN substrates are presented in Tables 11.2. to 11.4. The mean numbers of viable cells by Environmental Zone (EZ) ranged from  $1.74 \times 10^7$  to  $4.68 \times 10^7$  cfu per g. dry wt. soil (Table 11.2, Figure 11.5), with the highest in EZ 5 and lowest in EZ1. This showed that the variation in numbers of bacterial cells per sample was not evident at this broad level of grouping. The standard deviations and errors at this level of grouping were also low as the sheer number of samples analysed per Environmental Zone (between 90 and 226) and the broad ranging definitions of this grouping masked any subtle variations in the data. When the sample grouping is broken down in to smaller groupings such as the 39 ITE Land Classes (LC) or 22 Broad Habitat (BH) groups the variation in the data is more evident, as discussed below.

Table 11.2. Summary statistics for numbers of culturable heterotrophic soil bacteria per gram of soil after 7 days incubation on 10% nutrient agar at 20°C by Environmental Zone.

Environmental Zone	no. of samples	mean	median	minimum	maximum	sd	se
1	226	7.24	7.37	3.58	8.86	0.8	0.05
2	260	7.42	7.56	3.06	8.84	0.87	0.05
3	104	7.37	7.71	4.42	8.93	1.13	0.11
4	134	7.41	7.55	3.76	8.91	0.93	0.08
5	129	7.67	7.83	4.48	9.47	1.07	0.09
6	90	7.43	7.79	4.23	9.11	1.14	0.12
total	943	7.4	7.54	3.06	9.47	0.96	0.03

Figure 11.5. Mean number (+ 1.s.e) of viable bacterial cells per gram dry weight of soil after 7 days incubation at 20°C on 10% nutrient agar by Environmental Zone.

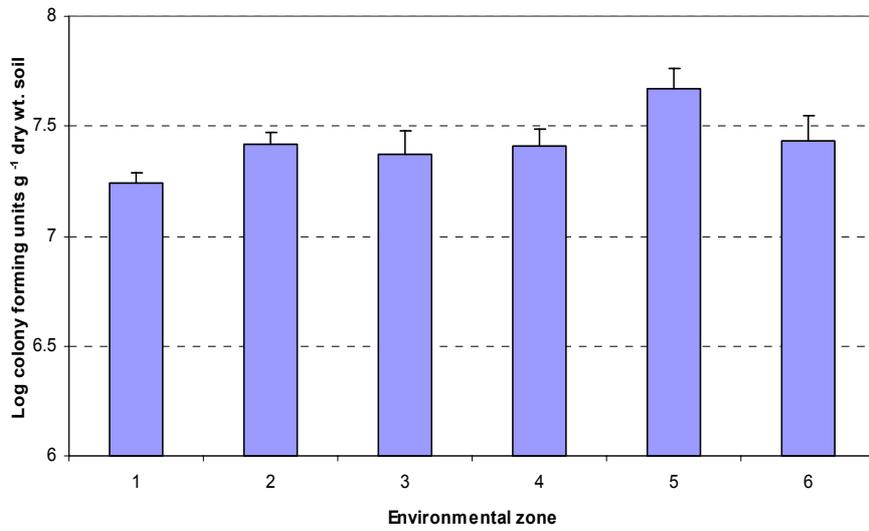
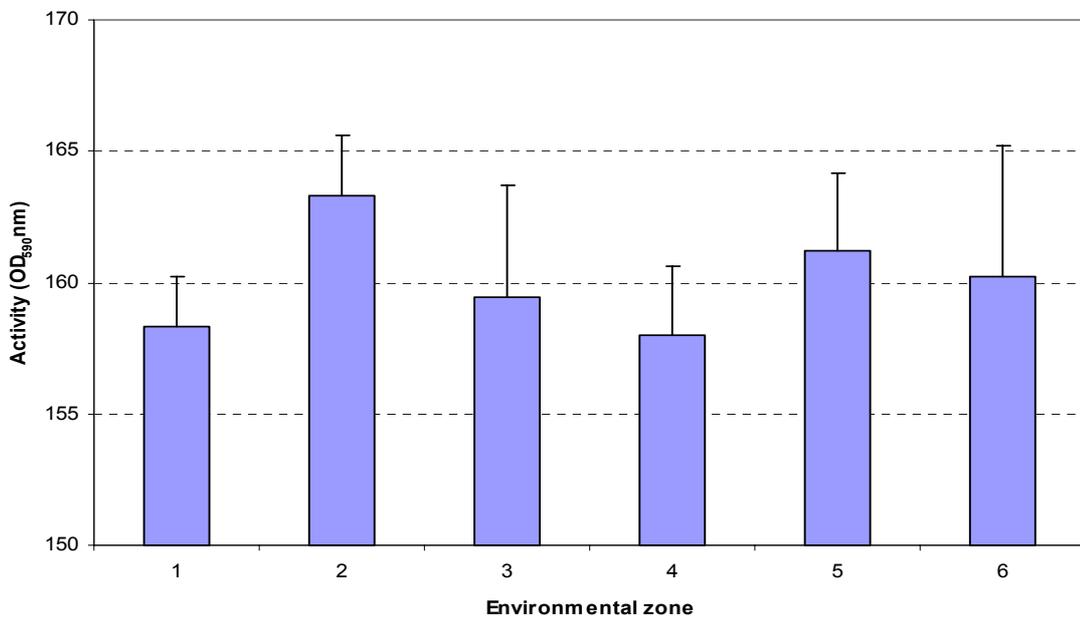


Figure 11.6. Mean global response of microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C by Environmental Zone. Bars indicate standard error of the mean.



The mean global activity by Environmental Zone (EZ) varied little, ranging from 158 to 163 (Table 11.4, Figure 11.6), with the highest activity in EZ 2 and lowest in EZ 1 and 4. However the standard deviations and errors for mean global activity at this level of grouping were quite high. When the sample grouping is broken down in to the 39 ITE Land Classes (LC) or 22 Broad Habitat (BH) groups the variation in the data was more evident.

Table 11.3. Density of heterotrophic soil bacteria per ml of dilution used to inoculate BIOLOG GN microplates, determined after 7 days incubation on 10% nutrient agar at 20°C. Summary data by Environmental Zone.

Environmental Zone	no. of samples	mean	median	minimum	maximum	sd	se
1	226	5.63	5.76	1.97	7.26	0.79	0.1
2	259	5.79	5.93	1.46	7.24	0.87	0.1
3	104	5.71	6.08	2.82	7.32	1.12	0.1
4	134	5.79	5.94	2.16	7.31	0.93	0.1
5	129	5.86	6.09	2.73	7.66	1.08	0.1
6	90	5.66	5.97	2.62	7.34	1.13	0.1
total	942	5.74	5.9	1.46	7.66	0.95	0.1

Table 11.4. Response of extracted soil microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C (OD<sub>590</sub>). Summary data by Environmental Zone.

Environmental Zone	no. of samples	mean	median	minimum	maximum	sd	se
1	228	158.36	156.49	57.91	284.8	28.15	1.86
2	263	163.33	164.57	22.12	249.67	36.5	2.25
3	107	159.45	161.51	17.07	288.01	43.74	4.23
4	128	157.99	157.4	23.59	225.37	29.65	2.62
5	129	161.24	158.07	34.58	302.9	33.4	2.94
6	89	160.26	156.57	22.88	285.43	43.08	4.57
total	944	160.39	159.77	17.07	302.9	34.98	1.14

### 11.3.3. Broad Habitat

Summary data by Broad Habitat mean number of viable cells, density of heterotrophic soil bacteria and response of extracted soil microbial communities to all 95 BIOLOG GN substrates are presented in Tables 11.5. to 11.7. Samples were grouped in to 17 out of a total of 22 possible Broad Habitat (BH) groups according to the vegetation and geological features of the sampling site (Table 11.5). The number of samples analysed per habitat ranged from 1 (BH groups 3, 16 and 19) to 306 (BH group 5, improved grasslands). The average numbers of viable bacterial cells per g dry wt. soil ranged from  $3.98 \times 10^6$  (BH group 16, n=1) to  $2.57 \times 10^8$  (BH group 18, n=2) as shown in Figure 11.7. Samples from woodland and grassland habitats generally had about  $3 \times 10^7$  cfu per g dry wt. soil. However samples from calcareous grassland sites (BH group 7) had a higher average number of viable bacterial cells per g dry wt soil ( $2 \times 10^8$ ) than samples from other grassland sites.

In terms of Broad Habitat groups, the mean global activity ranged from OD<sub>590</sub> 157 (BH group 9, n=16) to OD<sub>590</sub> 240 (BH group 19, n=1) as shown in Figure 11.8. In general the mean global activity showed little variation between Broad Habitat groups despite differences seen at this level in numbers of viable bacterial cells (Figure 11.7) and thus in BIOLOG inoculum densities.

Figure 11.7. Mean number (+ 1 s.e.) of viable bacterial cells per gram dry weight of soil after 7 days incubation at 20°C on 10% nutrient agar by Broad Habitat group.

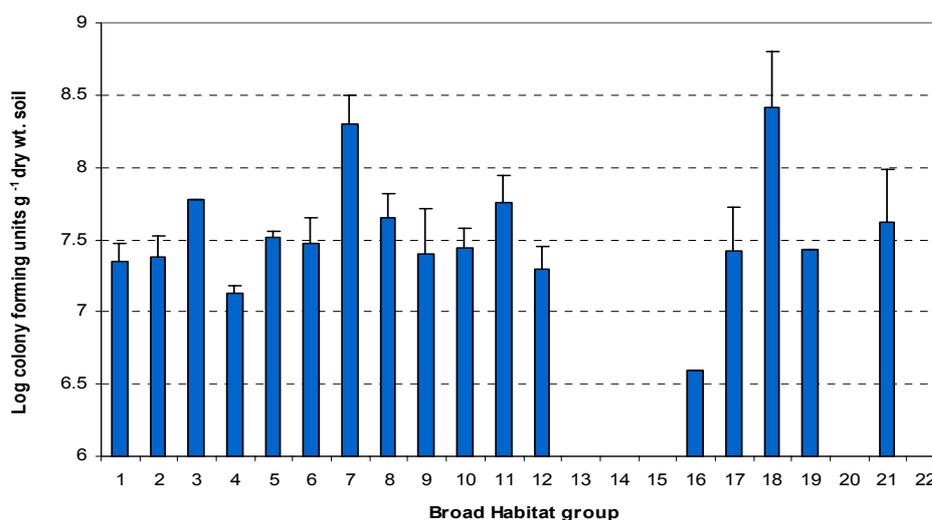


Table 11.5. Numbers of culturable heterotrophic soil bacteria per gram of soil after 7 days incubation on 10% nutrient agar at 20°C. Summary data by Broad Habitat.

Broad Habitat Group	Code	no. of samples	mean	median	Min.	Max.	sd
Broadleaved, mixed and yew woodland	1	61	7.35	7.48	3.58	8.88	0.93
Coniferous woodland	2	63	7.38	7.6	3.06	9.02	1.16
Boundary and linear features	3	1	7.78	7.78	7.78	7.78	.
Arable and horticultural	4	221	7.13	7.24	4.58	8.83	0.74
Improved grassland	5	306	7.52	7.61	4	8.82	0.77
Neutral grassland	6	42	7.47	7.85	3.76	8.84	1.2
Calcareous grassland	7	6	8.3	8.36	7.75	8.86	0.48
Acid grassland	8	54	7.65	7.96	4.25	9.27	1.26
Bracken	9	15	7.4	7.77	4.23	8.83	1.19
Dwarf shrub heath	10	75	7.44	7.74	4.56	9.13	1.18
Fen, marsh and swamp	11	32	7.76	8.09	5.17	9.47	1.03
Bog	12	69	7.3	7.47	4.48	9.47	1.25
Standing water and canals	13	.	.	.	.	.	.
Rivers and streams	14	.	.	.	.	.	.
Montane habitats	15	.	.	.	.	.	.
Inland rock	16	1	6.6	6.6	6.6	6.6	.
Built-up areas and gardens	17	10	7.42	7.43	5.33	8.53	0.94
Supralittoral rock	18	2	8.41	8.41	8.02	8.79	0.54
Supralittoral sediment	19	1	7.43	7.43	7.43	7.43	.
Littoral rock	20	.	.	.	.	.	.
Littoral sediment	21	6	7.62	7.65	6.58	8.5	0.9
Sea	22	.	.	.	.	.	.

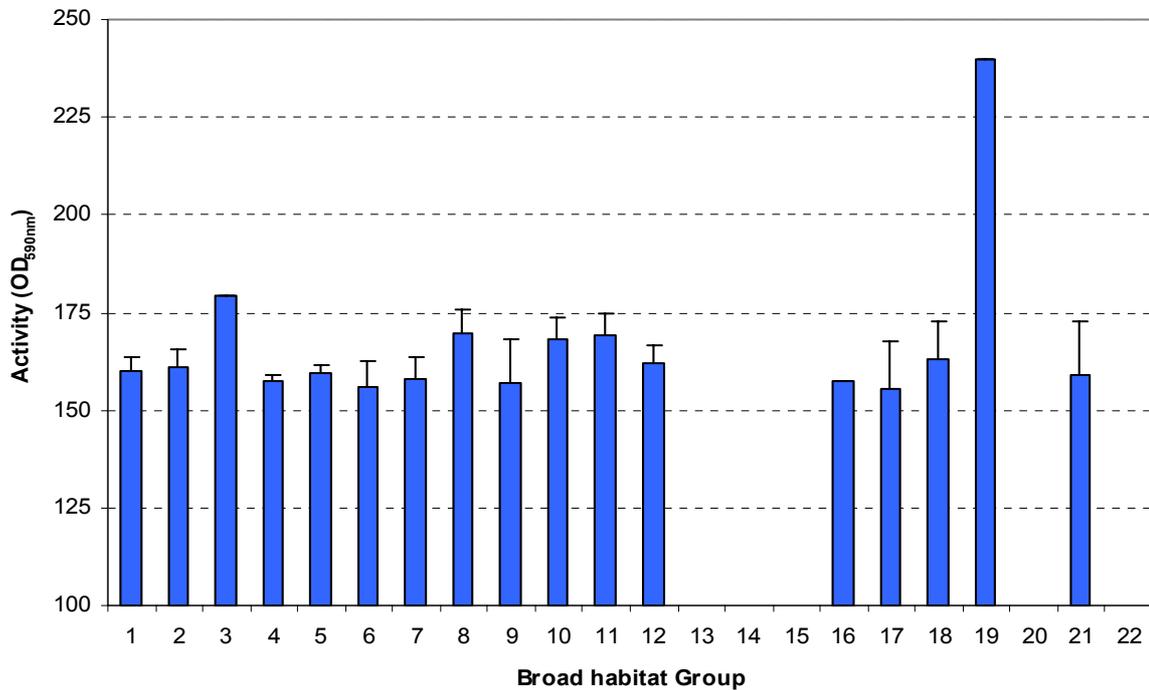
Table 11.6. Density of heterotrophic soil bacteria per ml of dilution used to inoculate BIOLOG GN microplates, determined after 7 days incubation on 10% nutrient agar at 20°C. Summary data by Broad Habitat.

Broad Habitat Group	code	no. of samples	mean	median	minimum	maximum	sd
Broadleaved, mixed and yew woodland	1	61	5.71	5.88	1.97	7.19	0.92
Coniferous woodland	2	63	5.62	5.92	1.46	7.1	1.12
Boundary and linear features	3	1	6.18	6.18	6.18	6.18	.
Arable and horticultural	4	221	5.53	5.63	2.97	7.23	0.74
Improved grassland	5	306	5.92	6.01	2.4	7.21	0.77
Neutral grassland	6	42	5.86	6.25	2.16	7.24	1.19
Calcareous grassland	7	6	6.7	6.76	6.15	7.26	0.48
Acid grassland	8	54	5.91	6.24	2.64	7.66	1.23
Bracken	9	15	5.74	6.16	2.62	6.93	1.14
Dwarf shrub heath	10	75	5.67	5.99	2.95	7.31	1.17
Fen, marsh and swamp	11	31	6.02	6.29	3.57	7.57	1
Bog	12	69	5.38	5.32	2.73	7.57	1.25
Standing water and canals	13	.	.	.	.	.	.
Rivers and streams	14	.	.	.	.	.	.
Montane habitats	15	.	.	.	.	.	.
Inland rock	16	1	5	5	5	5	.
Built-up areas and gardens	17	10	5.82	5.83	3.72	6.93	0.95
Supralittoral rock	18	2	6.66	6.66	6.42	6.89	0.33
Supralittoral sediment	19	1	5.83	5.83	5.83	5.83	.
Littoral rock	20	.	.	.	.	.	.
Littoral sediment	21	6	6.02	6.05	4.98	6.9	0.9
Sea	22	.	.	.	.	.	.

Table 11.7. Response of extracted soil microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C (OD<sub>590</sub>). Summary data by Broad Habitat.

Broad Habitat Group	code	no. of samples	mean	median	minimum	maximum	sd
Broadleaved, mixed and yew woodland	1	64	160.1	157.14	46.88	242.96	29.44
Coniferous woodland	2	64	160.8	158.74	50.82	261.19	39.9
Boundary and linear features	3	1	179.3	179.28	179.28	179.28	.
Arable and horticultural	4	221	157.4	155.85	64.94	251.86	26.92
Improved grassland	5	305	159.5	161.99	17.07	284.8	35.62
Neutral grassland	6	42	156.1	159.02	23.59	234.03	40.93
Calcareous grassland	7	6	157.9	155.04	138.44	175.54	13.85
Acid grassland	8	54	169.7	171.66	66.13	285.43	43.71
Bracken	9	16	156.9	159.92	22.88	223.56	44.71
Dwarf shrub heath	10	75	168.3	169.73	51.39	288.01	45.71
Fen, marsh and swamp	11	31	169.1	159.2	129.7	228.47	30.49
Bog	12	66	161.8	156.5	52.31	302.9	39.5
Standing water and canals	13	.	.	.	.	.	.
Rivers and streams	14	.	.	.	.	.	.
Montane habitats	15	.	.	.	.	.	.
Inland rock	16	1	157.5	157.51	157.51	157.51	.
Built-up areas and gardens	17	10	155.4	163.6	47.64	191.4	39.27
Supralittoral rock	18	2	162.8	162.8	152.87	172.74	14.05
Supralittoral sediment	19	1	240	240.04	240.04	240.04	.
Littoral rock	20	.	.	.	.	.	.
Littoral sediment	21	6	159	152.56	117.11	210.36	33.57
Sea	22	.	.	.	.	.	.

Figure 11.8. Mean global response of microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C by Broad Habitat group. Bars indicate standard error of the mean



#### 11.3.4. ITE Land Class

Summary data by ITE Land Class for mean number of viable cells, density of heterotrophic soil bacteria and response of extracted soil microbial communities to all 95 BIOLOG GN substrates are presented in Tables 11.8 to 11.10.

Samples grouped in ITE Land Class 22e (margins of high mountains, moorlands, often afforested, England) had the lowest mean number of viable cells per g dry wt. of soil ( $2.75 \times 10^6$ , n=10). Samples in Land Class 18s (rounded hills, some steep slopes, varied moorlands, Scotland) had the highest number of bacterial cells ( $9.12 \times 10^8$ , n=3). These two Land Classes are not dissimilar, in terms of altitude, soil type and vegetation, so it is surprising to find such a variation in the numbers of viable bacterial cells in samples from the two classes. Samples in Land Class 13s (variable, mainly flat land forms with heterogeneous land use, Scotland) also had a low mean number of viable cells per g dry wt. of soil. However, the definitions of the LC grouping are also very broad ranging (Barr, 1998) and thus it is difficult to draw conclusions at this level.

Samples grouped in ITE Land Class 13s (variable, flat land forms with heterogeneous land use, Scotland) had the lowest global activity (OD<sub>590</sub> 125.6, n=6) and samples in Land Class 18s (rounded hills, some steep slopes, varied moorlands, Scotland) had the highest global activity (OD<sub>590</sub> 207.6, n=3); Table 11.10. Samples in these Land Classes (both in Scotland) also had low and high numbers of viable bacteria and BIOLOG inoculum densities respectively; Tables 11.8 and 11.9.

Table 11.8. Numbers of culturable heterotrophic soil bacteria per gram of soil after 7 days incubation on 10% nutrient agar at 20°C. Summary data by ITE Land Class.

ITE Land Class	Code	no. of samples	mean	median	minimum	maximum	sd
1e	1	28	7.39	7.45	5.9	8.57	0.62
2e	2	44	7.51	7.49	3.58	8.86	0.81
3e	3	46	6.98	7.24	4	8.42	0.95
4e	4	14	7.26	7.45	6.31	8.57	0.78
5e	5	11	7.83	7.77	7.42	8.44	0.35
6e	6	34	7.46	7.7	4.85	8.82	1
7e	7	35	7.61	7.68	6.47	8.62	0.49
7s	8	19	7.94	8.04	7.07	8.74	0.49
8e	9	31	7.62	7.73	4.56	8.65	0.9
9e	10	44	7.35	7.47	4.85	8.36	0.66
10e	11	50	6.98	7.26	3.06	8.72	1.19
11e	12	49	7.18	7.27	5.73	8.37	0.65
12e	13	20	7	7.12	5.24	8.83	0.88
13e	14	26	7.5	7.79	6.28	8.58	0.63
13s	15	10	6.66	6.32	4.43	8.91	1.48
15e	16	16	7.79	7.9	6.6	8.36	0.53
16e	17	29	7.28	7.39	5.58	8.84	0.76
17e	18	20	7.71	7.76	5.27	8.78	0.89
17w1	19	10	7.38	7.57	4.42	8.26	1.08
17w2	20	.	.	.	.	.	.
17w3	21	14	8.02	8.02	6.18	8.82	0.65
18e	22	24	7.09	7.53	4.85	8.7	1.31
18s	23	3	8.96	8.83	8.58	9.47	0.46
19e	24	15	7.47	7.53	5.23	8.93	0.97
19s	25	5	8.19	8.12	7.64	8.7	0.41
21s	26	26	7.72	7.75	6.38	9.11	0.73
22e	27	10	6.44	6.26	4.92	7.98	1.13
22s	28	19	7.58	7.87	4.23	8.88	1.19
23e	29	11	7.28	7.26	4.56	8.83	1.27
23s	30	20	7.26	7.55	4.7	8.33	0.97
24s	31	25	7.15	7.83	4.25	8.94	1.5
25e	32	9	7.5	7.53	6.62	8.3	0.63
25s	33	43	7.41	7.54	3.76	8.81	0.94
26s	34	31	7.33	7.46	5.67	8.39	0.73
27s	35	31	7.41	7.57	4.52	8.7	0.95
28s	36	30	7.62	7.59	6.72	9.02	0.62
29s	37	27	7.71	7.9	4.48	9.27	1.29
30s	38	13	7.98	8.45	6.05	9.13	0.91
31s	39	23	7.71	8.17	5.3	9.14	1.04
32s	40	28	7.27	7.54	5.02	9.18	1.29

Table 11.9. Density of heterotrophic soil bacteria per ml of dilution used to inoculate BIOLOG GN microplates, determined after 7 days incubation on 10% nutrient agar at 20°C. Summary data by ITE Land Class.

ITE Land Class	Code	no. of samples	mean	median	minimum	maximum	sd
1e	1	28	5.74	5.85	4.29	6.96	0.66
2e	2	44	5.91	5.89	1.97	7.26	0.81
3e	3	46	5.38	5.63	2.4	6.82	0.95
4e	4	14	5.66	5.85	4.71	6.97	0.78
5e	5	11	6.22	6.17	5.82	6.84	0.35
6e	6	34	5.81	6.07	2.95	7.19	1.02
7e	7	35	6.01	6.08	4.87	7.02	0.49
7s	8	19	6.32	6.44	5.47	7.14	0.48
8e	9	30	5.96	6.07	2.95	7.05	0.88
9e	10	44	5.72	5.85	3.24	6.76	0.66
10e	11	50	5.36	5.63	1.46	7.12	1.18
11e	12	49	5.58	5.67	4.13	6.76	0.65
12e	13	20	5.39	5.51	3.63	7.23	0.88
13e	14	26	5.89	6.19	4.68	6.98	0.63
13s	15	10	4.99	4.72	2.83	7.31	1.53
15e	16	16	6.19	6.3	5	6.76	0.53
16e	17	29	5.67	5.79	3.98	7.24	0.76
17e	18	20	6.11	6.16	3.67	7.18	0.89
17w1	19	10	5.78	5.97	2.82	6.65	1.08
17w2	20	.	.	.	.	.	.
17w3	21	14	6.35	6.42	4.58	7.21	0.65
18e	22	24	5.48	5.93	3.24	7.1	1.31
18s	23	3	7.05	6.92	6.67	7.57	0.46
19e	24	15	5.72	5.87	3.62	7.32	0.97
19s	25	5	5.86	5.8	5.04	6.85	0.81
21s	26	26	5.9	6.05	4.48	7.21	0.77
22e	27	10	4.69	4.52	3.02	6.38	1.13
22s	28	19	5.78	6	2.62	6.98	1.11
23e	29	11	5.54	5.65	2.96	6.93	1.18
23s	30	20	5.53	5.79	3.1	6.73	0.94
24s	31	25	5.42	6.23	2.64	7.34	1.54
25e	32	9	5.89	5.92	5.02	6.7	0.63
25s	33	43	5.77	5.94	2.16	7.21	0.93
26s	34	31	5.73	5.86	4.06	6.79	0.74
27s	35	31	5.8	5.97	2.91	7	0.94
28s	36	30	5.79	5.8	4.15	6.75	0.61
29s	37	27	5.95	6.29	2.73	7.66	1.3
30s	38	13	6.07	6.54	4.15	7.23	0.91
31s	39	23	6.03	6.45	3.7	7.32	1.06
32s	40	28	5.47	5.78	3.12	7.25	1.33

Table 11.10. Response of extracted soil microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C (OD<sub>590</sub>). Summary data by ITE Land Class.

ITE Land Class	Code	no. of samples	mean	median	minimum	maximum	sd
1e	1	28	168.96	169.09	104.56	220.7	23.34
2e	2	47	157.71	153.41	106.36	284.8	26.76
3e	3	47	148.92	151.6	57.91	201.91	26.71
4e	4	14	166.15	162.56	128	237.47	29.14
5e	5	11	163.42	164.26	138.03	187.14	12.53
6e	6	35	162.13	162.08	66.13	211.17	26.34
7e	7	35	169.49	167.26	80.48	240.04	35.04
7s	8	19	156.71	159.15	102.62	178.92	16.83
8e	9	32	165.57	162.78	51.12	249.67	36.37
9e	10	42	170.89	168.64	95.43	251.86	27.8
10e	11	50	155.09	163.13	44.72	216.56	46.77
11e	12	49	162.3	160.49	95.68	245.04	25.33
12e	13	20	145.16	147.31	85.9	233.63	30.86
13e	14	26	177.28	178.55	101.08	234.03	27.02
13s	15	6	125.64	144.98	34.06	169.01	49.77
15e	16	16	164.59	155.85	137.75	228.47	25.91
16e	17	30	150.81	160.32	22.12	210.36	50.93
17e	18	20	174.22	172.16	47.64	242.96	38.71
17w1	19	10	132.74	148.63	29.77	166.3	42.05
17w2	20	.	.	.	.	.	.
17w3	21	16	166.46	160.29	133.39	232.95	25.23
18e	22	25	144.19	152.61	17.07	204.2	42.48
18s	23	3	207.64	211.14	184.49	227.27	21.6
19e	24	15	184.94	169.67	142.7	288.01	39.77
19s	25	5	159.25	156.25	144.23	177.84	13.13
21s	26	27	157.37	157.23	80.48	203.34	24.23
22e	27	10	150.6	131.05	62.85	241.03	56.23
22s	28	19	159.67	163.66	22.88	261.19	51.55
23e	29	11	154.65	169.89	51.39	208.73	51.12
23s	30	20	180.65	183.41	92.81	285.43	54.16
24s	31	23	146.4	139.88	52.31	240.53	37.86
25e	32	9	148.4	156.07	85.53	176.7	26.58
25s	33	41	158.73	156.56	23.59	225.37	37.44
26s	34	31	158.28	155.49	86.98	217.42	23.83
27s	35	31	163.76	162.83	110.56	210.59	21.38
28s	36	30	157.59	154.47	93.64	235.07	23.32
29s	37	27	173.64	160.78	131.12	302.9	35.8
30s	38	13	146.9	150.95	87.85	177.59	25.99
31s	39	23	160.55	155.9	103.28	217.21	28.54
32s	40	28	155.77	157.94	34.58	232.07	43.68

Figure 11.9. Mean number (+ 1 s.e.) of viable bacterial cells per gram dry weight of soil after 7 days incubation at 20°C on 10% nutrient agar by ITE Land Class.

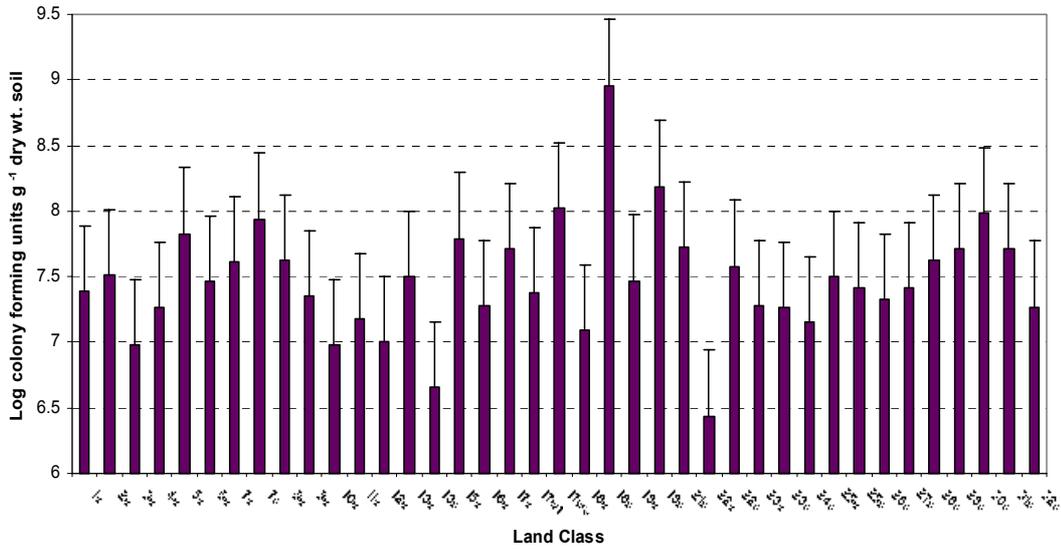
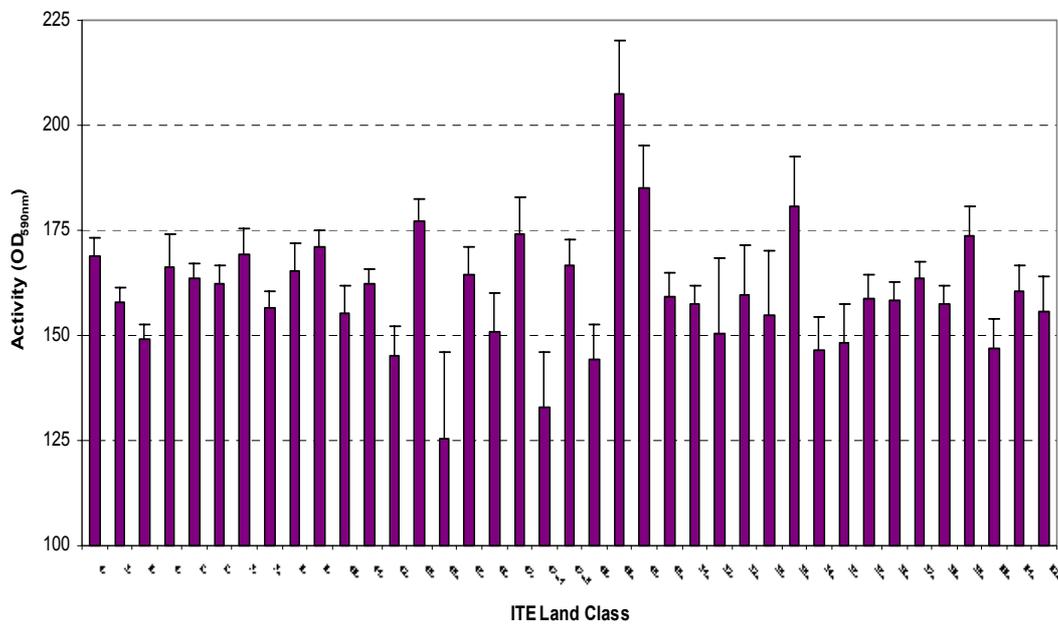


Figure 11.10. Mean global response of microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C by ITE Land Class. Bars indicate standard error of the mean



### 11.3.5. CVS Aggregate Vegetation Class

Summary data by CVS Aggregate Vegetation Class for mean number of viable cells, density of heterotrophic soil bacteria and response of extracted soil microbial communities to all 95 BIOLOG GN substrates are presented in Tables 11.11 to 11.13.

Above-ground vegetation dictates the type and amount of substrates entering the below-ground soil system through leaf litter and root exudates. In order to examine impacts of vegetation on soil microbial communities, samples were grouped in to 8 Aggregate Vegetation Classes (AVC) according to the predominant vegetation present at the sampling site (Table 11.11).

The numbers of samples analysed per vegetation class ranged from 29 (AVC 5, lowland woodland) to 208 (AVC 4, infertile grassland). However, there was little variation in the average numbers of viable bacterial cells per g dry wt. of soil according to this broad vegetation grouping (Figure 11.11). When the data is examined further (Figure 11.12) we can see that the median values for several vegetation classes (e.g., AVC 1, 3 and 4) were influenced by extreme and outlying low values for certain samples. The same was seen for mean values (graph not shown). In depth analyses of samples within each of the vegetation classes is necessary before relationships between vegetation and other site and soil parameters can be determined e.g., between vegetation and soil type or vegetation and soil organic matter. The time of sampling and soil moisture content may also play a role in determining the microbiological abundance and activity of samples within a certain vegetation class.

When samples were grouped according to the predominant vegetation of the sampling site, there was little variation in mean global activity (Figure 11.13). Samples from AVC groups 4 and 6 (infertile grassland, upland wooded) had the lowest mean global activities and those from AVC groups 5 and 8 (lowland wooded, heaths and bogs) had the highest activities. When the data is examined further (Figure 11.14) the median activity values for several vegetation classes (e.g., AVC 1, 3 and 4) seem to be influenced by extreme and outlying low and high values for certain samples. Extreme low values were also seen to influence numbers of viable bacterial cells in samples from these groups (see above). Again, a more in depth analyses of samples within each of the vegetation classes is necessary before relationships between soil microbial activities, site vegetation and other site and soil parameters can be determined.

Table 11.11. Numbers of culturable heterotrophic soil bacteria per gram of soil after 7 days incubation on 10% nutrient agar at 20°C. Summary data by Aggregate Vegetation Class.

Aggregate Vegetation Class	code	no. of samples	mean	median	minimum	maximum	sd
Crops and weeds	1	178	7.08	7.17	3.76	8.83	0.8
Tall grass and herb	2	59	7.28	7.36	4.56	8.64	0.76
Fertile grassland	3	192	7.56	7.6	4	8.81	0.69
Infertile grassland	4	208	7.57	7.71	4.42	8.93	0.85
Lowland wooded	5	29	7.24	7.44	3.58	8.79	1.02
Upland wooded	6	59	7.33	7.72	3.06	8.75	1.11
Moorland grass mosaics	7	113	7.54	7.87	4.23	9.47	1.26
Heath and bog	8	105	7.37	7.65	4.25	9.18	1.24

Figure 11.11. Mean number (+ 1 s.e.) of viable bacterial cells per gram dry weight of soil after 7 days incubation at 20°C on 10% nutrient agar by Aggregate Vegetation Class.

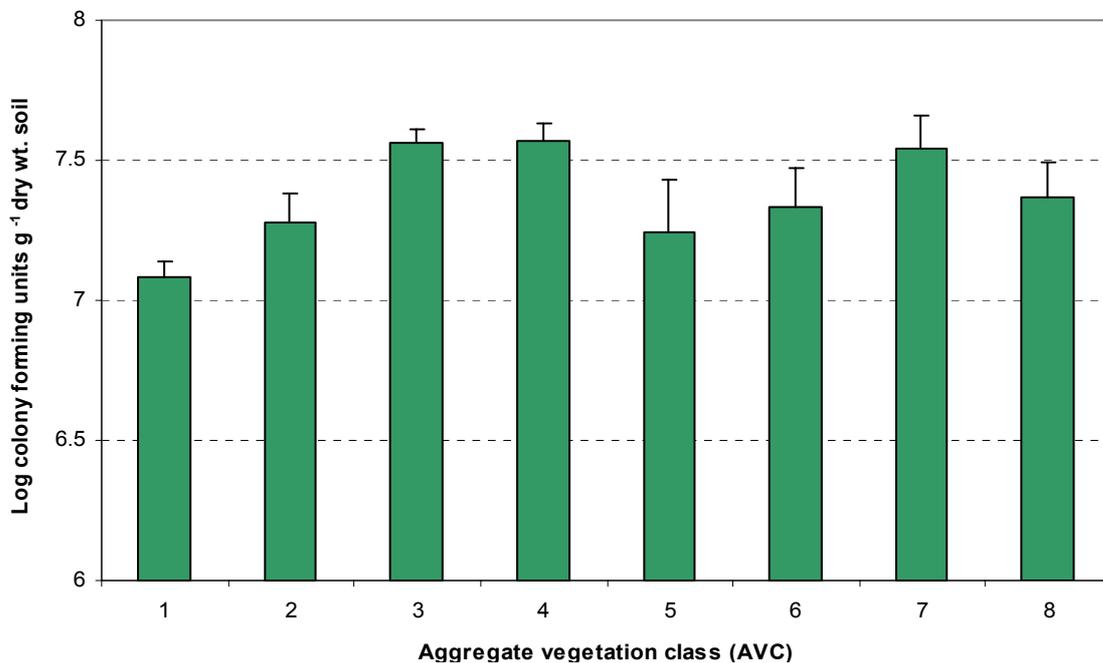


Table 11.12. Density of heterotrophic soil bacteria per ml of dilution used to inoculate BIOLOG GN microplates, determined after 7 days incubation on 10% nutrient agar at 20°C. Summary data by Aggregate Vegetation Class.

Aggregate Vegetation Class	code	no. of samples	mean	median	minimum	maximum	sd
Crops and weeds	1	178	5.47	5.56	2.16	7.23	0.8
Tall grass and herb	2	58	5.65	5.76	2.95	6.98	0.73
Fertile grassland	3	192	5.95	5.99	2.4	7.21	0.69
Infertile grassland	4	208	5.96	6.11	2.82	7.32	0.85
Lowland wooded	5	29	5.59	5.75	1.97	7.19	1.04
Upland wooded	6	59	5.63	5.96	1.46	7.1	1.1
Moorland grass mosaics	7	113	5.81	6.12	2.62	7.66	1.22
Heath and bog	8	105	5.46	5.57	2.64	7.32	1.22

Table 11.13. Response of extracted soil microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C (OD<sub>590</sub>). Summary data by CVS Aggregate Vegetation Class.

Aggregate Vegetation Class	code	no. of samples	mean	median	minimum	maximum	sd
Crops and weeds	1	178	155.2	153.26	23.59	249.67	28.98
Tall grass and herb	2	60	159.2	160.15	51.12	251.86	27.78
Fertile grassland	3	192	164.5	165.1	22.12	284.8	32.11
Infertile grassland	4	209	156.1	159.6	17.07	248.71	35.63
Lowland wooded	5	32	168.8	164.42	138.41	245.04	23.92
Upland wooded	6	60	156.6	153.73	50.82	261.19	41.53
Moorland grass mosaics	7	110	163.5	164.2	22.88	259.96	42.32
Heath and bog	8	103	167.2	161.38	80.48	302.9	39.93

Figure 11.12. A box whisker plot showing the median number of viable bacterial cells per gram dry weight of soil by Aggregate Vegetation Class. Minimum, maximum, 25 and 75% quartile and outlying values are also shown.

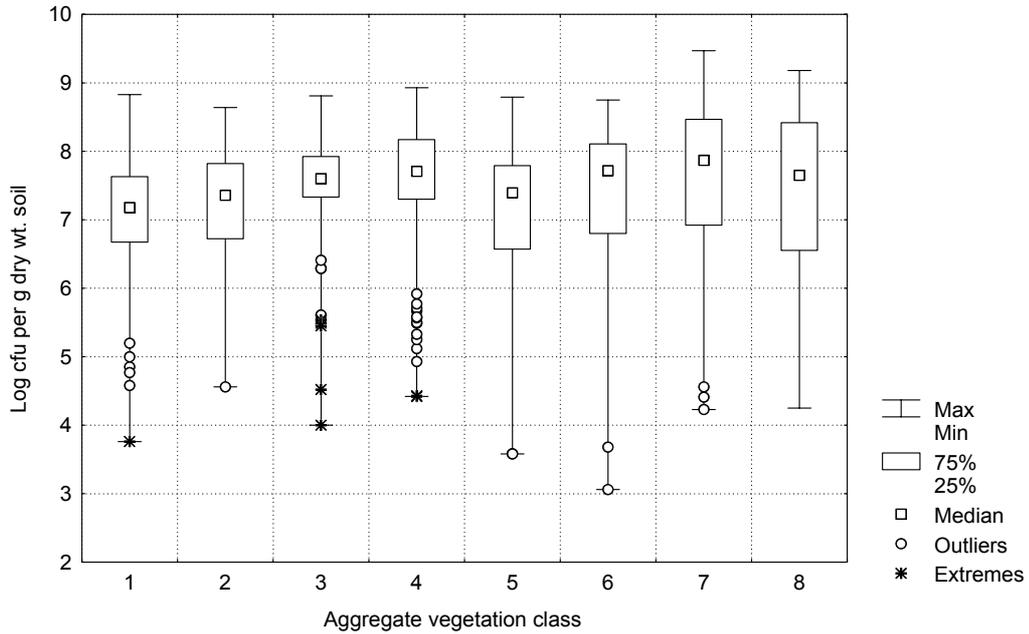


Figure 11.13. Mean global response of microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C by Aggregate Vegetation Class. Bars indicate standard error of the mean.

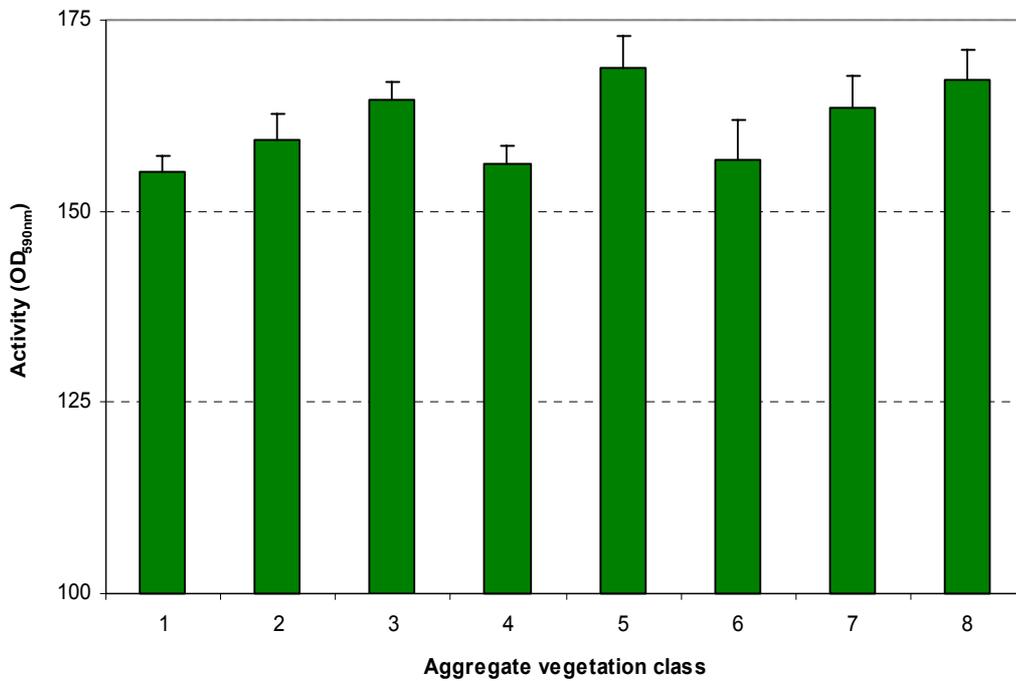
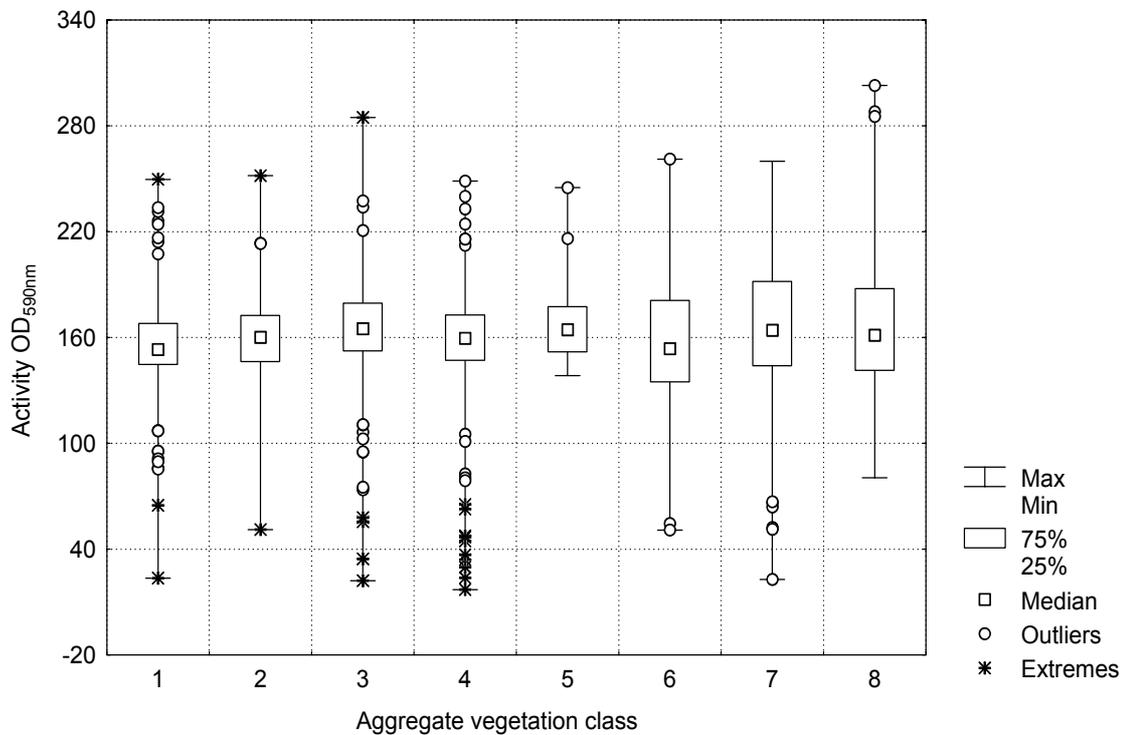


Figure 11.14. A box whisker plot showing the median level of global activity by Aggregate Vegetation Class. Minimum, maximum, 25 and 75% quartile and outlying values are also shown.



### 11.3.6. Major Soil Group

Summary data by Major Soil Group for mean number of viable cells, density of heterotrophic soil bacteria and response of extracted soil microbial communities to all 95 BIOLOG GN substrates are presented in Tables 11.14 to 11.16.

The soil type and texture (e.g., clay, sand, silt) can play a major role not only on the numbers of viable bacteria in the soil samples but also on the numbers of bacterial cells extracted from soil samples. Samples were grouped in to 7 out of a total of 10 Major Soil Groups (MSG) with between 24 (MSG 4, pelosols) and 170 (MSG 7, surface-water gley soils) samples analysed per group (Table 11.14). As for the vegetation class grouping above, little variation was seen in the average numbers of viable bacterial cells per g dry wt. of soil according to Major Soil Groups (Figure 11.15).

A box whisker plot of the data (Figure 11.16) shows that the median values for samples in each soil group were influenced by outliers with low numbers of viable bacterial cells per g dry wt. of soil. A similar trend is seen for the mean values for each group (graph not shown). A large number of outlying values were seen in samples belonging to soil group 5 (brown soils), further analysis need to be done to determine why certain samples contains very low numbers of viable bacterial cells. Vegetation, soil moisture and sampling season may influence the numbers of microorganisms and relationships with these parameters need to be further explored.

As noted for the vegetation class grouping above, little variation was seen in the mean global activities of samples when they were grouped according to Major Soil Groups (Figure 11.17). Samples grouped in to soil types 3, 4, 5 and 8 had a lower average global activity of OD<sub>590</sub> 158 and those in soil groups 6,7 and 8 had a slightly higher mean global activity of OD<sub>590</sub> 165. A box whisker plot of the data (Figure 11.18) shows that the median values for samples in each soil group were influenced by samples with extreme and outlying values for global activity. In particular, samples belonging to soil group 5 (brown soils) had a large number of extreme and outlying values. Relationships between samples in each soil group and other site and soil parameters need to be determined with further analysis of this data set.

Table 11.14. Numbers of culturable heterotrophic soil bacteria per gram of soil after 7 days incubation on 10% nutrient agar at 20°C. Summary Major Soil Group.

Major Soil Group	Code	no. of samples	mean	median	minimum	maximum	sd
Terrestrial Raw Soil	1	.	.	.	.	.	.
Raw Gley Soils	2	.	.	.	.	.	.
Lithomorphc Soils	3	81	7.55	7.71	4.25	9.13	0.99
Pelosols	4	24	7.22	7.24	6.12	8.37	0.54
Brown Soils	5	336	7.45	7.55	4.42	8.88	0.82
Podzolic Soils	6	128	7.5	7.69	4.41	9.47	1.05
Surface-water Gley Soils	7	170	7.3	7.47	3.06	9.18	0.98
Ground-water Gley Soils	8	122	7.26	7.43	3.58	8.84	1.01
Man-made Soils	9	.	.	.	.	.	.
Peat (organic) soils	10	82	7.41	7.62	4.23	9.14	1.19

Table 11.15. Density of heterotrophic soil bacteria per ml of dilution used to inoculate BIOLOG GN microplates, determined after 7 days incubation on 10% nutrient agar at 20°C. Summary data by Major Soil Group.

Major Soil Group	code	no. of samples	mean	median	minimum	maximum	sd
Terrestrial Raw Soil	1	.	.	.	.	.	.
Raw Gley Soils	2	.	.	.	.	.	.
Lithomorphc Soils	3	81	5.9	6.03	2.64	7.34	0.99
Pelosols	4	24	5.62	5.64	4.52	6.76	0.54
Brown Soils	5	336	5.83	5.94	2.82	7.26	0.81
Podzolic Soils	6	128	5.8	6.05	2.81	7.66	1.04
Surface-water Gley Soils	7	170	5.63	5.85	1.46	7.32	0.97
Ground-water Gley Soils	8	121	5.65	5.82	1.97	7.24	1.01
Man-made Soils	9	.	.	.	.	.	.
Peat (organic) soils	10	82	5.51	5.75	2.62	7.24	1.18

Table 11.16. Response of extracted soil microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C (OD<sub>590</sub>). Summary data by Major Soil Group

Major Soil Group	code	no. of samples	mean	median	minimum	maximum	sd
Terrestrial Raw Soil	1	.	.	.	.	.	.
Raw Gley Soils	2	.	.	.	.	.	.
Lithomorphic Soils	3	78	158.2	156.78	51.39	259.96	29.29
Pelosols	4	24	157.6	155.39	85.9	216.17	29.15
Brown Soils	5	336	157.1	158.43	17.07	245.04	31.94
Podzolic Soils	6	128	166.8	164.4	52.31	253.65	36.4
Surface-water Gley Soils	7	171	163.8	163.11	23.83	251.86	37.18
Ground-water Gley Soils	8	125	157.3	158.64	23.59	261.19	36.4
Man-made Soils	9	.	.	.	.	.	.
Peat (organic) soils	10	82	164.2	163.17	22.88	302.9	42.26

Figure 11.15. Mean number (+ 1 s.e.) of viable bacterial cells per gram dry weight of soil after 7 days incubation at 20°C on 10% nutrient agar by Major Soil Group.

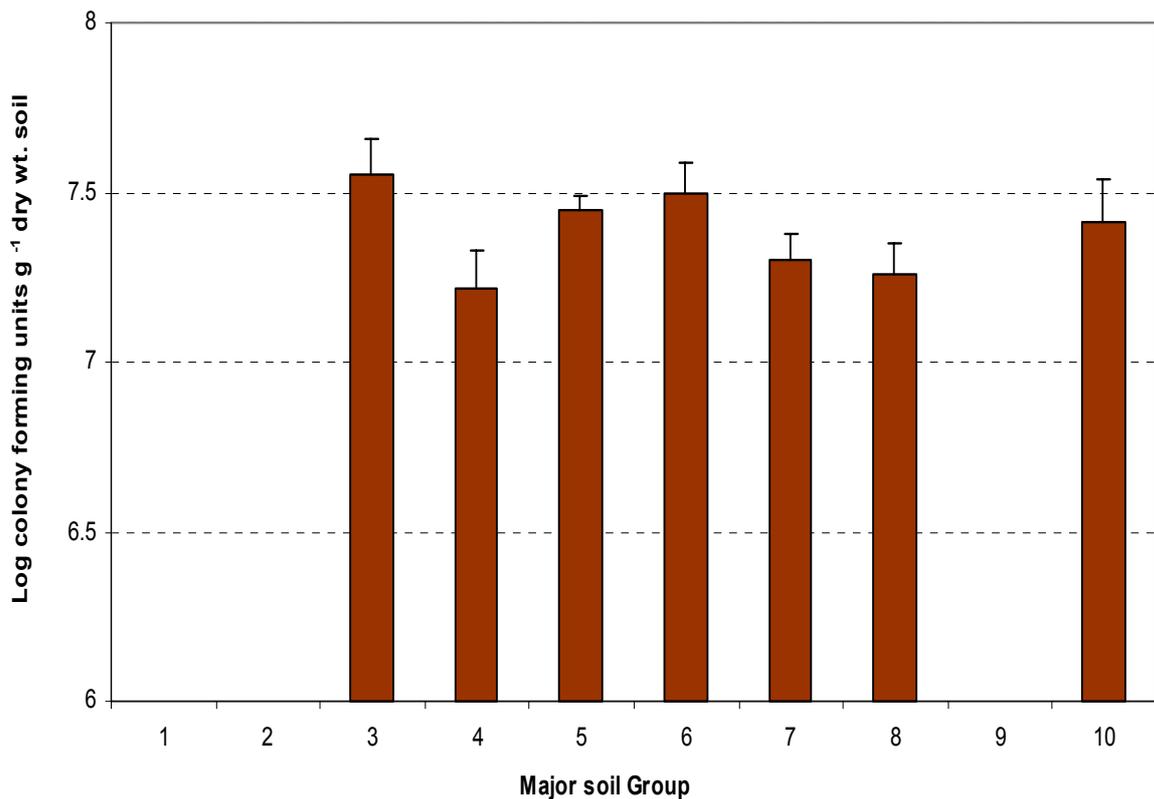


Figure 11.16. A box whisker plot showing the median number of viable bacterial cells per gram dry weight of soil by Aggregate Vegetation Class. Minimum, maximum, 25 and 75% quartile and outlying values are also shown.

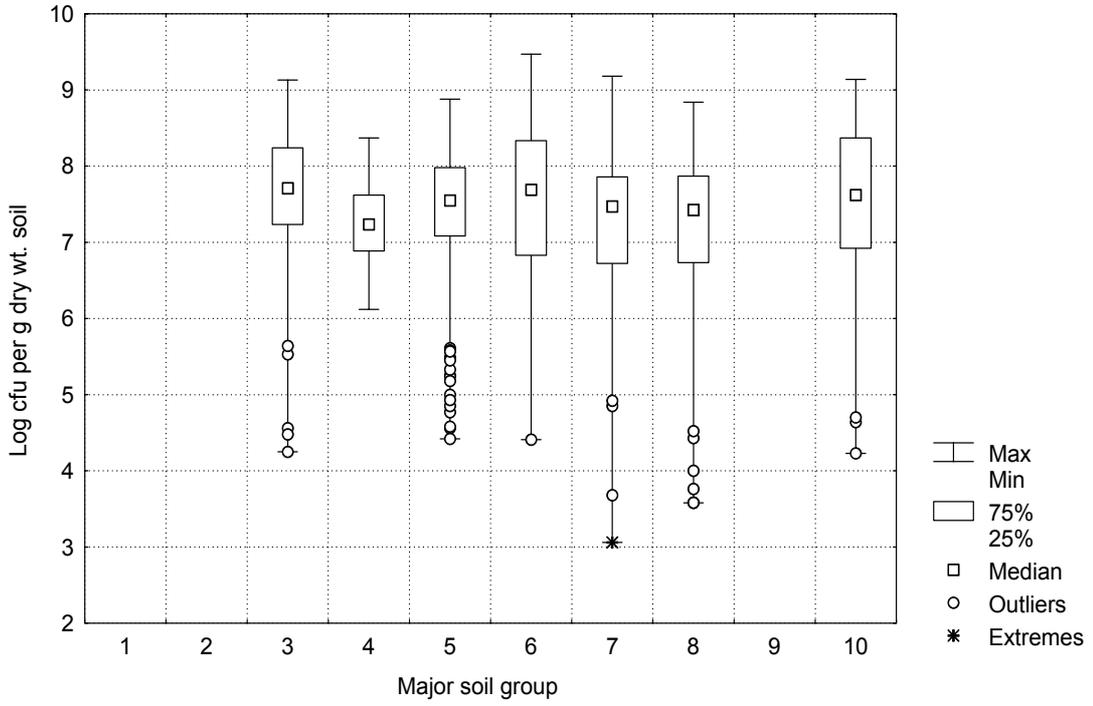


Figure 11.17. Mean global response of microbial communities to all 95 BIOLOG GN substrates after 7 days incubation at 20°C by Major Soil Group. Bars indicate standard error of the mean.

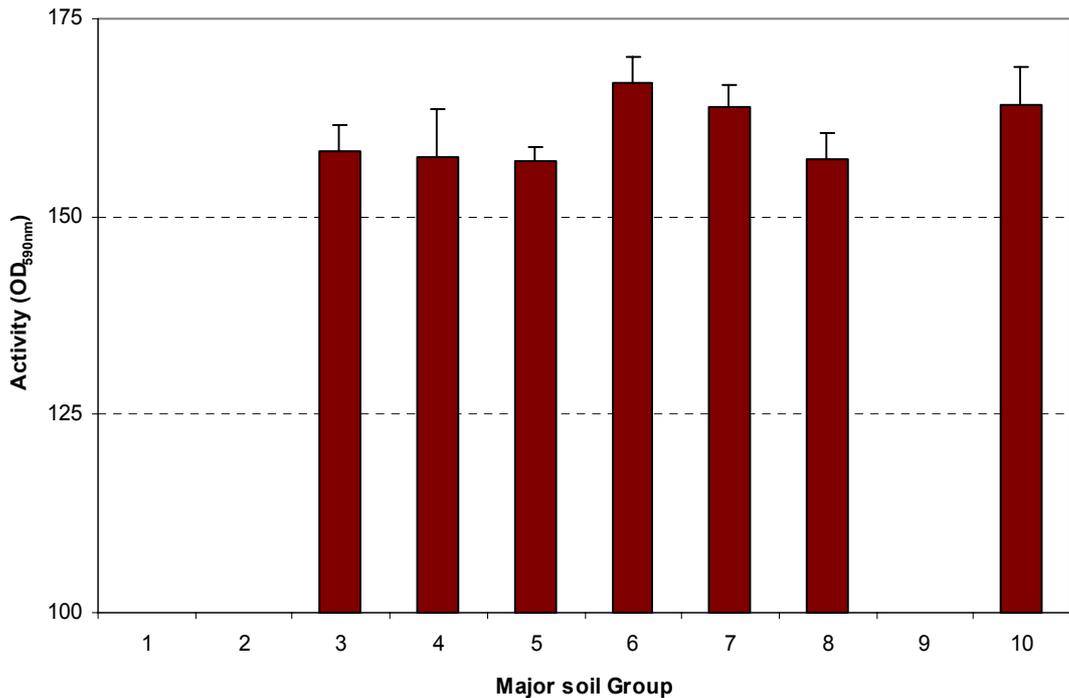
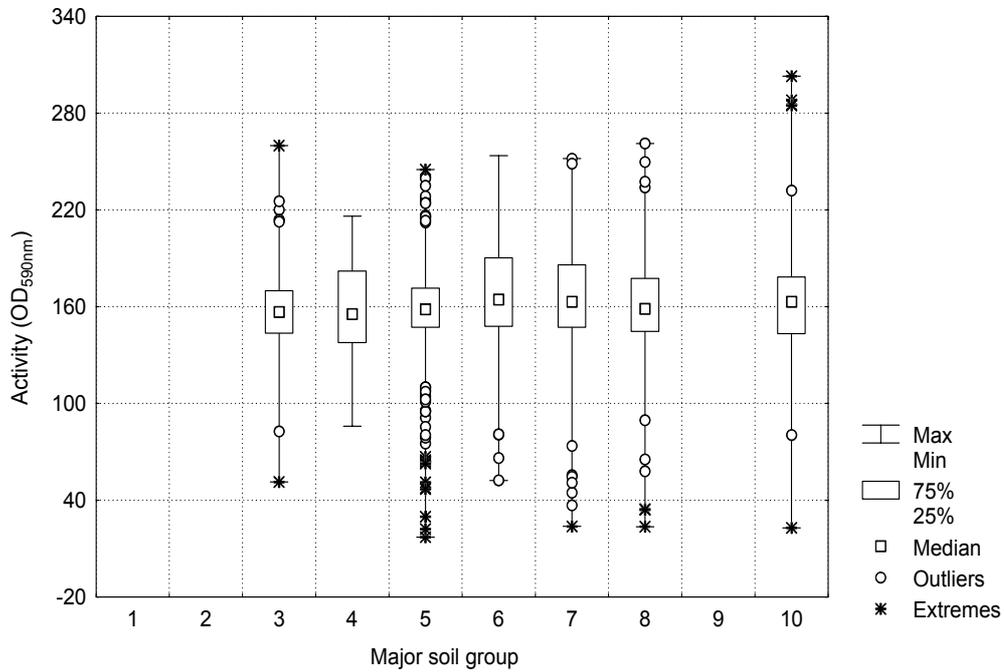


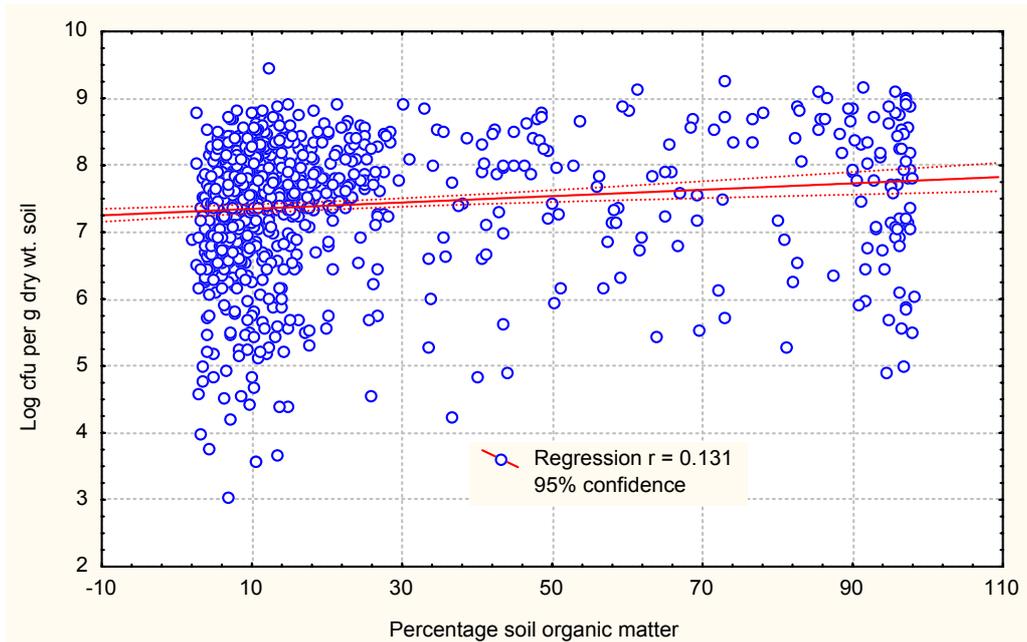
Figure 11.18. A box whisker plot showing the median level of global activity by Major Soil Group. Minimum, maximum, 25 and 75% quartile and outlying values are also shown.



### 11.3.7. Integrative Analyses

As soil organic matter provides the growth medium for the majority of soil microorganisms, we would expect there to be a relationship between SOM and numbers of viable bacterial cells. However, such a relation was not seen for the data set as a whole (Figure 11.9) where the majority of soil samples (83%) had an organic matter content below 30% as determined by the loss on ignition method. Similarly, soil pH and moisture can have an influence on the number and types of microorganisms present in soils; with fungal dominated systems at low pHs and more bacterial dominated systems at high pHs. However, regression correlation of the whole data set showed no relationship ( $r = 0.027$ ) between the numbers of viable bacterial cells and soil pH or soil moisture ( $r=0.165$ ). Such relationships may only hold for more restricted sample groupings, e.g., by vegetation or habitat, and more detailed analyses are thus necessary to determine the precise relationship, if any, between soil properties and microbial activity and abundance.

Figure 11.19. Relationship between numbers of viable bacterial cells per g dry wt. soil with soil organic matter content of samples.



#### 11.4. Conclusion

Over 940 soil samples were successfully analysed for numbers and functional diversity of heterotrophic bacteria. These data, along with all associated metadata, have been fully integrated into the Countryside Survey data management system that is maintained at CEH Merlewood. From here, microbial data can be linked to any other Countryside Survey data from the same 1 km square and/or the same X-plot.

The mean number of viable cells was  $2.51 \times 10^7$  cfu per g. dry wt. soil while the average global activities over all soil samples ranged from 17.1 to 302.9 ( $OD_{590}$ ). The data were skewed towards those with low numbers or activities. These non-normal distributions of the complete microbial datasets indicate that appropriate data transformations will be required before parametric analyses can be carried out.

The initial level of data analyses shows the range of values for viable bacterial cells activities for soil microbial communities extracted from a range of UK soils. This broad scale level of analysis did not allow us to see the subtle variations in the numbers and activities of soil microbial communities due to individual parameters e.g. soil type and vegetation and also differences in the response of microbial communities to certain types of substrates (e.g., sugars, carboxylic acids or amino acids). The data need to be examined in more detail. For example, the sample size and relatively even distribution of metabolic activity values for the microbial community in the complete dataset suggest that national range values could be established and used to place other samples (e.g., those from heavily polluted sites) into a broader context (c.f. Schipper and Sparling, 2000). The next stage will be to compare how published data fit into this distribution pattern. The potential to develop range values should be investigated for other microbial parameters using a similar approach.

The responses to the 95 BIOLOG GN substrates were analysed as a whole for the purpose of this report. Responses for each set of carbon sources should be analysed separately so that the discriminating power of different substrate groups can be evaluated. Further analyses of the BIOLOG data will also have to account for the correlation between global activities at low inoculum densities. Several different methods have been put forward in the literature for the data analysis of much smaller BIOLOG data sets.

## **12. OVERALL CONCLUSION**

### **12.1 Completion of MASQ**

The overall objective of the MASQ project was to produce good quality data for a range of soil properties from soil samples collected during Countryside Survey 2000. A unique aspect was the assessment of biological as well as chemical soil properties at the national scale to investigate the relationships between the two and also to assess the feasibility of incorporating biological assessments into large-scale soil monitoring programmes. In the latter context, the MASQ project focussed on microbial and invertebrate groups that could be sampled, extracted and characterised with relative ease while providing sufficient information to support detailed analyses. A key strength was seen to be the direct linkage between the soil data and other site factors since all data were obtained from within the same vegetation plots. As a result, the MASQ project has completed the single most comprehensive survey of soil biological and chemical properties to date.

The objective of producing good quality data has been met and a series of ORACLE based MASQ datasets have been established within The Countryside Survey 2000 Integrated Data System (CIDS). These datasets are fully compatible with the Countryside Information System (CIS). The MASQ datasets are managed by the Soil Ecology Section at CEH Merlewood under the auspices of Countryside Survey 2000 Module 13. MASQ data can be accessed in accordance with the data access policy agreed by the Countryside Survey 2000 Advisory Group.

The collection, processing and laboratory analysis of ca. 1200 soil samples for biological and chemical properties within a period of 20 months was a major achievement. This was the first time that a range of biological and chemical soil properties have been assessed simultaneously from a large number of sampling points at the national-scale. Detailed protocols were developed under the project, for sampling, analyses and data-management, and are available for future monitoring or national scale sampling programmes. Significant outputs have been the establishment of methods that meet rigorous QC standards. The validation exercises carried out in collaboration with the Environment Agency laboratories at Leeds, Nottingham and Llanelli prior to the analyses of the heavy metals and organic pollutants proved extremely valuable and the results will be published in the scientific literature.

The MASQ datasets now form a valuable resource that can be used as baseline data for future soil and other environmental monitoring programmes and a means to place specific site, region and country-scale issues within context e.g. regional, national, European and the wider international environment. Specifically, the MASQ data can now be used to support the development of national baseline datasets, inform the development of national soil sampling and monitoring programmes and investigate current and future issues regarding the assessment of soil quality in Great Britain. In particular, the potential for integrating soil biological properties into future soil assessments can be investigated since the feasibility of assessing soil biological properties in a national survey has now been established.

The next stage is to synthesis the relationships between the invertebrate and microbial properties of soils and other soil properties in the context of the wider environment e.g.

habitat, land use and geographical location. Research priorities to develop these issues are outlined in the relevant preceding sections and are summarised below.

## 12.2 Summarising results

The preceding sections provide an over-view of the individual biological and chemical properties obtained from the CS2000 soil sample and information on methods, quality control and the individual datasets. Summary statistics have been used to describe the datasets with analyses of each with respect to national scale stratifications used in CS2000 i.e. Environmental Zone, Broad Habitat, Major Soil Group and CVS Aggregate Vegetation Class. Thus illustrating the range and variability of individual soil properties within the British countryside, regions and with Broad Habitat, vegetation class and Major Soil Group.

Results from soil pH and soil organic matter contents from both 1978 and CS2000 follow expected patterns in the environment. The most acidic soils with highest soil organic matter contents were recorded in upland areas and in typical habitats generally associated with acid and organic rich soils i.e. heaths, bogs and moorlands, while soils with highest pH and lowest soil organic matter contents were found predominately in lowland agricultural habitats or calcareous and neutral grassland. Significance increases were detected in soil pH and SOM from 1978 to CS2000. **These results can only be substantiated once the variation between the 1978 and CS2000 datasets have been resolved, especially for SOM.** This is being done by cross-checking, on a sample-by-sample basis, the exact location of samples, land use changes and possible method differences from 1978 to CS2000.

The concentrations of heavy metals in the British countryside were generally lower than those reported previously by McGrath and Loveland (1992), perhaps reflecting the exclusion of urban and peri-urban areas from CS2000. There were trends in metal concentrations across the countryside with higher values, in general, being recorded in England and Wales compared to Scotland and in agricultural soils and habitats. The relationships between metal concentrations and soil organic matter contents were heavily influenced by the bi-modal distribution of soil organic matter in GB soils.

Preliminary data analyses indicated that the distribution of POPs in British soils was not homogeneous and that large variability can occur over relatively small distances (< 2 m). Significantly higher levels of PAHs and PCBs were detected in gley soils compared to brown soils. Concentration differences were also noted among habitats and vegetation classes. Further data analyses, in combination with chemical analyses of 120 soil samples, will, in the first instance, focus on understanding the distribution of POPs in the wider environment by investigating relationships with other soil properties and habitat characteristics that may influence the quality as well as quantity of POPs and the relationships with geographical location and proximity to sources.

Although there have been no equivalent large-scale geographical surveys, the numbers of major invertebrate taxa recorded from CS2000 soil samples can be considered typical of a soil invertebrate community, as recorded in various site-specific studies across Europe. Collembola and Acari are amongst the most common soil dwelling fauna, especially in semi-natural environments and the sampling and extraction methods used in this study would favour the collection of two groups (Collembola and Acari).

Therefore it is not surprising that the results confirm their pre-dominance at the national scale. It is, however, reassuring that, by using relatively simple sampling and extraction methods, these two groups of soil invertebrates were recorded in sufficient numbers to support further examination of distribution patterns and relationships with other soil properties and the wider environment.

The results analysed to date suggest that the least acidic, lowland agricultural and woodland soils tended to contain the most invertebrate taxa and number of Collembola while upland areas, with acidic (often podzolic) soils and vegetation associated with acidic soils, tended to have higher numbers of Acari and Oribatid mite species. A major highlight, in terms of biodiversity assessment, has been the identification of several new records to Britain and new species to science from the identification of Oribatid mite species. The prevalence of soil invertebrates, principally Collembola and Acari, in the CS2000 soil samples collected across the British countryside provides the basis to investigate the patterns and relationships of different levels of diversity to other soil properties and the wider environment.

The distribution patterns from the soil microbial assessments suggest that a national range for the number of viable bacterial cells and metabolic activities from soil samples could be established and used to place site-specific data. There was little difference in median numbers of culturable heterotrophic or global microbial activity, as assessed by BIOLOG, across the Environmental Zones. Trends in both culturable heterotrophic or global microbial activity were, however, evident between individual Broad Habitats, Land Classes, Aggregate Vegetation Classes and Major Soil Groups although large variation was observed within each category. Further analyses should determine whether this variation corresponds to land use, specific vegetation, location, sampling time etc. The BIOLOG dataset is an extensive resource offering many different options for investigative data analyses. The next stage must be to determine the importance of inoculum density on the global responses and to evaluate the discriminating power of the different substrate groups.

The results presented in this report have demonstrated that the MASQ datasets can be used to investigate the distribution patterns of, and relationships between, soil biological and chemical properties and their environment. The next stage will be to identify future research requirements that continue the development of appropriate methods and policy requirements for the monitoring and assessment of soil quality in Great Britain.

### **12.3 Future Research Priorities**

The following research priorities that have arisen from the analyses of the MASQ datasets. These are not seen as comprehensive list but rather a starting point from which research priorities that address research and policy requirements can be explored, identified and undertaken

- Examine changes in soil pH over time in the context of currently mapped critical loads for acidity and exceedance of these critical loads
- Examine changes in soil organic matter content in terms of controlling factors e.g. geographical location, land use and vegetation cover.

- Assessment impacts of changes in SOM and SOC on soil carbon stocks in the British countryside with respect to land use, management and location. This requires the analyses of a sub-set of CS2000 soil samples for total carbon contents to derive a suitable equation to convert SOM to SOC values.
- Apportion statistical significance to the variation recorded in heavy metal concentrations in soils and establish confidence limits to expected concentrations of heavy metals in soils across the British countryside.
- Establish the significance of environmental factors that may influence concentrations of heavy metals in soil such as geology, altitude, latitude and proximity to known emission sources.
- Establish how heavy metal concentrations in soils from CS2000 relate to regional or national data sources on heavy metal contents in soils and stream sediments.
- Examine the relationships between the concentrations and distribution patterns of POPs in the environment and data from the same sampling locations e.g. other soil properties and other CS2000 data such as vegetation and land use, and the size and location of previous and current sources of pollutants. This could include spatial correlation of PAH data with the size and location of sources, correlation of OC pesticide data with specific agricultural land uses such as cereals and sheep farming and studies of heterogeneity using the latest version of the Land Cover Map.
- Comparison of the OCPs and OCBS from the MASQ datasets with levels in birds of prey to determine whether there are any correlations between concentrations, compound patterns and geographical distribution to provide data on the transfer of POPs through food chains. Findings could inform studies of human as well as wildlife food chain.
- Test the global fractionation theory that the semi-volatile compounds move round the globe as a result of their physicochemical properties using MASQ datasets and related altitudinal and latitudinal gradients.
- Complete identification of Collembola species to provide a comprehensive dataset. Collembola is one group of soil invertebrates for which a great deal of information is available on distribution and occurrence at a site-specific level, often in relation to soil contamination.
- Investigate the relationships between the invertebrate occurrence and diversity with other soil properties and the wider environment. Firstly by examining variation and trends in invertebrates to determine whether outlier and extreme values correspond to other, more site-specific, environmental factors or higher-level spatial or temporal factors for example soil pH and organic matter content. Assess appropriate statistical analyses to determine the significance of the trends highlighted by these results. This should include examination of relationships between sample number, taxa occurrence and species richness

- Establish national range values for microbiological properties by apportioning significance to the variation recorded and comparing the MASQ data with published data fit. The potential to develop range values should be investigated for other microbial parameters using a similar approach.
- Responses for each set of carbon sources should be analysed separately so that the discriminating power of different substrate groups can be evaluated. Further analyses of the BIOLOG data will also have to account for the correlation between global activities at low inoculum densities.
- Assess the data analyses tools for interrogating the BIOLOG datasets. Several different methods have been put forward in the literature for the data analysis of much smaller BIOLOG data sets. The application of these to the CS2000 datasets before data analyses can commence in full; there have been, as yet, no equivalent analyses of such a large number of BIOLOG results.
- Investigate the relationships between the functional and taxonomic diversity of soil invertebrate and microbial biological properties in relation to the other soil properties and site-characteristics e.g. vegetation, soil type, location, land use and land use change. Specific questions that can be addressed from this are:
  - What are the characteristic soil biological properties across the range of vegetation types and soil types in the British countryside in soils with low levels of pollutants?
  - What is the discriminating power of different functional and taxonomic groups of soil biodiversity in relation to land use or land use change or levels of chemical and organic pollutants?
  - Is there any surrogacy between soil biological and chemical properties or between soil properties and vegetation?
- Examine the significance of small-scale heterogeneity (10 cm to 2 m up to 1 km) and large-scale spatial patterns? The Monkswood Wilderness study for the MASQ organic pollutants showed a large range in values over a small distance. This aspect could be examined, in the first instance, for all or any soil properties from MASQ by comparing variability between the five sample plots within each 1 km square and between each 1 km square.
- Examine the significance of short-term temporal variability versus single sample results and long-term change? For example, do seasonal differences in soil properties explain some of the variability reported since CS2000 was carried out over two years? This has most relevance for the soil biological properties but may also be important for the chemical properties. This can be investigated since the sampling and processing dates are available for all MASQ soil samples for date, season and year. An additional dataset is available from the Environmental Change Network terrestrial sites. This contains information on the bacterial counts, BIOLOG responses and the invertebrate communities, every two weeks over 6 months in 1998, using the same methods as CS2000.
- Analyses of stored CS2000 soil samples for other soil determinants.

- total C from a sub-set of stored samples to support conversion of SOM data to SOC
  - other soil chemical and physical properties to further support the investigative analyses into the relationships between soil biological properties and their environment e.g. bulk density, total carbon content, exchangeable cations/anions, total nitrogen content and texture
  - other potentially toxic organic compounds, with frequency of occurrence and an estimate made of their relative concentration.
  - biochemical properties to examine differences in bacterial and fungal communities (e.g. DNA, phospholipid fatty acids or ergosterol) by using remaining stored frozen CS2000 soil samples.
  
- Evaluation of methods used in MASQ project and other potential methods for assessing soil biological and chemical properties that could be used in future soil monitoring. This would require detailed consideration of the purpose of any soil data i.e. assessment of soil status versus soil function; long-term monitoring versus site-specific sampling. Methods that could be considered are those that:
  - determine availability of heavy metals in soils.
  - collect the more volatile POPs from soil samples
  - directly correlate with soil microbial diversity function e.g. rhizosphere substrates in a BIOLOG system that relate to soil microbial function and biochemical methods (e.g. DNA, phospholipid fatty acids or ergosterol).
  - extract the full complement of soil invertebrates e.g. automated extraction with flotation
  - enumerate soil invertebrates using digital imaging techniques
  - identify soil invertebrates using molecular techniques

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## List of Abbreviations

AAS	Atomic Adsorption Spectrophotometry
ASE	Accelerated Solvent Extraction
AVC	Aggregate Vegetation Class
BAP	Biodiversity Action Plan
BH	Broad Habitat
BIDS	Bath Information and Data Services
BRC	Biological Records Centre
Cd	Cadmium
CEH	Centre for Ecology and Hydrology
CEH MW	Centre for Ecology and Hydrology Merlewood
CFU	Colony Forming Units
CIDS	Countryside Information Data System
CIS	Countryside Information System
Cr	Chromium
CRM	Calcareous Reference Material
CS2000	Countryside Survey 2000
Cu	Copper
CVS	Countryside Vegetation System
DCM	Dichloromethane
DDE	Dichlorodiphenyldichloroethylene
DDT	Dichlorodiphenyltrichloroethane
DEFRA	Department for Environment, Food and Rural Affairs
EA	Environment Agency
EEC Directive	European Economic Community Directive
EZ	Environmental Zone
GC-ECD	Gas Chromatograph Electron Captor Detector
GC-MS	Gas Chromatograph Mass Spectrometer
GIS	Geographic Information System
H & S	Health and Safety
HCH	1,2,3,4,5,6-hexachlorocyclohexane
Hg	Mercury
HPLC	High Performance Liquid Chromatography
HTML	Hyper Text Markup Language
ICP-MS	Inductively Coupled Plasma-Mass Spectrometry
ICP-OES	Inductively Coupled Plasma-Optical Emission Spectrometers
ISE	International Soil Exchange
ITE	Institute of Terrestrial Ecology
IUPAC NO	<a href="#">International Union of Pure and Applied Chemistry</a> Number
LC	Land Class
LOD	Limit Of Detection
LOI	Loss On Ignition
MASQ	Monitoring and Assessing Soil Quality
MAX	Maximum
MEWAM	Methods for the Examination of Waters and Associated Materials
MIN	Minimum
MLURI	Macaulay Land Use Research Institute
MSG	Major Soil Group
MSG	Major Soil Groups

NERC	Natural Environment Research Council
Ni	Nickel
NLS	National Laboratory Service
NSI	National Soil Inventory
OC	Organochlorine
OCPs	Organochlorine Pesticides
OD	Optical Density
PAH	Polycyclic Aromatic Hydrocarbons
Pb	Lead
PCB	Polychlorinated Biphenyls
POPs	Persistent Organic Pollutants
QC	Quality Control
QSARs	Quantitative Structure Activity Relationships
RCEP	The Royal Commission on Environmental Pollution
RSSS	Representative Soil Sampling Scheme
SAS	SAS Institute Incorporated
SEC	Size Exclusion Chromatography
SEPA	Scottish Environmental Protection Agency
SETOC	Sediment Exchange for Tests on Organic Contaminants
SNIFFER	Scotland and Northern Ireland Forum For Environmental Research
SOM	Soil Organic Matter
SRM 1944	Standard Reference Material 1944
SSLRC	Soil Survey and land Research Centre
ST DEV	Standard Deviation
TIGER	Terrestrial Initiative in Global Environment Research
UNEP	United Nations Environment Programme
URGENT	Urban Regeneration and the Environment
UV	Ultra Violet
V	Vanadium
WWW	World Wide Web
Zn	Zinc

# Project Record on CD-ROM

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**Project Record Summary Tables**  
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*pHD 2000.xls*  
*pHF1978-2000.xls*  
*pHF2000.xls*

**SOILCARBON\_SUMMARY\_TABLES**

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*LDEC\_1978.xls*  
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**METALS\_SUMMARY\_TABLES**

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**INVERTS\_SUMMARY\_TABLES**

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*MITE\_SUMMARY\_TABLES.xls;*  
*TAXA\_SUMMARY\_TABLES.xls*

**MICROBES\_SUMMARY\_TABLES**

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