Environmental Change Research Centre

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Sampling and analytical protocols for the Open Air Laboratories (OPAL) Water Projects

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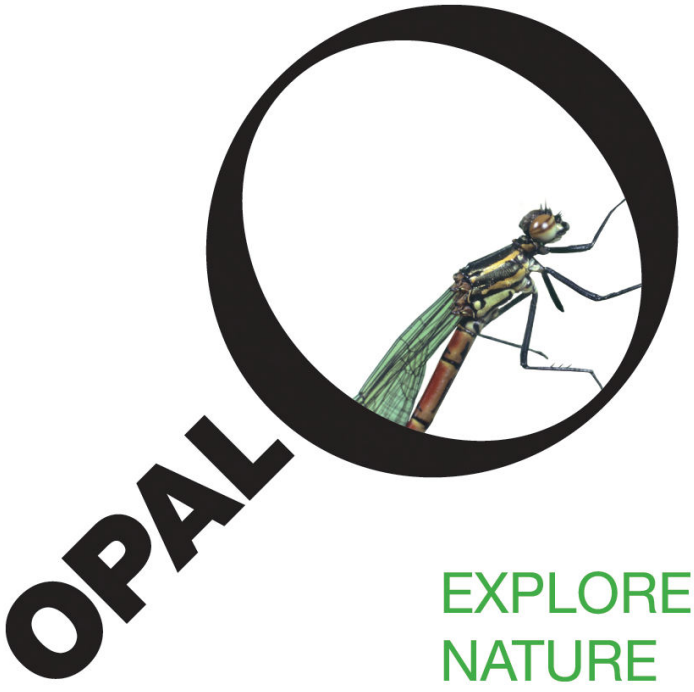

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Introduction

One of the main aims of the Open Air Laboratories (OPAL) project is to develop a greater understanding of the state of the natural environment and its biodiversity especially in the most impoverished parts of England. To this end, the project aims to increase national monitoring, providing new data on a range of sites in locations across the country; provide more information on disadvantaged areas and polluted environments and raise awareness of ecosystem health and how individual actions affect this.

As part of the OPAL National Water Centre's activities our aim is to set up a monitoring programme at a lake in each of the nine regions of England (Figure 1 and 2). This monitoring programme involves quarterly measurements over 4-5 years (up to the summer 2012) supplemented by other less frequent monitoring and a few 'one-off' sampling activities, mainly in the first year of the project. This report describes the field and laboratory protocols for the sampling and analytical procedures undertaken as part of the OPAL water projects.

Quarterly sampling

Every three months water samples will be taken for major chemistry (major cations, anions, nutrients and other determinands – see Appendix D), trace metals (mercury, lead, cadmium, copper, nickel and zinc), persistent organic pollutants (POPs; e.g. PAHs, PCBs and PBDEs) and suspended solids. We will also sample for zooplankton, phytoplankton and epiphytic diatoms and undertake physical measurements such as a Secchi disc reading (for water transparency) and temperature, conductivity, oxygen and light profiles.

Annual sampling

Annually we will undertake a macrophyte and invertebrate survey of each lake and take a few samples of these for trace metals analysis.

'One-off' sampling

i) Sediment coring. Two sediment cores will be taken from each lake. These will allow us to put the contemporary observations and monitoring into an historical context. One sediment core will be radiometrically dated and analysed (e.g. macrofossils; diatoms) to look for biological changes and to assess changes in the inputs of deposited pollutants (e.g. geochemistry, trace metals and fly-ash particles).

ii) Toxicity tests. Surface sediment samples will be taken for metals, POPs and fly-ash particle analysis but also for direct toxicity tests using larval midges (*Chironomus riparius*) and juvenile cladocerans (*Daphnia magna*). These results will be linked to levels of metals and POPs in these sediments and to similar data from previous studies (e.g. Rippey et al. 2008).

iii) Fish samples. We will collect 5-10 fish from each site from which the muscle will be analysed for POPs while the kidney, liver and muscle will be analysed for trace metals.

iv) Bathymetry. We will undertake a bathymetric survey of each lake using an echo sounder linked to a GPS. This will provide a basin morphology for each lake as well as lake volume, mean depths and hypsographic curves.

All field sampling is subject to a standard University College London Risk Assessment (Appendix A) while all laboratory procedures are covered by the Safety Protocols for the Institutes in which they occur.

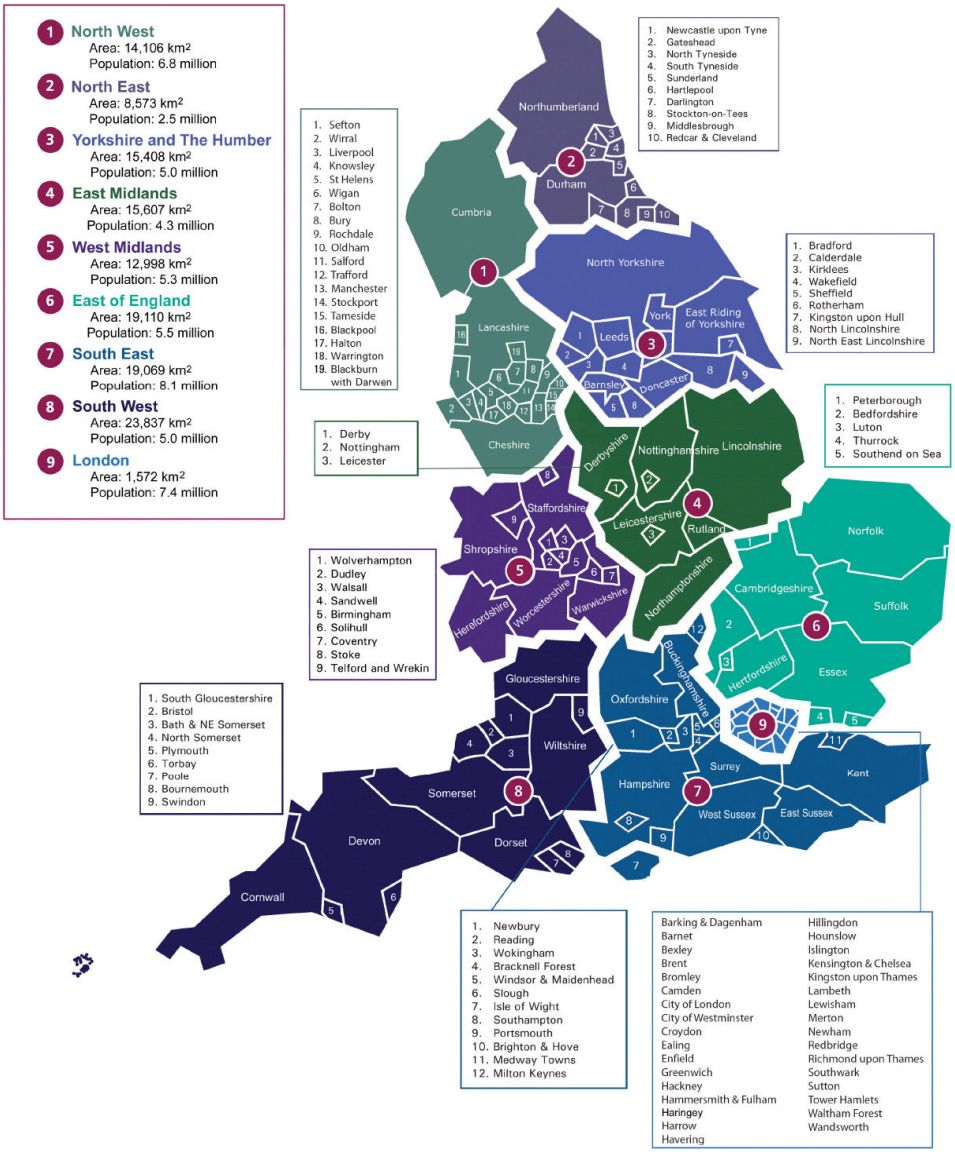

Figure 1. The nine OPAL regions of England

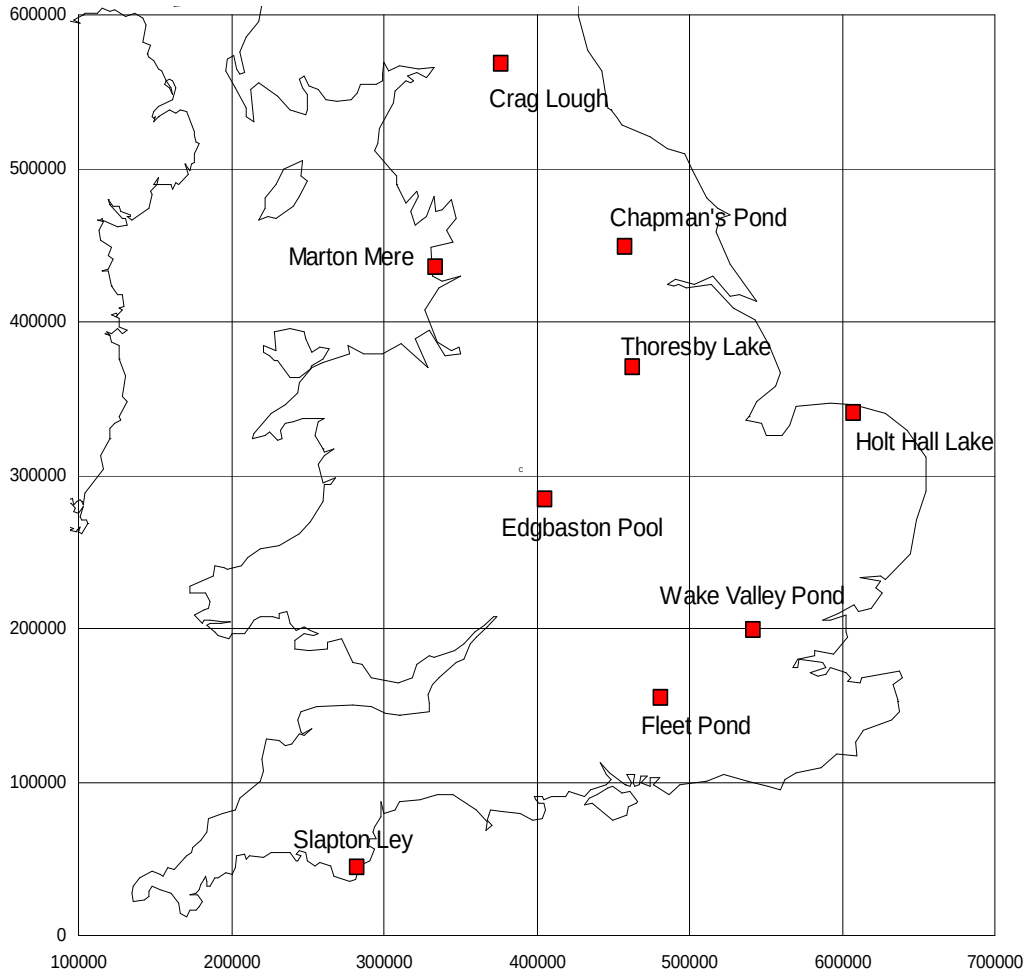

Figure 2. Locations of the nine selected OPAL sites

OPAL region	Site name	Nearest town	Grid Reference
North west	Marton Mere	Blackpool	SD 343 352
North east	Crag Lough	Haltwhistle	NY 765 680
Yorkshire and Humber	Chapman's Pond	York	SE 588 491
East Midlands	Thoresby Lake	Mansfield	SK 630 704
West Midlands	Edgbaston Pool	Birmingham	SP 055 842
East of England	Holt Hall Lake	Holt	TG 076 398
South east	Fleet Pond	Fleet	SU 820 550
South west	Slapton Ley	Slapton	SX 825 432
London	Wake valley Pond	Epping	TQ 421 987

CLEANING PROCEDURES FOR SAMPLE COLLECTION EQUIPMENT

1. Water Sampling

a) Water samples taken for analysis by the National Laboratory Service (Environment Agency). (Analytical Protocols Section 12; Appendix D) are collected in clean bottles supplied by the NLS.

b) Water samples for trace metals are collected in bottles (250ml; NALGENE ®) containing 2.5ml (1% vol) Aristar® grade nitric acid. Returned bottles after analysis are rinsed in distilled water, washed and soaked overnight in a 5% Tween solution, then rinsed in distilled water. The cleaned bottles are then soaked for 24hrs in a 4% HCl solution bath. On removal the bottles are rinsed again in distilled water and given a final rinse with de-ionised water before being dried in a static air drying cabinet. The Aristar® grade nitric acid is added to the bottles a few days before fieldwork to reduce degradation of the bottle interior.

c) Water samples for mercury are collected in bottles (125ml; Teflon) containing 1.25ml (1% vol) Aristar® grade hydrochloric acid. Sample bottles are cleaned and re-filled with hydrochloric acid prior to fieldwork by NILU (Norwegian Institute of Air Research) in Norway.

d) Water samples for persistent organic pollutants (POPs) are collected in 20L PVC carboys. After analysis, on return from the University of Birmingham, the unclean carboys are rinsed in distilled water, washed, scrubbed and left to soak for 24hrs in a 5% Tween solution. Cleaned carboys are then rinsed thoroughly with de-ionised water and dried in a static air drying cabinet.

e) The gusher pump and tubing used for water sampling are washed between sites with a 5% Tween solution and rinsed with distilled water. On return to UCL the pump and tubing are washed again with a 5% Tween solution and rinsed with distilled water. The whole apparatus is dried in a static air drying cabinet before storage.

2. Biological and other sampling

a) All bottles used for zooplankton, phytoplankton and suspended sediment are washed in a 5% Tween solution, rinsed in distilled water and dried.

b) The cleaning and washing of materials for POPs sampling of sediments and fish are outlined in the relevant sections.

FIELD SAMPLING PROTOCOLS

1. Sampling Location

All routine (quarterly) water and biological sampling takes place from a location towards the centre of each lake. These locations were fixed upon completion of the bathymetric surveys and are located in the deeper areas of the lakes in order to provide a maximum depth for the profile measurements (see Section 3). This sampling is undertaken from a small inflatable boat, rowed to the sampling location and secured by a small anchor. Other sampling (e.g. epiphytic diatoms; aquatic macrophytes; macro-invertebrates) is undertaken from around the shore-line at fixed points to allow repeat sampling at the same points. All locations are fixed by GPS and recorded on a site sheet completed for each sampling visit (Appendix B).

The transfer of species from one site to another is a real possibility when sites are visited sequentially within a short period of time. This is a particular concern when invasive species maybe transferred. To prevent this all OPAL sampling involving boat work will follow ECRC Invasive or Harmful Species Disinfection Protocol (Appendix C).

2. Water Sampling

Water samples for major chemistry, trace metals (including mercury), persistent organic pollutants (POPs) and suspended solids are taken from the central sampling location (Section 1; above). All samples are taken from 50 cm water depth by use of a manually operated pump with a hose lowered to the correct depth. The intake hose is covered with a mesh to prevent filamentous algae and other debris from entering the pump. Large volumes are required for the POPs sampling and so it is not feasible to fill these large containers at the appropriate depth by hand. Therefore, to ensure standardisation all water samples will be taken in this way. All sampling apparatus will be cleaned accordingly.

2.1. Suspended solids

The mass of suspended material in the water will be determined by collecting a known volume of water (usually 2 litres) and filtering this through a pre-weighed Whatman GF/C filter. This filter is then dried at 40 °C to constant weight and re-weighed. The mass of suspended solid per litre of lake water is then calculated from the mass difference.

3. Physical Measurements

All these measurements are taken from inflatable boat at the central sampling point.

3.1. Underwater Light Profile

Underwater light levels are measured in the lake water column with a submergible LI-COR LI-250 Light Meter. Light levels are measured at regular depth intervals (dependant on lake depth, usually every 0.5m) to the bottom of the lake. Light levels are measured as the probe is lowered and returned to the surface, with the mean value recorded. An above water (ambient) light level measurement is also recorded.

3.2. Secchi disc (water clarity)

A Secchi depth measurement is also used to assess light penetration in the water column. This involves lowering a quartered black and white metal disc attached to a tape measure. The disc is lowered into the water until it can no longer be seen. At this point the depth is recorded. The disc is then raised again until the disc becomes visible once more and this depth is also recorded. The 'Secchi depth' is the mean of the two recorded values.

3.3. Conductivity Profile

Conductivity (expressed in micro-Siemens; μS) of lake water is a measure of the resistance of a solution to electrical flow. Resistance declines with increasing ion content (e.g. salinity). Conductivity measurements are commonly used to assess water quality changes (e.g. seasonal variation) and characterise lakes due to catchment properties. Conductivity is measured using a Hach HQ 30D conductivity probe on a cable at regular depth intervals (dependant on lake depth, usually every 0.5m) from the surface to the sediment/water interface at the bottom of the lake.

3.4. Oxygen Profile

Oxygen dissolved in lake water is essential to the metabolism of aerobic aquatic organisms. Levels of dissolved oxygen in the water reflect inputs from both the atmosphere and photosynthesis, which allows an approximate evaluation of biological processes occurring in the lake. Dissolved oxygen concentrations are measured using the Hach HQ 30D DO probe at regular depth intervals (dependent on lake depth, usually every 0.5m) from the surface to the bottom of the lake.

3.5. Temperature Profile

Water temperatures are measured at regular depth intervals (dependent on lake depth, usually every 0.5m) from the surface to the bottom of the lake. Water temperature is measured at the same time by the probes measuring conductivity and dissolved oxygen. Air temperature is also measured with a thermometer during lake water sampling.

4. Bathymetric Survey

A detailed bathymetric survey was carried out at each site using a combined echo sounder and global positioning system (GPS). The system is mounted on a small inflatable boat with combined soundings and GPS fixes taken every second as the boat traverses the lake. The boat is propelled with either a low speed electric motor or oars. Oars are nonetheless always used at lake margins and areas of aquatic plants to avoid disturbance. This approach results in a large number of readings for the lake basin from which a bathymetry can then be calculated. A geographic information system (GIS) is used to store and manipulate data and interpolation is used to construct a gridded depth 'surface'. The resolution of this grid is dependant upon the size of the lake but is usually around 10m. Lake volumes, mean depths and hypsographic curves (showing cumulative volume or lake area against depth) can then be calculated from the bathymetric data.

We also aim to record lake level changes at each site. Where these data are already recorded (e.g. where stageboards are present readings are taken at each visit and we will encourage visitors to do this too. If a stageboard is not present at a site we will investigate the feasibility of installing one or at least a marker which we can use to take readings over the course of the OPAL project. Again we will encourage visitors to take additional interim readings. Any lake level device installed will be levelled to Ordnance Datum.

5. Sediment Coring

Two sediment cores from each site were extracted using a Livingstone-type piston corer (Livingstone, 1955). The location of the cores are dependant on the nature of the lake and its known history (dredged areas, areas of know disturbance). Cores taken for palaeolimnological analyses are usually extracted from the deepest part of the lake, where sediment accumulation and optimal conditions for preservation are greatest. Cores from littoral areas are used for macrofossil analysis. Livingstone-type piston cores consist

of an in-line piston and collection chamber operated and connected to the lake surface by solid metal rods. A small, stable platform usually consisting of two small inflatable boats strapped together is anchored above the selected core site during core retrieval. Cores were extruded vertically at the field site in 1cm intervals. Samples are stored, cool in sealed plastic bags and returned to the laboratory for analysis.

6. Biological sampling

6.1. Invertebrates

Benthic macroinvertebrate samples are collected once each year. A semi-quantitative timed (2 minute) net sweep is carried out on three typical habitats at each site focussing on emergent and submerged vegetation. These samples are placed in a plastic bucket and preserved with Industrial Methylated Spirits (IMS). Sorting of the sample may also take place on site should time permit. Otherwise this is undertaken back at the laboratory.

6.2. Diatoms

At some lake types it has been standard to collect benthic diatoms from the surface of stones (epilithon). However, the waterbodies that form part of the OPAL study do not in all cases have an appropriate stony littoral, making the standardised collection of epilithon impossible. As a result, benthic diatoms are sampled quarterly at all sites from the base of an emergent plant. Where present the common (or Norfolk) reed (*Phragmites australis*) will be sampled. In the event that this is absent alternative emergent vegetation such as *Schoenoplectus lactustris* or *Carex* spp. is sampled. Dead stems are avoided. A sample of living reed from the outer edge of the reed bed, so as to avoid the effects of shading, is cut and the uppermost submerged section of plant collected. Three such sections of reed are collected, placed in a Sterilin tube and preserved.

6.3. Phytoplankton

Phytoplankton is collected from the same central point of the lake at which the water chemistry samples are collected. A flexible 2 m long tube with a weight attached to the bottom is lowered into the water. The top is then sealed with a bung and the tube withdrawn. The contents of the tube is emptied into a large bottle and preserved with Lugols Iodine. Thus, a large proportion of the photic zone will be sampled and any vertical heterogeneity in the phytoplankton community accounted for. Once preserved the phytoplankton is settled out overnight and the supernatant decanted thereby reducing the volume of the sample for storage prior to analysis.

6.4. Zooplankton

Zooplankton is sampled quarterly at each site in two ways: (i) open water and (ii) the littoral. For the open water sample a quantitative estimate of zooplankton abundance is obtained by tube sampling. A 2.5 m plastic tube (internal diameter ≈ 7.5 cm) is used to take a sample of the entire water column. Five tube samples are taken along a visual transect from at, or near, the centre of the lake towards the margins. Samples are then pooled and filtered through a 50 μm net. This will provide a quantitative estimate of zooplankton abundance. Start and end points of each transect are recorded by GPS so that sampling is undertaken in approximately the same regions of the lake each time.

For the littoral sample, the tube sampler is used to sample the shallower water of the lake margins and is carried out in three distinct locations. The tube will be placed at an angle covering the entire water column and collecting the greatest volume of water possible. The samples are then passed through a 50 μm net and pooled. The samples are then preserved with IMS.

6.5. Fish

As the sampling of fish in OPAL is not an attempt to provide a quantitative estimate of fish density or biomass the sampling methods are fairly simple. Fyke nets, fitted with otter guards to prevent the capture of otters, were deployed prior to dusk and left overnight. Fishing with rod and line was used to supplement the fyke netting. Fishing effort was concentrated around dawn and dusk as this is when fish are most active, although it was not limited to this period. Five to ten fish caught from each site above the appropriate size / weight limit were measured, weighed, sexed and a scale sample collected for aging. Samples of fish muscle were taken and frozen on site for the analysis of POPs. Samples of fish kidney, liver and muscle were also collected on site and frozen for subsequent trace metals analysis. Fish stomach contents were also recorded and, where appropriate, stored for metals analysis. All fish collection information will be recorded on site using the Fish Sampling Sheet (See Appendix E).

6.6. Submerged macrophytes

Macrophytes will be surveyed using the common standard methodology developed for the surveying of submerged vegetation by the conservation agencies (JNCC, 2005). A number of 100 metre sections per site are chosen, and at each of these sections both a wader survey and a boat survey are conducted. The wader survey consists of five transects perpendicular to the shore at 20 metre intervals along the 100 metre section of shoreline. On each transect macrophyte composition and abundance is measured at 25, 50, 75 and > 75cm. Further, at 50 m along the 100 metre shoreline section, a boat survey will be conducted where up to 12 points are sampled between c. 80 cm depth and the maximum depth of plant growth, or the centre of the lake. Species will be identified on site and voucher specimens of selected taxa (e.g. fine

leaved *Potamogeton* species and *Chara* species) will be collected where required in order to confirm identification.

6.7. Other biological organisms

Biological organisms additionally collected in the fyke nets were opportunistically sampled for metals analysis thereby providing additional information on metals within the food-web of these sites. In particular, the signal crayfish (*Pacifastacus leniusculus*) was caught in large numbers at Fleet Pond. In Britain, this species represents one of the biggest threats to the native crayfish and was introduced into England and Wales in the late 1970s and early 1980s for farming purposes. Subsequently, however, it escaped from many farm sites into which it was introduced and rapidly colonised. Not only is this species a competitor to the native crayfish, but it also carries a fungal disease, the crayfish plague, to which the native crayfish has no defence (http://www.defra.gov.uk/fish/freshwater/crayfish.htm).

7. Sampling of biota for trace metal analysis

Aquatic plants for trace metal analysis were collected from around the lake margins. Species selection was driven by presence at the site but the same species was used at all sites where possible. Plastic gloves are worn during sample collection and the samples stored in re-sealable plastic bags. The sampled vegetation is rinsed with deionised water, stored cool and freeze-dried prior to analysis.

8. Sampling for toxicological studies

The protocol for the toxicological studies follows that described in Rippey et al. (2008). Seven sediment cores were taken from the profundal area of each lake during using a gravity corer fitted with a perspex tube of internal diameter of 74 mm. Cores were retrieved from within as small an area as possible. The 0-5 cm section of each core was extruded vertically in the field, amalgamated and homogenised in a hexane-washed glass container. The core tube and all utensils in contact with the sediment were hexane washed and kept contaminant-free until use by wrapping in hexane-washed aluminium foil. The 0-5 cm sediment layer is likely to be the most relevant to benthic invertebrates as the 0-3 cm layer was used by Dave (1992) and Long et al. (1998) and the 0-6 cm layer by Besser et al. (1996).

Approximately 100 g of the homogenised wet sediment was transferred into hexane-washed amber jars. This sub-sample was frozen and freeze-dried before analysis for persistent organic pollutants (POPs – see Analytical Protocols Section 5). A second sub-sample of approximately 10 g wet sediment was stored in sealable plastic bags, freeze-dried and analysed for organic content (Dean 1974) (Analytical Protocols

Section 1) and for trace metals (Analytical Protocols Section 4.2). A third sub-sample of approximately 2 g wet sediment was stored in sealable plastic bags and air-dried prior to the analysis of spheroidal carbonaceous particles (SCPs; Analytical Protocols Section .3). The remainder of the sediment sample was then used for the sediment toxicity tests. The sediment samples were stored in polypropylene bottles at 4 ºC before use. The maximum holding time prior to analysis is 8 weeks.

ANALYTICAL PROTOCOLS

1. Lithostratigraphic analysis of sediments

1.1. Visual inspection

Significant changes are noted following the extraction of the core as the core tubes are made of transparent plastic. These changes with depth are checked during extrusion and sampling of the cores. The colour of the sediment samples are described using a Munsell colour chart and their basic composition described using Troels-Smith notation (Troels-Smith, 1955).

1.2. Water content and density

Sediment samples from aquatic environments have variable water contents. Recently deposited sediments have a higher water content as they have not been compacted by burial. Similarly, the water content of samples in a stratigraphic sequence varies with the porosity and permeability of clastic sediments. Water content in sediments is measured by the loss of mass from a weighed sample of wet sediment after heating (105ºC for 24hrs). Sediment density is calculated by measuring the mass of a known volume (2 cm^3) of sediment taken from a core sample.

1.3. Organic and carbonate content

Measurement of the organic/inorganic content of sediments is highly valuable for interpreting recent depositional changes and contemporary sediment/biogeochemical processes. A rapid and reliable assessment of organic content is achieved by the combustion at 550 ºC of a known mass of dried sediment. This 'loss-on-ignition' takes place in a muffle furnace over a period of 2 hours (cf. Dean, 1974; Heiri *et al.* 2001). The amount of organic matter in a sample is expressed as a percentage of the dried sediment. Further heating (950 ºC for 2 hours) and combustion is similarly used as a proxy measurement of carbonate content (Heiri *et al.* 2001).

2. Radionuclide dating of sediment cores

Lead-210 (half-life 22.3 years) is a naturally produced radionuclide, derived from atmospheric fallout (termed unsupported ^{210}Pb). Cesium-137 (half-life 30 years) and ^{241}Am (half-life 432 years) are artificially produced radionuclides, introduced by atmospheric fallout from nuclear weapons testing. ^{137}Cs was

additionally introduced into the atmosphere by the Chernobyl reactor fire in Ukraine in 1986. and nuclear reactor accidents. Sediment samples from each core were analysed for ^{210}Pb, ^{226}Ra, ^{137}Cs and ^{241}Am by direct gamma assay in the Bloomsbury Environment Institute at University College London, using an ORTEC HPGe GWL series well-type coaxial low background intrinsic germanium detector. Lead-210 is determined via its gamma emissions at 46.5keV, and ^{226}Ra by the 295keV and 352keV gamma rays emitted by its daughter isotope ^{214}Pb following three weeks storage in sealed containers to allow radioactive equilibration. Cesium-137 and ^{241}Am are measured by their emissions at 662keV and 59.5keV respectively. The absolute efficiencies of the detector are determined using calibrated sources and sediment samples of known activity. Corrections are made for the effect of self absorption of low energy gamma rays within the sample.

Supported ^{210}Pb activity is assumed to be equal to the measured ^{226}Ra activity and unsupported ^{210}Pb activity is calculated by subtracting supported ^{210}Pb from the measured total ^{210}Pb activity. ^{210}Pb radiometric dates are calculated using the CRS (constant rate of supply) and CIC (constant initial concentration) dating models (Appleby and Oldfield, 1978) and validated using dates determined from the ^{137}Cs and ^{241}Am stratigraphic records. Definitive chronologies based on an assessment of all the data were determined using the methods described in Appleby and Oldfield (1983) and Appleby (2001).

3. Spheroidal carbonaceous particle (SCP) analysis of sediments

Spheroidal carbonaeous particles (SCPs) are a component of fly-ash, the particulate by-product of high temperature fossil-fuel combsution. They have no natural sources and therefore provide an unambiguous indicator of atmospherically deposited contamination from these sources. Analysis for SCPs in sediment material follows the standard method set out in Rose (1994) whereby a sequential mineral acid attack is employed to remove unwanted sediment fractions. Nitric, hydrofluoric and hydrochloric acids are employed to remove organic material, silicates and carbonates respectively. A known aliquot of the resulting suspension, mainly carbonaceous material in water, is evaporated onto a microscope coverslip, mounted and the SCPs enumerated under a light microscope at 400x magnification. SCP concentrations are expressed as 'number of particles per gram dry mass' (or gDM^{-1}). The technique has a detection limit of 50 – 80 gDM^{-1} and a mean recovery rate of > 95%. Reproducibility is good, with the 95% confidence interval being less than 10% of the mean (based on n = 15 samples) (Rose 1990; 1994). A SCP reference sediment (Rose, 2008) and analytical blanks are included with every sediment digestion.

4. Trace metal analysis

4.1. Waters

Cd, Pb, Zn, Cu and Ni in lake waters are measured by inductively coupled plasma mass spectrometry (ICP-MS). Standard reference samples e.g. Standard Reference Material® 1640, and acidified blanks are analysed frequently (typically every fifth sample) for QA/QC purposes. Recent typical detection limits for these analyses are as follows (all values in $\mu g\ L^{-1}$):

Cu	Ni	Cd	Pb	Zn
0.18	0.11	0.02	0.01	0.20

All Hg analyses on water samples are analysed by CV-AFS at NILU (Norwegian Institute of Air Research) in Norway. Samples are stored in the dark at +5°C for a maximum of 3 months prior to analysis. BrCl is used to convert stable mercury forms to water soluble species, which in turn are reduced to Hg° with $SnCl_2$. Hg analysis is performed using a Tekran 2600 CV-AFS, where the Hg° is concentrated on a gold trap before being detected in the AFS detector. The detection limits are 0.5 ng Hg L^{-1}. The procedure (NILU-U-60) for sampling of precipitation followed by determination of total mercury using CV-AFS is accredited by Norwegian Accreditation in accordance with ISO IEC-17025.

4.2 Sediment and biological material

Initial screening of sediment core geochemistry was undertaken by X-ray fluorescence (XRF) using a Spectro XLAB 2000 in the Department of Geography, University College London. Extraction for more detailed analysis follows the following methodology. Sediment samples are treated with 8 ml concentrated Aristar HNO_3 and heated at 100°C for 1 hour in rigorously acid leached 50 ml Teflon beakers. For Hg measurements, after digestion, the supernates are carefully transferred into polyethylene tubes. The residue in the beakers is then washed with deionised distilled water and the supernates transferred into the same tubes. Pb, Cd, Zn, Cu and Ni are measured using atomic absorption spectroscopy (AAS) whilst Hg is measured by cold vapour atomic absorption spectrometry (CV-AAS) following reduction of Hg in the digested sample to its elemental state by 2 ml fresh $SnCl_2$ (10% in 20% (v/v) HCl). Certified standard reference materials (e.g. Buffalo River sediment SRM2704, Stream sediment GBW07305) are included in all digestion and analytical procedures. For AAS, reference materials and sample blanks are analysed every 20 samples. Biological samples (aquatic biota, terrestrial mosses) for trace metals analysis are extracted and analysed as for sediments.

5. Persistent organic pollutants (POPs)

The concentrations of selected chemicals from three groups of POPs were measured. These include seven polychlorinated biphenyl congeners (PCBs), 28, 52, 101, 118, 138, 153 and 180; fifteen polycyclic aromatic hydrocarbons (PAHs), acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benz[*a*]anthracene, chrysene, benzo[*b*]fluoranthene, benzo[*j*+*k*]fluoranthene, benzo[*a*]pyrene, indeno[1,2,3-*cd*]pyrene, dibenz[*ah*+*ac*]anthracene, benzo[*ghi*]perylene; and five polybrominated diphenyl ether congeners (PBDEs), 47, 99, 100, 153 and 154.

The extraction and purification methods employed for the determination of PCBs, PAHs and PBDEs in sediments are based on those of Rippey et al (2008), while the determination of the same analytes in fish flesh and water are based on those of Ayris et al (1997), Harrad and Smith (1998), and Harrad et al (2004). Briefly, accurately weighed 10 g aliquots of freeze-dried sediment (1 g of freeze-dried fish flesh) are treated with known quantities of internal standards (PCB congeners 34, 62, 119, 131, and 173, deuterated PAHs (acenaphthylene, phenanthrene, fluoranthene, benzo[*a*]pyrene, and benzo[*ghi*]perylene) and $^{13}C_{12}$-PBDEs congeners 47, 99, and 153) and extracted with hexane in an accelerated solvent extractor (ASE, Dionex, UK). For water samples, the entire sample (40 L) is gravity filtered through a filter paper (Whatman's pore size 1 μm) followed by a pre-extracted polyurethane foam (PUF) plug. Following filtration, the filter paper and PUF plug are combined and extracted in a soxhlet for 8 h with hexane. In each case, after cooling, the crude extract is concentrated to approximately 40 ml; 10 mL is reserved for PAH analysis, while the remainder is used for the determination of PCBs and PBDEs.

The PCBs and PBDEs fraction is eluted through a 20 g florisil column with 200 ml of CH_2Cl_2. The eluate is reduced to 2 ml hexane in a Kuderna Danish apparatus, washed with 2 ml conc. H_2SO_4, before elution through a column of florisil and $AgNO_3$-impregnated alumina (2 g of each) with 100 ml CH_2Cl_2 to remove residual sulphur. The entire eluate is then concentrated to 50 μL in nonane containing the recovery determination standards PCB 29 and 129 ready for GC-MS analysis. The PAH fraction is eluted through a florisil column (2 g) with 30 ml CH_2Cl_2 and concentrated to a final extract volume of between 200 μL to 2 ml CH_2Cl_2 containing p-terphenyl as a recovery determination standard (the exact volume depends on the anticipated concentrations in a given sample). GC-MS analysis of all analytes is conducted on a Fisons MD-800 instrument operated at unit mass resolution in EI selected ion monitoring mode, and fitted with a 60 m SGE BP-5 capillary column (0.2 mm x 0.25 μm). In all cases, 1 μL of the final sample extract is injected in splitless mode. The limit of detection for individual PCBs, PBDEs and PAHs is 0.1-0.2, 0.2-0.4, and 1 μg kg^{-1} dry weight, respectively. Method accuracy and reproducibility is evaluated by on-going replicate analyses of appropriate certified or standard reference materials.

6. Sediment macrofossil analysis

In the absence of reliable historical information on past aquatic macrophyte communities, analysis of sedimentary macro-remains of plants (the seeds, fruits and remains of stems, leaves and rhizomes) may provide a means of determining changes in the aquatic flora of a site (Birks 1980). Recent work has indicated that plant macrofossils provide a reliable means for tracking shifts in the dominant components of the submerged aquatic flora in shallow lakes (Davidson et al. 2005).

Typically, at least 15 levels from cores are examined for macrofossils. Samples are sieved at 350 and 125 μm, the exact sample volume being measured by water displacement. The entire residue on the 350 μm sieve is examined under a stereo-microscope at magnifications of x10 to x40 and plant and animal macrofossils enumerated. A quantitative sub-sample, approximately one tenth of the sample from the 125 μm sieve, is also analysed for smaller remains, such as leaf spines. All material is identified by comparison to reference material.

7. Diatom analysis

Analysis of diatoms (*Bacillariophyceae*: unicellular, siliceous algae) in sediment cores is a useful means with which to infer environmental trends. Diatoms are particularly good indicators of past limnological conditions, for example pH, nutrient concentrations and salinity, and can also be used for inferring changes in lake habitats. At least 15 levels from each of the cores are prepared and analysed for diatoms using standard techniques (Battarbee et al. 2001). A minimum of 300 diatom valves are counted on each slide at 1000x magnification and diatom concentrations were estimated using the microsphere method (Battarbee and Kneen 1982). Diatom species results are expressed as percentages of the total diatom sum. Contemporary diatoms from epiphytic samples are treated and enumerated in the same way.

8. Invertebrate analysis (contemporary)

Invertebrate sorting will take place in the laboratoty where all animals will be picked and identified to the highest practicable taxonomic level. Identification will use a variety of keys and guides (e.g. Croft 1986; Friday 1988) and will use a stereo-microscope at of x10 to x40 and a compound microscope up to x400 for body parts where this is necessary. The data produced will be estimates of abundance for each species/taxa found and though more complete species lists could be generated using other approaches this technique standardises effort to allow comparison between years. Additional expert identification may be available for some groups.

9. Zooplankton analysis (contemporary)

Zooplankton are enumerated using a Sedgewick-Rafter counting cell on a compound microscope at x 40 to x100 magnification. A minimum of 100 will be identified from a sub-sample of known volume. Cladocera will be identified with reference to Flössner (1972) and Alonso (1996). All individuals will be identified to species where possible. Species-level identification for some Daphnids is difficult and some guilds of *Daphnia hyalina* agg. may be used where species level identification in that group is not possible. Cyclopoid crustaceans will be assigned to only family level i.e. Calanoid or Cyclopoid. The data produced will be quantitative estimates of zooplankton abundance.

10. Phytoplankton analysis

Phytoplantkon are analysed using an inverted microscope. A sub-sample of known volume is taken from the well-mixed whole sample and placed in a counting chamber. The sample is then given at least four hours to settle and the phytoplankton enumerated at a number of magnifications:

- a low magnification (e.g. x 40 or x100). This is a whole chamber count to pick up large taxa, followed by;
- transect counts at an intermediate magnification (x250), which enumerate "intermediate-sized" taxa i.e. those that are too small for the low-magnification count but too large to be reasonably counted using fields of view at high magnification, followed by;
- a high magnification count (x400 or greater) using fields of view. This identifies the small taxa. Approximately 100 fields of view are counted (i.e. about 400 units assuming the recommended sample concentration).

This approach allows the generation of a quantitative estimate of phytoplankton diversity and abundance in a standardised way so as to allow comparison between samples.

11. Sediment toxicity tests

Toxicity tests for the sediments follow Rippey et al. (2008). Two sediment toxicity tests are undertaken. A 10-day chironomid survival and growth test and a 7-day cladoceran survival and reproduction test (ASTM 2000; US EPA 2000).

In the chironomid test, eight replicates of a reference sediment and five for each sediment sample are used each with 10 organisms per replicate chamber. All sediment and control samples are observed

for the number of adults emerging from the sediment. The number of live and dead animals in each test chamber is enumerated at test termination by sieving the sediment through a No. 35 (500 μm) sieve. The criterion for test acceptability is mean survival in the reference sediment ≥70%.

In the cladoceran test, ten replicates of reference sediment and sediment samples are used with one juvenile added to each replicate beaker. All beakers are monitored daily for survival of *Daphnia* and for production of young. The criteria for test acceptability is mean survival of ≥80%, an average of 20 or more young per surviving female in the reference sediment and production of at least three broods in 60% of the surviving organisms.

In both tests, the hardness of the water used is 160 to 180 mg/L as $CaCO_3$ while the reference sediment is a natural sediment collected from a small pond in Columbia, Missouri, a sandy loam (74% sand, 20% silt, and 6% clay) with an organic carbon content of 0.2 to 0.7%. The test water is monitored for temperature, dissolved oxygen, pH, conductivity, total ammonia, alkalinity and hardness. The test chambers are maintained in a temperature-controlled water bath adjusted to maintain a sediment temperature of 23 ± 1 ºC for chironomids and 25 ± 1ºC for cladocerans, each under a 16 hour light: 8 hour dark photoperiod. The statistical significance of animal survival is determined by hypothesis testing (p = 0.05). Analysis of each end-point between samples is evaluated by first analysing the data for normality and homogeneity of variance with Shapiro-Wilk's Test and either Bartlett's, Hartley's, or Lavene's Test, before comparison of means. Analysis of variance is utilised for the reproduction data along with Dunnett's procedure for comparing the means. Survival data are analysed using Fisher's Exact test.

12. Major water chemistry

All routine water chemistry analysis is undertaken by the National Laboratory Service (Environment Agency). The water analyses, methods and minimum reporting values (MRV) for each analysis is given in the following Table. More details are given in Appendix D.

Analyte	Method	MRV	Units
Conductivity at 20°C	Electrometric	10.0	μS cm^{-1}
pH	Electrometric	0.05	pH Units
Nitrogen: Total as N	Automated colourimetric	0.2	mg L^{-1}
Nitrogen: Total Oxidised as N	Automated colourimetric	0.005	mg L^{-1}
Nitrogen: Organic as N	Calculation	0	mg L^{-1}
Nitrite as N	Automated colourimetric	0.001	mg L^{-1}

Nitrate as N	Calculation	0	mg L^{-1}
Dissolved Organic Carbon (DOC) as C	Automated colourimetric	0.2	mg L^{-1}
Phosphorus: Total as P	Colourimetry	0.003	mg L^{-1}
Orthophosphate, reactive as P	Automated colourimetric	0.001	mg L^{-1}
Alkalinity to pH 4.5 as $CaCO_3$	Automated colourimetric	5	mg L^{-1}
Chloride	Automated colourimetric	1	mg L^{-1}
Chlorophyll	Acetone extract / UV Spectrophotometry	0.5	μg L^{-1}
Aluminium: Total	Inductively Coupled Plasma Mass Spectometry.	10	μg L^{-1}
Aluminium: non-labile, dissolved	UV/Visible Spectrophotometry	0.004	mg L^{-1}
Calcium	Optical emission spectroscopy	1.0	mg L^{-1}
Magnesium	Optical emission spectroscopy	0.3	mg L^{-1}
Potassium	Optical emission spectroscopy	0.1	mg L^{-1}
Sodium	Optical emission spectroscopy	2.0	mg L^{-1}
Sulphate as SO_4	Optical emission spectroscopy	1.0	mg L^{-1}

13. Lead isotopes

Pb isotopes in sediment material is measured by inductively coupled plasma-mass spectrometry (ICP-MS). Lead concentrations in the samples are measured by quadrupole ICP-MS (Agilent 7500c), with calibration against a synthetic Pb standard. The isotopes ^{206}Pb and ^{207}Pb are measured using a high resolution magnetic sector single collector ICP-MS (Axiom). In order to quantify precision, ten replicate analyses are performed for each sample. A solution of the National Institute of Standards and Technology (NIST) common Pb isotopic reference standard "Standard reference material 981" (natural Pb; $^{206}Pb/^{207}Pb$ = 1.0933) was measured after every three samples for calibration and mass bias correction.

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APPENDIX A.

UCL DEPARTMENT OF GEOGRAPHY
Fieldwork Risk Assessment

FIELDWORK RISK ASSESSMENT

Project title:	OPAL – Open Air Laboratories (Aquatic Monitoring and Water Research Centre)
Location(s):	9 Water bodies in England (Detailed site information available)
Project Supervisor:	Dr. Neil Rose
Brief description of the work:	Palaeolimnological research and regular environmental/ecological monitoring.

Declaration

We the undersigned have assessed the activity and the associated risks and declare that the risks will be controlled by the methods listed. Those participating in the work have read the assessment. The work will be reassessed whenever there is significant change and at least annually.

Name (Block Capitals)

NEIL ROSE SIMON TURNER TOM DAVIDSON

Signed:

Supervisor: Dr Neil Rose
Researcher(s): Dr Simon Turner, Dr Tom Davidson

Date:

Review date for risk assessment:

Purpose

The purpose of this document is to identify the risks which relate to the planned fieldwork, and to describe the control procedures which will be adopted.

The risk assessment document identifies a set of hazards which may be relevant to the project. For each hazard category the risk is identified and appropriate control measure(s) described.

Environment	Hazards involved with environment must be considered	
Hazard (s)	***Risk(s)***	***Control Measure(s)***
Weather	General/Miscellaneous	• Consult a daily weather forecast before setting out. Check Met Office forecast, Tel: 0891-500401. • Wear clothing suitable for expected weather conditions. However, be prepared for sudden changes and where possible take a change of clothes. • Strong winds and cold weather reduce energy levels; take adequate food and drink supplies.
	Hypothermia - This results from dangerous loss of body heat. The main land based cause is wind chill, through inadequate clothing. Immersion in cold water can rapidly lead to hypothermia.	• Ensure clothing is appropriate and use a survival suit in extremely cold weather. • Wear a woollen hat - this will significantly reduce heat loss. • Wear lots of layered clothing - remove layers to reduce sweating. • Wear woollen gloves under waterproof gloves. • Eat plenty of food and drink plenty of fluids.
	Frostbite - This results from lack of circulation in the extremities caused by severe cold. It is most common when hands or feet are wet. High altitude can exaggerate the problem as circulation tends to be less efficient in these environments.	• Wear woollen gloves under waterproof gloves. • Make sure shoes/boots are loose enough to allow room for two pairs of warm socks and still not be tight. Restricting circulation with tight shoes will make you more prone to frostbite. • Be aware that the altitude you will be working at will affect your susceptibility to frostbite. • Ensure that footwear is waterproof.
	Poor Visibility - This can be due to driving rain, snow etc. or fog. Working at dawn or dusk can also lead to visibility problems.	• If problems are due to adverse weather conditions, seek shelter and wait for weather to clear. • If working at dawn or dusk, ensure a torch is carried. • Wear high visibility clothing, so that you can be seen easily.
	Sunburn - This can occur even in cold conditions, especially where there is reflected light - e.g. close to or on water, at high altitude, in snowy conditions. Any exposed area of the skin is susceptible (esp. face and hands).	• Use a high factor Sun block • If working in full sun, do not expose skin unnecessarily. • Make sure back of neck is covered. • Wear Sun glasses to protect eyes.
	Dehydration- This can occur in hot or cold weather - wind can be a	• Drink plenty of fluids - it is recommended that more than 3 litres per person per day is necessary in hot weather.

	contributing factor. Heatstroke/heat exhaustion - These are due to the body over heating and are often accompanied by dehydration.	• Do not drink alcohol and avoid caffeinated drinks. • Avoid working in full sun. • Drink plenty of fluids. • Always wear a sun hat. • Keep back of neck covered. • If feeling particularly hot, find some shade and rest

Manual Handling	**Do manual handling activities take place? (Tick relevant box)**	**NO** **YES X**	**Move to next hazard -**

Hazard(s)	***Risk(s)***	***Control Measure(s)***
Unstable loads in Vehicle	Equipment must be stacked properly in the vehicle to reduce moving whilst the vehicle is in motion. This may injure the driver/passenger or may fall when the doors of the vehicle are opened for unloading.	• Ensure that heavy equipment is on the bottom of the pile. • Pack awkward shaped items into square boxes if possible. • Tie down any loose items. • Do not stack equipment higher than the seats.
Back Injury – Lifting	Incorrect lifting technique used.	• Stand close to object, with feet apart. • Keep back straight. • Bend knees. • Keep head up. • Avoid twisting or bending or repetitive work. • Seek proper training in manual handling techniques
Back injury – Carrying equipment to site.	Incorrect carrying techniques	• Where possible carry equipment in a rucksack. • Do not overload rucksacks. • Do not carry equipment further than is necessary - get as close to site as possible by other means. • Do not single-handedly attempt to carry anything that is "too heavy". This will vary from individual to individual - know your limits and do not be persuaded to over stretch that limit. • Be aware of awkward shaped loads and unevenly balanced loads.
Injury to arms and hands – Carrying equipment to site	Strains, sprains, breaks, cuts and grazes	• Where possible dismantle large pieces of equipment into smaller, lighter components. • Where possible carry equipment in a rucksack. • Wear protective gloves. • Keep sleeves rolled down.

		• Be aware of sharp edges/corners. • Be aware of pieces of equipment which may pinch the skin - hinges, lids etc.

Chemical/Biological	**Are chemical or biological hazards involved? (Tick relevant box)**	**NO** **YES X**	**Move to next hazard -**
Hazard(s)	***Risk(s)***	***Control Measure(s)***	
Reagents	Risk of personal injury/long term health effects.	• Read all relevant COSHH guidelines and risk assessments pertaining to reagents being used and follow instructions for use. • Wear gloves and safety glasses • Wash hands frequently. • Clear up any spillages immediately using methods indicated in COSHH guidelines.	
	Risk of environmental damage due to inappropriate disposal.	• Do not pour any chemical into drains, rivers etc. • Dispose of all reagents in accordance with Departmental guidelines (see Departmental Safety Arrangements) • Waste should be brought back to the Department in clearly labeled containers for disposal. • Do not mix different types of waste reagent - they may react violently or need to be disposed of by different methods. • Do not put waste reagents into "food" containers - e.g. Soft drinks bottles, Ice cream tubs etc.	
	Risk of endangering other people's health and safety.	• Do not put waste or any reagent into any inappropriately labeled container. • Label all reagents clearly. • Do not leave chemicals unattended in the field. • Follow guidelines for correct disposal. • Clear up any spillages immediately using methods indicated in COSHH guidelines. • Do not put reagents into "food" containers - e.g. soft drinks bottles, ice cream tubs etc.	

Pathogens **For additional information, consult "Safety in Biological Fieldwork" 3rd edition, 1990 Institute of Biology, 20 Queensbury Place, London SW7 2DZ**	Health risks from working in aquatic environments	• Be aware of health risks from water borne pathogens e.g. Hepatitis 'A', Weil's Disease, Polio and toxic cyano-bacteria • Hepatitis 'A' and Polio vaccinations are highly recommended. • Ensure Tetanus vaccinations are up to date • Wear waterproof gloves. • Clean hands frequently. Always wash hands before eating, preferably using antiseptic medical wipes.
	Risks from agricultural diseases	• Be aware that some fields may be cordoned off due to soil borne pathogens. These are easily spread from field to field if you walk or drive through them. • Do not enter fields with DEFRA notices on them, even if the land owner gives permission. • Consult DEFRA for further information.
Plants	Risk of Poisoning	• Do not eat any wild plants as they may be poisonous or carry parasites. • If handling plant material, wear gloves. • Wash hands frequently • Plants may be coated with any airborne pollutant present in the area – avoid brushing against plants with bare arms/legs.
Plants (cont.)	Cuts and grazes	• Be aware of any plants with thorns, spines etc. • Clean and cover any cuts or grazes sustained to prevent infection. • Be aware of low, over-hanging branches. • Be aware of tree roots and creepers that may cause you to trip.
Invasive Species	Disinfection Protocol for Invasive or Harmful Species	• Invasive Species encountered during fieldwork should not be transferred to other water bodies. • All equipment must be cleaned, disinfected and dried after use. • ECRC guidelines

		(http://www.ecrc.ucl.ac.uk/content/view/75/158/) will be followed.

Working on or near water	**Does the work involve being near or on water? (Tick relevant box)**	**NO** / **YES X**	**Move to next hazard -**

Hazard(s)	***Risk(s)***	***Control Measure(s)***
Boat Handling	Equipment - failure to check or use correctly resulting in personal injury.	• All Named OPAL personnel have RYA National Powerboat Level 2 Certificates. • The Helm is responsible for all decisions within the boat. • Ensure the boat is fully inflated and that valves are sealed correctly. • Inflate the keel (where appropriate) in windy conditions for extra stability. • Wear survival suits in rough, cold conditions. Be aware that conditions in the middle of a lake may be much rougher than those at the launch site, which is often a sheltered bay. • A minimum of two people should be in the boat while in operation. • A tool kit including a knife, pliers, spare shear and split pins must be carried on board. • A set of oars must always be carried. • A boat pump, anchor and rope, a safety line and a bailer should be carried.
	On water bodies where permission to use motor engines has been granted and use is appropriate	• Keep ropes coiled and stowed neatly inside the boat. • Check that the motor is securely attached to the transom and is also attached by a rope to the boat for additional security. • Ensure that there is sufficient fuel in the tank before setting out. • If using an electric engine, treat the battery with care. Always place the battery in a water tight box, lift carefully and place in a stable position on board. Avoid skin contact with any leaked acid. Carry material suitable to soak up any acid spills, e.g. cat litter and strong plastic bags to put the contaminated material in. • Keep the bow rope inside the boat to avoid it tangling in the propeller.
	Risks of Fire – petrol engine	• Ensure fuel tank is horizontal and stable. Open pressure release valve when the motor is running. • No other fuel allowed on board. • Do not smoke/use naked flames near inflatable boats or engines.
	Sinking/drowning	• Buoyancy aids must be worn at all times. • Submerged obstacles can damage the propeller and the bottom of the boat. Always keep a look out, especially in shallow water and rocky areas. • In gusty winds, short steep localised waves of

		considerable size can develop. If the boat length exceeds the wave length, the boat cannot ride the waves. Shelter should be sought. • Ensure safe anchoring prior to cutting the engine.
Working near water	Slips, trips and falls	• The use of waders must be accompanied with the wearing of a lifejacket. • Use a pole to probe ahead to assess the stability of shoreline terrain. • If stuck in mud, do not struggle as this causes deeper sinking. Roll on back and spread weight evenly whilst attempting to "sledge" to firmer ground. • Take special care on slippery rocks around lake shores and river banks. Always look ahead at ground when walking around the water's edge. • When sampling in flowing water environments, be careful of slippery or steep banks and fast currents. If the current is fast or the water looks deeper than knee height, do not enter the water. If you must enter the water, a rope should be tied around your waist and secured to the bank.

Lone, isolated or out of hours working	**Is lone working involved? (Tick relevant box)**	**NO** **YES X**	**Move to next hazard -**

Hazard(s)	***Risk(s)***	***Control Measure(s)***
Miscellaneous	Difficulties in summoning help when required; risk of abuse/attack	• Where possible work, as a minimum, in pairs. • Where possible carry a radio or mobile phone. • Leave details of the field site and a work plan (include contact name and address) with colleagues in the department or at home prior to any trip. • Specify dates and times of departure and return • If your plans change, inform someone as soon as possible. • Do not carry valuables or large sums of money unless you need to. • Carry a personal alarm (This advice is directed to males as well as females - all are equally vulnerable when alone!) • Instigate a "check-in" system with a colleague or supervisor - Phone in at regular intervals. If you do not phone or return at a certain time arrange for suitable action to be taken.
Accommodation during fieldwork	Risks to personal safety	• At reception, try to avoid letting other people overhear your name and room number. • Do not go into other people's rooms unless you know it is absolutely safe. • Do not allow people into your room unless you know

		who they are. • If you hear a disturbance, stay in your room and phone for help.

Ill Health	**The possibility of ill health must be considered**	
Hazard(s)	***Risk(s)***	***Control Measure(s)***
Accidents	Risk of Injury	• Ensure that one of the fieldwork team is trained in First Aid. • Carry a First Aid kit and know how to use it. • Medical Supplies or treatment - be aware of where these can be bought or received if there is an accident • Have plans of action and be aware of where help can be sought should an accident occur in a remote location. • Remember that it is essential to fill out an accident report and return it to the Departmental Safety Officer on return. It may help to make notes as soon after the incident as is possible.
Medical Conditions and Fitness	Risk of illness whilst in the field	• Ensure any necessary medication is carried at all times • Ensure someone else is aware of medical conditions and will recognise signs and symptoms. They should also be informed of the location of medication. • Diabetics should ensure sufficient food is carried in case there is a delay in returning. • Medical Supplies or treatment - be aware of where these can be bought or received if a medical condition or illness arises
	Fatigue leading to lack of concentration, accidents and risk of injury	• Do not try to do too much in one day, especially if the work is to be followed by a long drive home • Be aware that working at high altitude can quickly lead to fatigue due to reduced oxygen intake. • Lack of sleep can lead to accidents - ensure sufficient rest is taken.
	Lack of Physical Fitness - leading to risk of personal injury/illness	• Know your limitations - do not be forced to over-stretch your limit. • Do not be afraid to tell someone if you feel unwell or cannot carry on with a task. • Plan your work within your limits. • If you feel unwell - stop.
Allergies	Anaphylactic Shock - severe cases of allergic reaction can result in breathing difficulties	• Seek medical attention immediately. • Be cautious of the first signs of allergic reaction and DO NOT ignore them. • Allergic reactions can cause discomfort and in severe cases anaphylactic shock. • If aware of an allergy, carry any necessary

		medication. Be aware that some forms of anti-histamine can cause drowsiness. If affected do not continue with fieldwork.
	Insect Bites and stings	• Be cautious of the first signs of allergic reaction and DO NOT ignore them. • If aware of an allergy, carry any necessary medication. Be aware that some forms of anti-histamine can cause drowsiness. If affected do not continue with fieldwork. • Be cautious of the first signs of allergic reaction and DO NOT ignore them.
Food and Drink	Lack of food and Drink - various risks including dehydration, fatigue, fainting etc.	• Do not forget to stop for food breaks • Drink plenty of fluids, particularly in hot weather. • Always carry plenty of water. • Carry food supplies with you if necessary. • Breakfast is particularly important before a day in the field.
	Alcohol - Risks of dehydration; inability to work efficiently due to hangover; in cold weather, alcohol consumption can lead to hypothermia	• Avoid drinking excessive amounts of alcohol on the evening before going into the field. • Avoid drinking alcohol during fieldwork. • Be aware that alcohol can impair judgment and will remain in the system for several hours after consumption.

Equipment	**Will equipment be used? (Tick relevant box)**	**NO** **YES X**	**Move to next hazard -**

Hazard(s)	***Risk(s)***	***Control Measure(s)***
Using Equipment	Risks of personal injury, damage to equipment	• Read Risk Assessments associated with each piece of equipment • Read the instruction manual - follow the instructions! • Read manufacturers safety information • Do not try to bypass or tamper with any safety device. • Seek instruction from trained personnel. • Do not use damaged or faulty equipment. • Ensure the equipment is suitable for the work - try not to improvise! • Do not use electrical equipment in wet/damp conditions or if you have wet hands.
Use of sharps (scalpels)	Risk of personal injury/injury to others during use.	• Use safe techniques when using sharps. Think and plan carefully how the work can

Use of sharps (scalpels) cont.	Risk of personal injury/injury to others during use (cont.)	be done to reduce the risks of exposure and sharps injuries. • Use forceps to hold tissues or materials. • Use appropriate PPE (gloves, specs or face shield etc). • Use double gloves. • Don't hold materials with hands unless it's essential. Use forceps or clamps to hold materials when cutting etc. • Keep the other hand without the sharp as far apart as possible from the hand holding and using the sharp. This reduces the risks of a stab or cut injuries. The further apart your two hands are the less likely you are to injure yourself in a sharps accident. • Dispose of waste materials carefully and using the correct route. • Use puncture resistant sharps bins to dispose of used sharps. • Dispose of sharps immediately after use. • Take the sharps bin to the sharps not the other way around. Put a sharps bin where you are going to use the sharps. • Lock the lid onto the sharps bin before use and do not overfill sharps bins. • Disinfect or sterilise equipment and work surfaces after use where applicable. • Do not leave sharps lying around for other people to have an accident and injure themselves.
Equipment Failure	Risk of personal injury/ injury to others	• Do not use equipment found to be damaged or faulty. • Do not attempt to repair equipment if you are untrained. • Report any faults as soon as possible. • Label faulty equipment clearly so that no one else tries to use it. Try to write down the "symptoms" of the failure and any action taken.
Checking Equipment	Risks of personal injury/injury to others	• Always check equipment before use - do not assume it has already been checked. • Check external appearance for signs of wear - electrical cables, ropes, cracks in casing etc. • Ensure that any built in safety devices are operating correctly. • Check for sharp edges. • Do not use equipment that is damaged or

		found to be faulty. • Report any faults found to the relevant people.

Dealing with the Public	**Will you be dealing with the public? (Tick relevant box)**	**NO** **YES X**	**Move to next hazard -**

Hazard(s)	***Risk(s)***	***Control Measure(s)***
Unexpected Behaviour	Risk of personal attack/abuse due to misunderstanding of nature of work.	• Be aware of any delicate issues involved with discussions or interviews e.g. before asking a farmer questions regarding his land management, explain why you need to know. • Ensure landowners and their employees know who you are and what you are doing.
	Aggressive Behaviour	• Do not underestimate the importance of body language. • Talk yourself out of problems; placate rather than provoke. • Do not turn your back on someone who is behaving aggressively. • Stay Calm, speak gently and slowly. • Do not be enticed into an argument. • Avoid an aggressive stance. Crossed arms, hands on hips or raised hands will challenge and confront. • Keep your distance. • Never try to touch someone who is angry - this will not calm the situation. • Keep your eye on potential escape routes
	Physical attack	• Try to get away as quickly as possible. Move towards a place where you know there will be other people. • Carry a personal alarm - set it off as close to the aggressor's ear as possible and then throw it out of reach. • Shout and scream - shout something practical like "call the police!" or "Fire!" - people rarely react to cries of "help!" or "rape!"
Public Places	Causing offence, leading to abuse/attack	• Do not stand in places where you will be causing an obstruction. • Always carry your ID card and be prepared to identify yourself. • Consider your dress carefully - is it suitable for the location. • Make sure you have sought permission

		from relevant authorities to work in your chosen location.

Other Hazards	**Have you identified any other hazards? (Tick relevant box)**	**NO** / **YES**	
Hazard(s)	***Risk(s)***		***Control Measure(s)***
Other	Specific Hazards		Due to the duration of this project (5 years) unforeseen and other specific hazards will be assessed and risk assessments produced.

Extra sources of information

Departmental safety and risk web sites (http://www.geog.ucl.ac.uk/)
Local experts e.g. Environment Agency, Field Studies Council, Local Constabulary, recent visitor to sites.
UK Meteorological Office, tide tables, port authorities
UCL Safety Advisory Unit
UCL Occupational Health Service
Local organisations, e.g. Field Centres
Community representatives
OPAL Regional Personnel.

I have read, understood and agree to abide by the risk assessments and the safe working guidance provided for the project detailed overleaf.

Print Name	Signature	Date
Dr Neil Rose		
Dr Simon Turner		
Dr Tom Davidson		

APPENDIX B. Site sheet (example Edgbaston Pool)

OPAL SITE & SAMPLE SHEET	Date:		Weather:	Air T ºC	Pressure	Cloud	Wind
	Worker/s:						

SITE NAME: Edgbaston Pool – OPAL West Mid — **Lake Water Level =**

SITE CODE: EDGB

Depth Profiles @ SU 05487 83990 — Light meter | Time:

Water Depth = — **Secchi depth =**

Depth	Cond.	ºC	DO
Surfac			
0.5 m			
1 m			
1.5 m			
2.0 m			
2.5 m			

LM Up	
LM Down	
Surface	
0.5 m	
1 m	
1.5 m	
2.0 m	

OBSERVATIONS		CHECKLIST FOR WATER & BIOLOGICAL		
What?	Where (Use GPS)?			
		WATER		
				Bottle No.
		Major Chemistry	Depth: 0.5 m	
		Trace Metals	Depth: 0.5 m	
		Hg Sample	Depth: 0.5 m	
		POPs	Depth: 0.5 m	
		Susp. Solids	Volume:	
		Filtered?		
		Chlorophyll	Volume:	
		Filtered & Frozen?		
		BIOLOGICAL		
		Invertebrates?		
			Depth of tube	
		Phytoplankton		
	Previous locs..	**Zooplankton**		
	SP 05580 83989	Littoral 1		
	SP 05551 84024	Littoral 2		
	SP 05404 84047	Littoral 3		
		Zooplankton		
	SP 05487 83990	Open Water 1		
		Open Water 2		
		Open Water 3		
		Open Water 4		
	SP 05550 83991	Open Water 5		
	Previous loc..	**Diatom Scrape**		
	SP 05584 83993	Location (GPS):		
	Typha	Substrate:		
		CHECK - Preservatives Added to Biological Samples?		

APPENDIX C.
ECRC Invasive or Harmful Species Disinfection Protocol

Invasive and/or Harmful Species Likely to be Encountered in the UK include:
Plants:

- *Crassula helmsii* (Australian Swamp Stonecrop)
- *Elodea canadensis* and *E. nuttallii* (Canadian Pondweed and the similar Nuttall's Pondweed)
- *Azolla filiculoides* (Water Fern)
- *Lemna* sp. (Duckweeds)
- *Hydrocotyle ranunculoides* (Floating Pennywort)
- *Myriophyllum aqauticum* (Parrot's Feather)
- *Lagarosiphon major* (Curly Water Thyme).

Molluscs:

- *Dreissena polymorpha* (Zebra Mussel)
- *Corbicula fluminea* (Asiatic Clam)

Crustaceans:

- *Eriocheir sinensis* (Chinese Mitten Crab)
- *Austropotamobiuos pallipes* (Signal Crayfish)
- Several other exotic crayfish species.

Pathogens:

- *Aphanomyces astaci* (Crayfish Plague)

Disinfection of all equipment used in the field is necessary after visiting:

- Sites having previous records of invasive or harmful species
- Sites where invasive or harmful species are observed
- Sites with fish or crayfish farms
- Sites with boating or fishing facilities
- Sites connected to the canal network
- Sites within, and within half an hour’s drive of, urban areas
- Sites having direct public road access
- Any site in the Norfolk Broads and surrounding area
- Any site outside Great Britain

Disinfection should involve:

- checking all equipment and removing pieces of plants or invertebrates
- washing all equipment with a dilute bleach solution in an area with appropriate drainage
- washing all equipment using a hot high pressure hose
- rinsing all equipment
- thoroughly drying all equipment

APPENDIX D.
Environment Agency water chemistry methodologies

Determinand:	**pH AND CONDUCTIVITY**
Matrix:	Freshwater, effluent and saline
Method of Analysis:	Electrometric
Instrumentation:	Manual and automated SKALAR robotic system.
Principle:	pH which is the negative logarithm to the base 10 of the hydrogen ion activity. Measurement of the electrochemical potential of a cell which is responsive to the hydrogen ion activity and which contains the test solution as electrolyte. The electrical conductivity is a measure of a solutions ability to conduct electricity, and is dependant on the concentration of dissolved mineral salts present and the temperature of the solution The conductivity is measured by inserting an electrode containing two platinum electrodes into the test solution. These are connected to a conductivity meter and the result displayed in μS/cm. .
Range of Application:	pH: 0-14 pH units Conductivity: 1-100.000 uS/cm
Sample Container:	1 litre PET (min volume 200ml)
Storage/Preservation:	Samples are stored in a cold room at 2 - 8C, prior to analysis
MRV:	N/A
QC within Laboratory:	Error Target - 20% total error Precision - Better than 5% RSD Bias - Better than 10% Bias Performance testing - WRc NS30
External Quality Control:	Aquacheck

Determinand:	**DISSOLVED ORGANIC CARBON**
Matrix:	Freshwater, saline and effluents
Method of Analysis:	Automated Colourimetric
Instrumentation:	SKALAR Air Segmented Flow
Principle:	The automated pre-treatment of the sample with acid and the entraining of the acidified liquid with a high velocity stream of nitrogen results in the removal of any inorganic carbon. The carbonate free sample is mixed with an oxidising agent and subjected to UV radiation. The CO_2 generated reacts with a weakly buffered phenolphthalein indicator and the decrease in colour is proportional to the organic carbon concentration Measured spectrophotometrically at 550 nm.
Range of Application:	Up to 10 mg/l. The range may be extended by dilution
Sample Pre-treatment:	Samples are stored in a cold room at 2 - 8C, prior to analysis. Samples are filtered prior to analysis.
Sample Container:	1 litre PET
Storage/Preservation:	Samples are stored in a cold room at 2 - 8C, prior to analysis
MRV:	0.2mg/l
QC within Laboratory:	Error Target - 20% total error Precision - Better than 10% RSD Bias - Better than 5% Bias
External Quality Control:	Aquacheck

Determinand:	**TOTAL NITROGEN**
Matrix:	Freshwaters and Effluents.
Method of Analysis:	Automated Colourimetric
Instrumentation:	SKALAR Air-Segmented Flow Analyser
Principle:	In this automated method, the sample is mixed with an oxidised agent at a raised temperature, mixed with a buffer and brought into a UV digester. This converts both organic and inorganic nitrogen compounds to nitrate. The sample then uses copper cadmium wire to reduce nitrate to nitrite. A colour complex is formed by further reaction and this is measured at 540nm.
Range of Application:	Up to 5.0mg/l. Range may be extended by dilution.
Sample Container:	1 litre PET
Storage/Preservation:	Samples are stored in a cold room at 2 - 8C, prior to analysis
MRV:	0.2mg/l
QC within Laboratory:	Error Target - 20% total error Precision - Better than 10% RSD Bias - Better than 5% Bias
External Quality Control:	Aquacheck

Determinand: **NUTRIENTS**
ALKALINITY, AMMONIA, CHLORIDE, ORTHO-PHOSPHATE, TOTAL OXIDISED NITROGEN, NITRITE and SILICATE.

Matrix: Freshwaters and Effluents

Method of Analysis: Automated Colourimetric

Instrumentation: Discrete analyser

Principle: The analyte of interest reacts with different reagents to form coloured compounds at characteristic wavelengths which are measured spectrophotometically.

Range of Application: The stated range may be extended by dilution.

Determinand	Low Range mg/l	Very Low Range mg/l
Ammonia as N	0-2	0-0.2
Total Oxidised Nitrogen as N	0-20	0-0.2
Nitrite as N	0-1	0-0.02
Orthophosphate as P	0-2	0-0.1
Chloride	0-200	N/A
Silicate as SiO_2	0-20	0-1.0
Alkalinity as $CaCO_3$	0-200	N/A

Sample Container: 1 litre PET

Storage/Preservation: Samples are stored in a cold room at 2 - 8C, prior to analysis.

MRV:

Determinand	Low Range mg/l	Very Low Range mg/l
Ammonia as N	0.03	0.007
Total Oxidised nitrogen as N	0.2	0.007
Nitrite as N	0.004	0.0007
Orthophosphate as P	0.022	0.0015
Chloride	1	N/A
Silicate as SiO_2	0.2	0.012mg/l
Alkalinity as $CaCO_3$	15	N/A

QC within Laboratory:
Error Target - 20% total error
Precision - Better than 10% RSD
Bias - Better than 5% Bias

External Quality Control: Aquacheck

Determinand:	**TOTAL PHOSPHORUS**
Matrix:	Freshwater and Effluents
Method of Analysis:	Colourimetry
Instrumentation:	Aquakem
Principle:	Condensed phosphates are hydrolised in the presence of dilute acid and organophosphates are oxidised with persulphate to convert them to orthophosphate. Orthophosphate is reacted to form a blue compound which is measured colourimetrically at 760nm and 880nm.
Range of Appliation:	Up to 5.0mg/l. The range may be extended by dilution of the sample.
Sample Container:	1 litre PET (min volume 200ml)
Storage/Preservation:	Samples are stored in a cold room at 2 - 8C, prior to analysis
MRV:	0.02mg/l (0.003mg/l low level)
QC within Laboratory:	Error Target - 20% total error Precision - Better than 5% RSD Bias - Better than 10% Bias Performance testing- WRc NS30
External Quality Control:	Aquacheck

Determinand:	**CHLOROPHYLL A AND PHAEOPHYTIN**
Matrix:	Freshwater, saline and effluents
Method of Analysis:	UV Spectrophotometric determination
Instrumentation:	UV Spectrophotometer
Principle:	The sample is filtered to remove plant material. Chlorophyll a is extracted with acetone and its concentration determined spectrophotometrically by carrying out absorbance measurements at 665nm for chlorophyll a. The concentration of phaeophytin is estimated after acidification and re-measuring the absorbantions.
Range of Application:	Up to 50ug/l total chlorophyll a for a 1000ml sample. The range may be extended by dilution of the sample.
Sample Container:	1 Litre green plastic bottle
Storage/Preservation:	Samples are stored in a cold room at 2 - 8C, prior to analysis in dark bottles. Samples may be pre-filtered on site and stored in foil packets.
MRV:	0.5ug/l Chlorophyll
QC within Laboratory:	Error Target - 20% total error Precision - Better than 10% RSD Bias - Better than 10% Bias
External Quality Control:	Aquacheck

Determinand: **BARIUM, BORON, CALCIUM, IRON, LITHIUM, MANGANESE, MAGNESIUM, POTASSIUM, SODIUM, STRONTIUM AND SULPHATE**

Matrix: Surface waters, groundwaters, sewages and trade discharges

Method of Analysis: Inductively Coupled Plasma Optical Emission Spectroscopy (ICPOES)

Instrumentation: Optima 5300 or Vista Pro

Principle: A plasma torch maintained at a temperature of between 6000˚C and 10,000˚C by the reaction between radio frequency (RF) waves (provided by a RF generator) and argon. The constituent elements introduced into the torch are excited in the source and emit spectra characteristic to specific elements. The concentration of an element is measured by comparison of the intensity of light emitted from the sample with that emitted by standards of known concentration.

Range of Application: The range may be extended by use of dilution or linearity check.
All values are for both total and dissolved analysis

Determinand	mg/l	Determinand	Mg/l
Barium	1	Manganese	2
Boron	2	Potassium	20
Calcium	200	Sulphate	250
Iron	2	Sodium	200
Lithium	10	Strontium	5
Magnesium	50		

Sample Container: 125ml plastic narrow neck bottle

Storage/Preservation: Samples requiring dissolved metals analysis should be filtered through 0.45 micron membrane filters before acidification.

MRV: All values are for both total and dissolved analysis.

Determinand		Determinand	
Barium	10 ug/l	Manganese	10 ug/l
Boron	100 ug/l	Potassium	0.1mg/l
Calcium	1 mg/l	Sulphate	10 mg/l
Iron	30 ug/l	Sodium	2 mg/l
Lithium	100 ug/l	Strontium	20 ug/l
Magnesium	0.3 ug/l		

QC within Laboratory:

Error Target	-	20% total error
Precision	-	Better than 5% RSD
Bias	-	Better than 10% Bias

External Quality Control: Aquacheck

Determinand:	**ACTIVE ALUMINIUM**
Matrix:	Surface waters and ground waters.
Method of Analysis:	Colouring reaction followed by analysis ultra violet / visible Spectrophotometer
Instrumentation:	Ultra Violet / Visible Spectrophotometer
Principle:	The reaction of active aluminium with pyrocatecol violet to form a blue – coloured complex the concentration of which is measured by spectrophotometry at 570nm.
Range of Application:	0-200 µg/l . The range may be extended by use of dilution
Sample Container:	125ml plastic narrow neck bottle
Storage/Preservation:	Samples must not be refrigerated and analysis should start as soon as possible.
MRV:	4 µg/l
QC within Laboratory:	Error Target - 20% total error Precision - Better than 5% RSD Bias - Better than 10% Bias
External Quality Control:	None available

Determinand: **TOTAL AND DISSOLVED METALS BY ICP-MS ALUMINIUM, CADMIUM, CHROMIUM, COPPER, LEAD, NICKEL, ZINC, ANTIMONY, BERYLLIUM, COBALT, MOLYBDENUM, SILVER, TIN, TITANIUM, VANADIUM**

Matrix: Surface waters, groundwaters, sewage and trade effluents and leachates

Method of Analysis: Inductively Coupled Plasma Mass Spectometry.

Instrumentation: Perkin ELAN 6000 and 6100

Principle: The sample to be analysed, as a solution is nebulised and dispersed into a stream of argon gas. The gas stream is injected into the cone of a high temperature plasma sustained by radio frequency fields. Energy is transferred from the plasma to the sample, dissociating, atomising and ionising it in turn. Ions are separated according to their mass to charge ratios as they pass through a quadrupole mass filter. They are detected via pulse ion counting with a channel electron multiplier. The number of registered ions from a given isotope depends directly on the concentration of the relevant element in the sample.

Range of Application: Range may be extended by use of dilutions or linearity check.
All values are for both total and dissolved analysis.

Determinand	**µg/l**	**Determinand**	**µg/l**
Aluminium	500	Silver	25
Copper	500	Beryllium	25
Lead	500	Cobalt	25
Chromium	50	Molybdenum	100
Cadmium	50	Antimony	25
Nickel	500	Tin	30
Zinc	500	Titanium	250
		Vanadium	25

Sample Container: 125 ml plastic narrow neck bottle.

Storage/Preservation: Samples are stored in a cold room at 2 - 8C, prior to analysis. Samples requiring dissolved metals analysis should be filtered through 0.45 micron membrane filters before acidification.

MRV: All values are for both total and dissolved analysis.

Determinand	**µg/l**	**Determinand**	**µg/l**
Copper	0.5	Silver	1.0
Lead	0.4	Beryllium	1.0
Nickel	5.0	Cobalt	1.0
Zinc	5.0	Molybdenum	3.0
Chromium	0.5	Antimony	1.0
Cadmium	0.1	Tin	2.5
Aluminium	10	Titanium	2.0
		Vanadium	2.0

QC within Laboratory:	Error Target	-	20% total error
	Precision	-	Better than 10% RSD
	Bias	-	Better than 10% Bias

External Quality Control: WRc NS30**,** Aquacheck

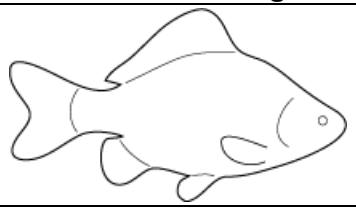
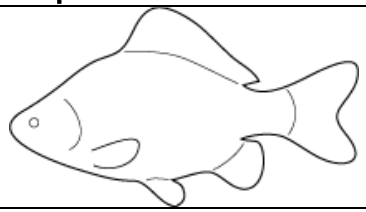
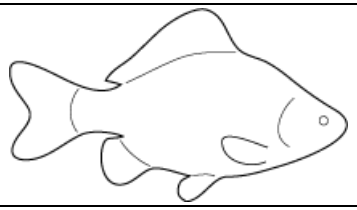
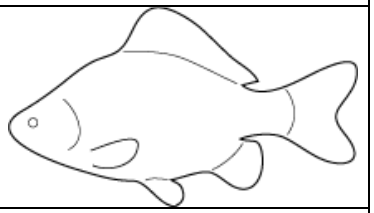
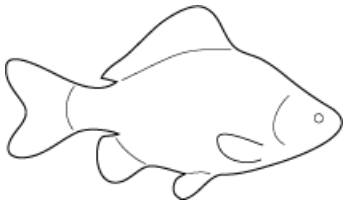
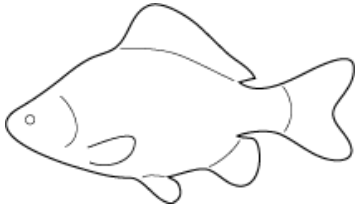
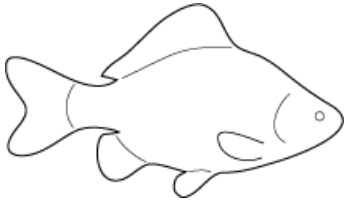
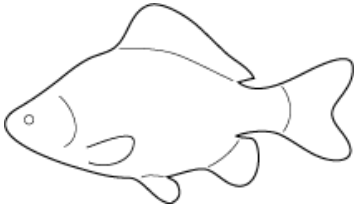
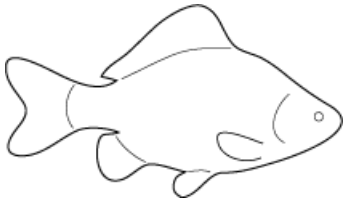
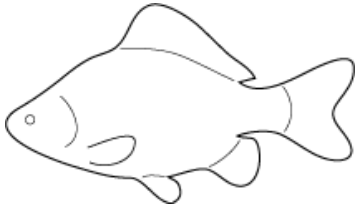
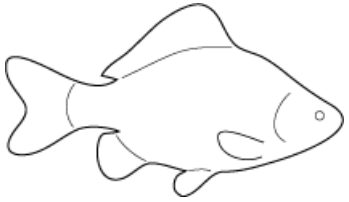
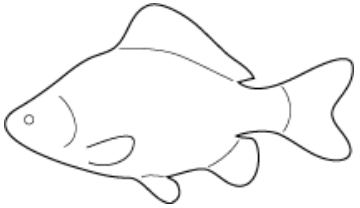
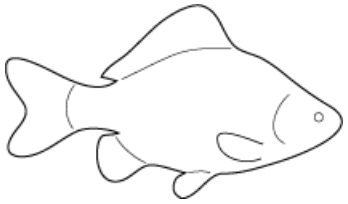
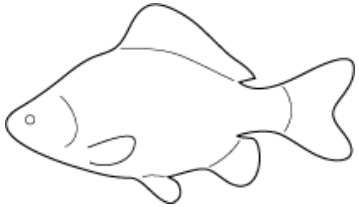
APPENDIX E.

OPAL – FISH SAMPLING FIELD DATA SHEET (i)

SITE:				**LOCATION (GPS):**					**Investigators:**			
Date:												
SAMPLE COLLECTION												
How were fish captured:												
Sampling Duration		Start:		End:			Duration:					
					METALS					**POPS**	**Age**	**Notes**
	Species	**Length (cm)**	**Mass (g)**	**Sex (M/F)**	**Liver**	**Kidney**	**Flesh**	**Stom. Contents**		**Flesh**	**Scales**	
FISH 1												
FISH 2												
FISH 3												
FISH 4												
FISH 5												
FISH 6												
FISH 7												
FISH 8												
FISH 9												
FISH 10												
Return												
Return												
Return												
Return												
Return												

APPENDIX E.

OPAL – FISH SAMPLING FIELD DATA SHEET (ii)

Anomalies/Infections/Damage on sampled fish? Mark and detail					
FISH 1			**FISH 5**		
FISH 2			**FISH 6**		
FISH 3			**FISH 7**		
FISH 4			**FISH 8**