A protocol for plant macrofossil analysis of peat deposits

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SUMMARY

Analyses of plant macrofossils can be used to reconstruct the development of the local vegetation on peatlands, and thus to elucidate successional processes. In the case of ombrotrophic peatlands, such analyses can also be used to generate palaeoclimate data. Identification of plant macrofossils in peat deposits is essential for accurate ^{14}C dating. We present a brief overview of the sample pre-treatment procedure and available techniques for estimating macrofossil composition, and we recommend identification guides.

KEY WORDS: ombrotrophic bogs, palaeoclimate, palaeohydrology, *Sphagnum*.

1. INTRODUCTION

Plant macrofossils, with a median size range of 0.5–2 mm, are visible to the naked eye (Birks 2007, Figure 1). Unlike pollen and non-pollen microfossils, many of them can be identified to species level, enabling more accurate palaeoenvironmental reconstructions. Because of their size and/or weight, plant macrofossils are not usually transported far from the parent plants, and in peat deposits represent the former *in situ* vegetation. Excellent preservation is possible in raised bog deposits. Macrofossil analyses of fen (Hughes & Barber 2003) and blanket peats, as well as archaeological deposits (Chambers *et al*. 2007), are also commonplace. They have been used extensively to reconstruct bog surface wetness (BSW) as evidence for climate change (van Geel *et al*. 1996, Barber *et al*. 1998, Hughes *et al*. 2000, Mauquoy *et al*. 2008), to trace mire development pathways (Hughes & Barber 2003), in studies of long-term vegetation development to inform conservation management (Chambers *et al*. 2007), to investigate the rate and nature of carbon sequestration in peat deposits (Heijmans *et al*. 2008), and to reconstruct archaeological contexts. Numerous plant macrofossil diagrams have been generated using European and North American peat deposits, but application of the technique has not been confined to these parts of the world. Macrofossils with excellent preservation have also been identified in southern South American peatlands (Mauquoy *et al*. 2004), Ile de la Possession (Van der Putten *et al*. 2008) and South Georgia (Van der Putten *et al*. 2009).

Various techniques are available for estimating the abundance of macrofossils in peat deposits. The simplest techniques assign *ordinal values*, for example: 1 = rare, 2 = occasional, 3 = frequent, 4 = common and 5 = abundant (Walker & Walker 1961, Barber 1981). The most detailed (and the most time consuming) techniques estimate absolute numbers, calculated as either concentrations (number of objects per unit volume) (Janssens 1983, Booth *et al*. 2004) or influx (based on age-depth models). The Quadrat and Leaf Count (QLC) technique (Barber *et al*. 2003) adopts an intermediate approach which delivers quantitative estimates of the major peat components (%) and numbers (n) of fruits, seeds and charcoal fragments. The different macrofossil components of a single peat sequence may be presented at different levels; for example Väliranta *et al*. (2003) express mosses, dwarf shrub remains and cyperaceous roots as percentages, small leaves and bud scales as ordinal values, and fruits and seeds as absolute numbers. However, before deciding which technique(s) to adopt, thought must be given to the goal of the macrofossil analysis. If the intention is simply to determine the approximate composition of the peat samples, for example to inform selection of the best location for a 'master' core in stratigraphic survey of a peatland, the ordinal technique is likely to be the optimum method. On the other hand, if the goal of the research is to reconstruct a detailed record of changes in BSW as a palaeoclimate proxy, then the QLC technique or absolute estimates will be required to enable subsequent conversion of the data into reconstructions of mire surface wetness using either the Dupont index (Dupont 1986) or ordination techniques (for example DCA or PCA). Ordination techniques work best with data expressed as

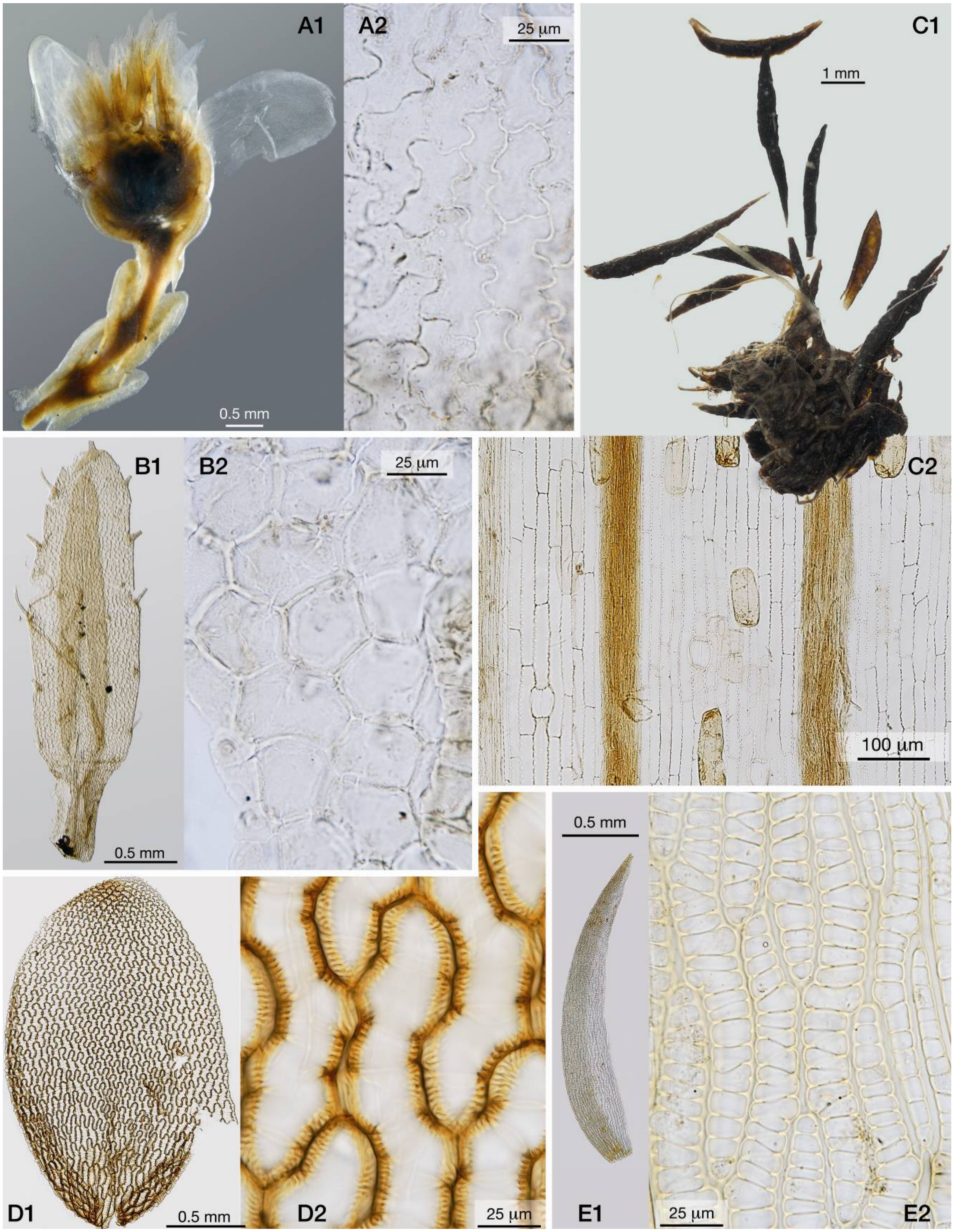

Figure 1. Examples of plant macrofossils commonly encountered in bog peat. A1: *Calluna vulgaris*, stem with flower; A2: *Calluna vulgaris*, leaf epidermis; B1: *Erica tetralix*, leaf; B2: *Erica tetralix*, leaf epidermis; C1: *Eriophorum vaginatum*, part of stem with *in situ* spindles and separate spindles; C2: *Eriophorum vaginatum*, epidermis; D1: *Sphagnum austinii*, leaf; D2: *Sphagnum austinii*, leaf detail; E1: *Sphagnum* sect. *Cuspidata*, leaf; E2: *Sphagnum* sect. *Cuspidata*, leaf detail.

percentages. If it is necessary to include species that were originally quantified using ordinal values, the data can be degraded to presence-absence format to allow this, but the resultant ordinations are usually less satisfactory than those conducted on percentage data because significant information is lacking. In the sections below we describe the QLC methodology, since this is the technique that is most commonly used for palaeoclimate reconstruction.

2. SAMPLE PRE-TREATMENT

The preparation of peat for macrofossil analyses is simple and straightforward. For accurate reconstruction of former peat-forming plant assemblages we recommend a sample size of *ca*. 5 cm^3. The sample should be warmed (boiling is not necessary) with 5% KOH/NaOH for 30–45 minutes to dissolve humic and fulvic acids, then disaggregated on a sieve (100 or 125 µm) using a 'squeezy' bottle of distilled water for rinsing. When sieving, the residue in the sieve should be kept just below the water surface in order to minimise damage to any plant macrofossils and charcoal fragments. This is especially important when *Sphagnum* is present, in order to avoid the detachment of stem leaves, which are highly distinctive in many species (e.g. *Sphagnum fimbriatum*) and thus enable identification of *Sphagnum* remains to species level. We do not recommend the use of stains in sample preparation because the colours of macrofossils can be helpful in the identification process.

3. DETERMINATION OF MACROFOSSIL COMPOSITION

The first stage of the analysis estimates volume percentages of *Sphagnum* and other main peat components - for example ericaceous rootlets, *Eriophorum* remains, other mosses (where present) - for the whole sample. Ideally, the pre-treated sample is poured into a trough (e.g. a *ca.* 20×10 cm glass beaker or bowl) and sufficient distilled water is added to just float the remains, which are then scanned using a low power (×10 – ×50) stereo-zoom microscope with a 10×10 square grid graticule inserted into one of the eyepieces. If a large beaker/bowl is unavailable or there is insufficient space under the stereo-zoom microscope to accommodate such a receptacle, petri-dishes may be used but the material must then be examined in parts; a little of it is poured into a petri-dish, gently stirred, inspected, then more is poured into another petri-dish and the procedure repeated until all of the remains from the sample have been scanned. The trough or petri-dish is moved randomly to 15 different views, plant macrofossil types are estimated as percentages for each view using the graticule, and the results are averaged to represent the whole sample.

Sub-samples which contain well preserved epidermal tissues of monocotyledon species should be mounted on microscope slides (temporary preparations can be made using water) and identified at ×100 – ×400 magnification. However, there is usually no need to make microscope slides of *Eriophorum vaginatum* remains because, with experience, its characteristics can be recognised under the stereo-zoom microscope. A random selection of at least 100 *Sphagnum* leaves should also be mounted on slides, identified at ×400 magnification, and the results expressed as percentages of the total identifiable *Sphagnum* estimated in the first stage of the macrofossil analysis. Where several parts of a single species are represented in the plant macrofossil assemblage - for example the roots, leaf bases/leaves and seeds of *Rhynchospora alba* - we recommend that each of these parts is logged as a separate pseudo-taxon. Adopting this recording convention will aid interpretation of the resultant macrofossil diagram; for example, the roots of a species typically penetrate older peat strata and it is helpful to know whether the first occurrence of a taxon is represented by above-ground or below-ground vegetative parts or by seeds that could be more widely transported.

Fruits/seeds and macroscopic charcoal fragments are simply counted and expressed as the total number (n) present in the sample (i.e. in the trough or all of the petri-dishes). If multiple petri-dishes are used, heavier fruits/seeds and macroscopic charcoal fragments are more likely to be found in the last part of the sample examined. Charcoal fragments can be placed into size (length) classes, e.g. <0.5 mm, 0.5–1 mm, 1–1.5 mm, 1.5–2 mm and >2 mm. Volume percentages of 'above-ground' remains are often very low, for example *Andromeda polifolia* leaves, *Calluna vulgaris* leaves, *Calluna vulgaris* stems, *Empetrum nigrum* leaves, *Erica tetralix* leaves and *Vaccinium* spp. These remains should also be counted separately.

When the macrofossil analysis is complete, the sample should be stored in a sealed plastic bag or tube with a few drops of 5% HCl to prevent further decomposition and contamination by bacteria or fungi. Where possible, sub-samples should be stored in the dark in a cold room at 3–4°C so that they remain available for subsequent ^{14}C dating.

4. IDENTIFICATION GUIDES

Examples of commonly occurring macrofossils are presented in Figure 1. The use of a reference collection of type material is highly recommended, along with the identification plates in Mauquoy & van Geel (2007). There are also good plant macrofossil plates in Grosse-Brauckmann (1972, 1974, 1992), and drawings in Katz *et al*. (1977). Branch and stem leaves of *Sphagnum* and the branch leaves of brown mosses can be identified using Smith (2004). The drawings in Daniels & Eddy (1990) are also very good for *Sphagnum* identification although the taxonomy is now out of date. For the study of *Sphagnum* macrofossils from eastern North America, Bastien & Garneau (1997) is very useful. European wood samples may be identified with the aid of Schweingruber (1990), whilst many European seed types can be found as colour plates in Cappers *et al.* (2006) or online at www.seedatlas.nl (access code required).

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