The Analysis Of SOCs In Vegetation: Influences Of Plant Type And Preparation/ Extraction Technique.

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Introduction

It is important to know the levels at which semi-volatile organic compounds (SOCs) are found in vegetation, and the kinetics of their transfer from the air because this will determine: I) the amount which will enter the grass/cow/human food chain, II) processes of air-surface exchange of SOCs which are of relevance to global cycling and air concentrations, and III) the validity of using vegetation as a predictor of local air concentrations. The largest compartment within a plant leaf for storage of SOCs is the cuticle, a surface structure composed of the polymer cutin embedded with and overlain by cuticular waxes [1]. The cuticle is also directly in contact with the air, making it the site of air/plant partitioning of SOCs. Cuticle structure and thickness vary between plant species, from 0.87μm in grasses to 4.7μm in pine needles, for example [2]. Other lipophilic compartments that exist within the plant, such as intracellular lipids and structural polymers, are separated from the air by the cuticle and the watery matrix of plant cells, which could reduce the importance of such structures by slowing the rate at which they accumulate SOCs. It could also impede the extraction of SOCs from within them. It is clear from the literature that a great variety of extraction methods have been used by researchers for vegetation analysis [3-6]. Some are relatively ‘gentle’, such as the dipping of leaves in solvent, compared with others that are more exhaustive e.g. soxhlet extraction. In this paper we will present evidence that the extraction procedure which is adopted for studying SOCs in plants will be critical in influencing the amount and reproducibility of compound removed from the plant.

Method

A series of method development experiments were carried out which compared: I) the amount of PCBs extracted by hexane/acetone soxhlet extraction of whole grass with the amount extracted from both finely chopped and blended grass, II) the amount of PCBs extracted from grass by soxhlet extraction in a 4:1 hexane/acetone mixture with the amount extracted by shaking over different lengths of time, III) the amount of PCBs extracted from grass by soxhlet extraction using 2 different solvents (DCM, and 4:1 hexane/acetone mixture) for different lengths of time (4h, 8h, 16h), IV) the amount of PCBs extracted by soxhlet extraction in DCM from whole and blended pine needles and lichens, V) the amount of PCBs extracted from whole pine needles and lichens by different lengths of time of DCM soxhlet extraction (4h and 8h) VI) the amount of PCBs extracted from pine needles and lichens by soxhlet extraction in DCM with the amount extracted by sonication, VII) the amount of PAHs extracted from blended grass by soxhlet extraction using different solvents (DCM, hexane and a 4:1 hexane/acetone mixture) for different lengths of time (4h and 8h), VIII) the amount of PAHs extracted from blended grass by soxhlet extraction in DCM with the amount extracted by sonication, IX) the amount of PAHs extracted from mixed deciduous leaves by soxhlet extraction in DCM for different lengths of time (4h and 8h), and X) the amount of PAHs extracted from mixed deciduous leaves by soxhlet extraction in DCM with the amount extracted by sonification.

Results

The results show that the dipping/shaking method only extracts a small percentage of the PCBs within grass compared with soxhleting (Fig.1). It was found that cutting and grinding up grass before extraction increased the amount of PCBs that were extracted, with grinding having the greatest effect (Fig.2). This effect was observed over the whole range of PCB congeners, indicating that it wasn't due to contamination from laboratory air because of the increased handling time. An 8 hour hexane/acetone soxhlet was found to be the most efficient method for extracting grass (Fig.2).

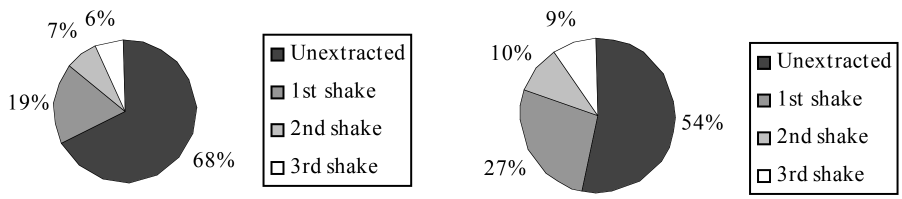


Fig.1 The amount of PCBs extracted from whole (left) and cut (right) grass as a percentage of the amount extracted by an 8 hour hexane/acetone soxhlet of ground grass.

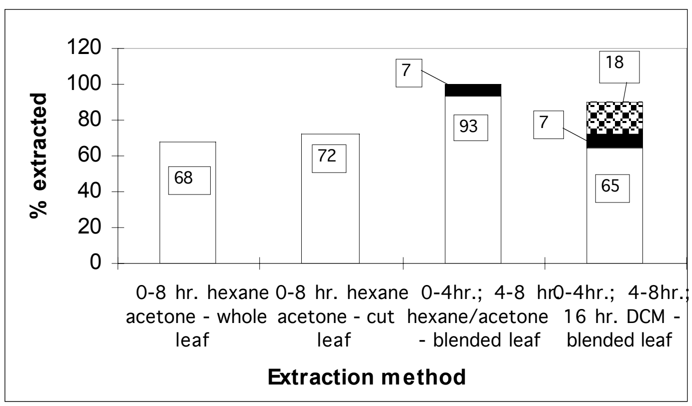
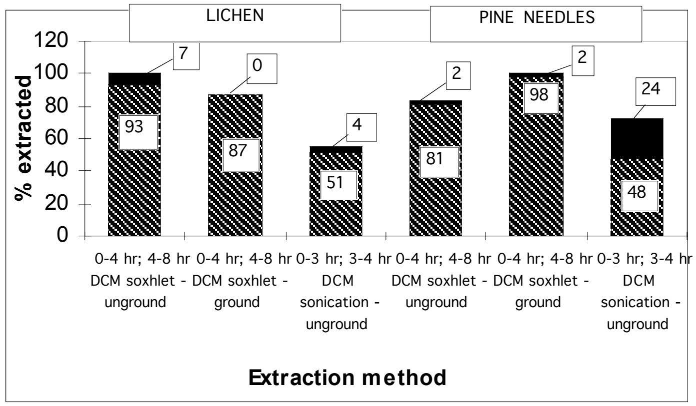


Fig.2 A comparison of soxhleting grass using different preparation techniques, solvents and extraction times.

The most efficient method found for extracting PCBs from lichens and pine needles was the 4 hour DCM soxhlet (Fig. 3). Although more PCBs were extracted if the extraction was continued for another 4 hours, this amount was a small percentage compared to that extracted in the first 4 hours. Also, soxhleting using ground lichens and pine needles extracted more than using unground ones. However, this extra amount was mainly lighter PCBs, which is likely to be the result of contamination during the grinding step. Sonication in DCM was found to be much less efficient than soxhleting, and the results were much more variable (unpublished data). The amount extracted in the 4^{th} hour was small compared to the amount extracted in the first 3 hours, so increasing the extraction time would probably not result in much greater recoveries.

Fig.3 <u>A comparison of the relative efficiency of extracting ground and un-ground lichens and pine needles by soxhleting in DCM and sonicating in DCM.</u>



The most efficient method for extracting PAHs from grass of those tested was soxhleting for 4 hours in DCM, with only 1% extracted in an additional 4 hour period (Fig.4). This extracted more than either soxhleting in a 4:1 hexane/acetone mixture for 4 hours or sonicating in DCM for 3 hours. Neither of these methods improved in efficiency when the extraction time was increased.

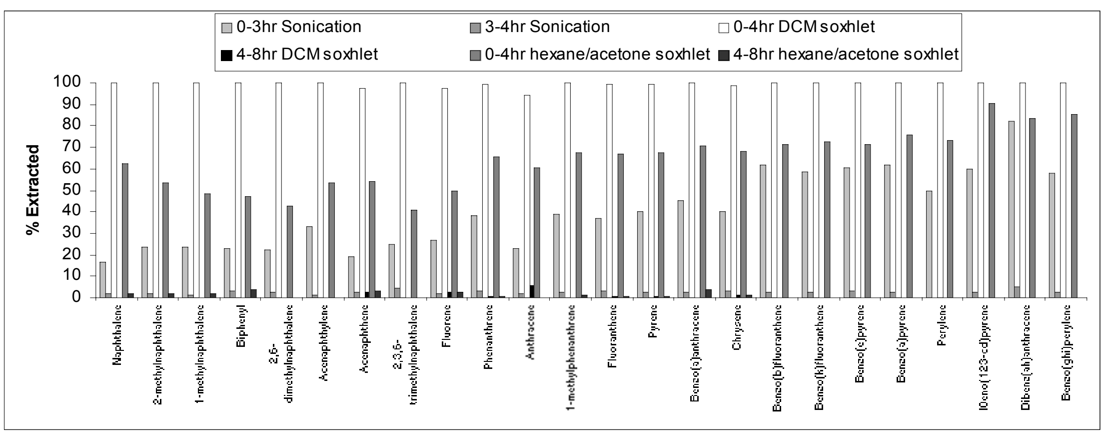


Fig.4 <u>A comparison of the efficiency of soxhleting using DCM with soxhleting using hexane/acetone and sonication for extracting PAHs from grass.</u>

When the results of soxhleting and sonication were compared for leaf litter, both methods extracted similar amounts of PAHs overall, but sonication was much less reproducible, with greater variability in the amount of the heavier PAHs that it extracted (see Fig.5).

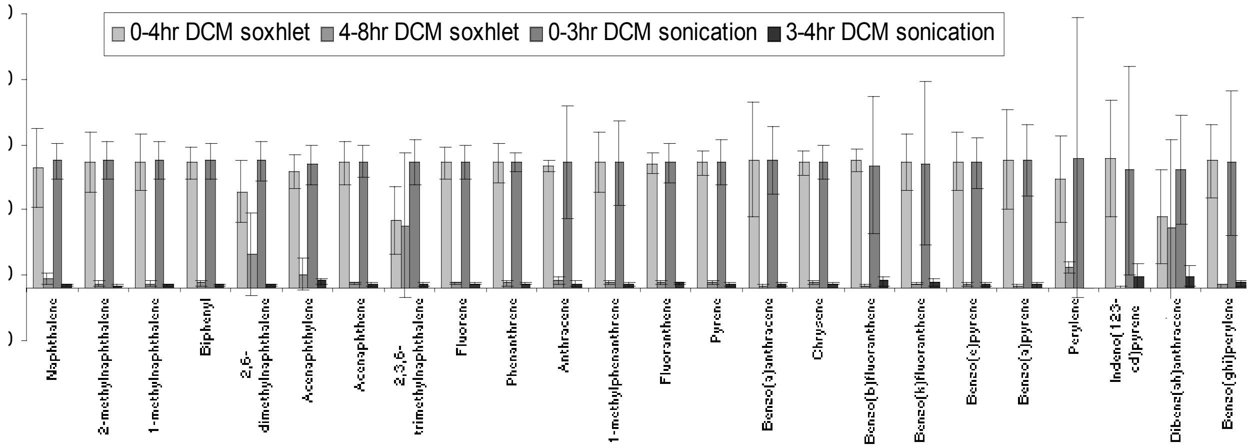


Fig.5 A comparison of the efficiency and variability of soxhleting and sonication for extracting PAHs from leaf litter.

Discussion

The following observations can be made from this study: I) there are species differences in the extractability of SOCs from vegetation; II) the sample preparation technique affects the amount of SOCs extracted; III) methods vary in their efficiency and reproducibility of extracting SOCs from vegetation; and IV) there is variability in the extractability of SOCs from vegetation, with PCBs behaving differently to PAHs. This will have important implications for: I) comparisons of different data sets in the literature; II) seeking to understand the distribution of SOCs within plants; III) seeking to understand the kinetics of uptake; and IV) modelling air/plant partitioning and global fate of SOCs.

Acknowledgements

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