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A method of processing soil core samples for root studies by subsampling

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Abstract Root studies are generally believed to be very important in ecological research. Soil coring is a valuable approach to root research, but it requires a very large amount of processing time. We present here a method for processing soil cores consisting of the combination and homogenization of several soil cores from a plot, with subsequent subsampling for root extraction. The required subsample size was determined for a topsoil and a subsoil sample from a groundnut field and was found to be 5-10% of the total soil sample. Advantages and limitations of the method are discussed.

Key words Root biomass · Root extraction · Root length · Sample homogenization · Soil coring

Introduction

There is general agreement among scientists about the importance of root studies in ecological research. Among the major root research topics are (1) C and nutrient cycling in ecosystems (McClaugherty et al. 1982), (2) competition between plants (Ruhigwa et al. 1992), (3) uptake of water and nutrients by plants (Bowen 1985), and (4) the effects of natural and anthrophogenic stress factors on plant communities (Sandhage-Hofmann and Zech 1993).

A method frequently used in quantitative root studies consists of obtaining soil cores and extracting roots by washing and subsequent hand-sorting under suitable magnification (Caldwell and Virginia 1989; Persson 1990; Vogt and Persson 1991). This method can yield quantitative information about the mass and length of live and dead roots, the root systems of different plant species at the same site, and the number and condition of root nodules, among others. A problem associated with this method is, however, the "tremendous amount of processing time involved" (Persson 1990).

With typical core diameters of 2-10 cm, a single core represents only a small portion of the root zone of a plot (Caldwell and Virginia 1989). To obtain representative information about a site, it is necessary to treat several samples. As the heterogeneity of site and vegetation increases, there is a higher variability between the cores from one site, thus requiring a higher number of samples. The same may be true with increasing soil depth (Mackie-Dawson and Atkinson 1991).

Persson (1990) estimated that sorting a single core sample may take as much as 4-8 h. In consequence, the processing time required can become prohibitive in quantitative root studies, especially when replicated field trials with several treatments are under study, and when the study of root dynamics necessitates the repetition of sampling over time.

The following method permits a reduction in the time necessary for processing soil cores for root studies, thus allowing a better characterization of root systems, especially in heterogeneous sites, and/or an increase in the number of plots under study at the same time.

Materials and methods

To obtain soil samples of a known volume for root extraction, soil cores are taken in the field by driving a sharpened iron cylinder into the ground at several points in a plot, using the usual coring equipment (Persson 1990). If different soil horizons are to be studied, the cores are cut into depth sections, or cores are taken successively from increasing soil depths. Cores taken from the same depth are combined to form a single sample per plot. The total volume of the combined sample is calculated from the volume and number of the single cores. The combined sample is then homogenized as much as possible. This may be done by spreading the soil on a plastic awning or, for smaller samples, in a tub. Soil aggregates are crushed , and longer roots are cut into pieces of a few centimeters in length. The soil and roots are mixed thoroughly. During this homogenization, all roots of more than 2 mm in diameter are collected if they are of interest to the study, as coarser roots are not adequately represented in a small subsample.

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A subsample is formed by taking soil from several randomly chosen points in the homogenized sample. The subsample and the total sample are weighed to the nearest gram. Only the subsample is taken for root extraction, following the usual methods (Caldwell and Virginia 1989; Persson 1990). A second subsample may be taken as a control. The water content of the soil is measured by drying one or two other subsamples at 105 °C to constant weight (approximate-ly 24 h).

From the known volume of the total sample and the weight of the total sample and of the subsample for root extraction, the volume of this subsample is calculated. The root parameters measured in the subsample, like root mass, root length, or number of root tips, can thus be related to the corresponding soil volume as well as to the soil dry weight.

The bulk density of the soil sample can also be calculated, and further subsamples may be analyzed for chemical, physical, or biological soil parameters. These may be related to the observed root variables. Coming from the same total sample, the subsample values should show close relationships with each other.

Whenever a distinction between live and dead roots is intended, care must be taken that fine roots do not dry out during the homogenization and subsampling process, nor in other steps of the processing, as even after rewetting it might be difficult to distinguish between live and dead material (Persson 1990). The homogenization and subsampling should be carried out rapidly, and in a cool and shadowy place. The subsample for root extraction should be wetted after weighing to prevent any further drying. Homogenization and subsampling of a sample of 10-15 kg may take about 20-30 min.

The number of soil cores required to characterize the root systems in a plot depends on the size and the heterogeneity of the plot and the vegetation. It should be determined at the beginning of a sampling program by extracting the roots separately from a number of soil cores coming form the same plot (Vogt and Persson 1991).

The required size of the subsample from which the roots are extracted depends on the size and homogeneity of the total sample. It may be identified at the beginning of a study by the following simple procedure. A sample is taken from the site in question, or from a similar site. It should be the same size as the samples to be studied. This first sample is treated as described above. After homogenization, several subsamples are taken successively from the total sample, and the roots in each subsample are extracted and sorted and their length and mass measured. To find the minimum size of the subsample required for root extraction, the root parameters of the individual subsamples are successively combined and plotted versus their volume or mass as a percentage of the total sample. The minimum subsample size is the percentage of the total sample that yields results which are sufficiently close to those of the total sample.

The procedure was tested using data from a groundnut field on a Plinthic Lixisol (FAO/Unesco 1988) in Central Ivory Coast. The groundnuts had been sown in double rows with 15×15 cm between plants within the double rows and 60 cm between the double rows. A topsoil sample (0-10 cm) and a subsoil sample (10-30 cm) were taken about 2 months after sowing, using a soil corer 8 cm in diameter.

To obtain representative samples for the whole plot of 70 m^2 , four samples were taken between the double rows, and four samples were taken within the double rows. From the latter, two samples were taken directly on a groundnut plant in order to include the taproot in the sample, and two samples were taken in the middle between four plants. The eight samples from each depth were combined to give 5200 g of soil ($105 \,^{\circ}$ C dry weight) and $4021 \,\mathrm{cm}^3$ for the topsoil sample, and 12400 g and 8042 cm³ for the subsoil sample. In view of the sampling method, these samples were regarded as extremely heterogeneous. After the homogenization, six subsamples for root extraction were taken from the topsoil sample and seven subsamples from the subsoil sample. In each case, the combined weight of the subsamples was about 20% of that of the total sample. Live and dead roots of <2 mm diameter were extracted by washing and hand-sorting under $10 \times \text{magnification}$. The length of the live roots was measured according to Tennant (1975), and the dry weight (70 °C for 48 h) of live and dead roots was obtained with a precision of 0.1 mg.

Results and discussion

For the topsoil and the subsoil sample, the results from the subsamples were combined successively in the same order in which they had been taken from the total samples and were plotted versus the mass or volume of the combined subsamples as a percentage of the total samples (Figs. 1, 2). For both root length and root weight, the results obtained with an increasing subsample size showed that with a subsample representing 5-10% of the total sample an almost constant value was obtained, indicating that this proportion was sufficient for reliable subsampling. Complete extraction and hand-sorting of the roots



Fig. 1 Changes in the length of live roots < 2 mm in diameter from topsoil and subsoil of a groundnut plot with increasing size of the subsample for root extraction



Fig. 2 Changes in the mass of live and dead roots <2 mm in diameter from topsoil and subsoil of a groundnut plot with increasing size of the subsample for root extraction

from the total samples of 5200 g for the topsoil and 12400 g for the subsoil would have taken several days for one person.

By reducing the processing time for the soil samples in the manner described, the number of soil cores from a plot can be increased, and the variability in the results which is caused by heterogeneity of the site can be reduced accordingly. However, the final processing of only a part of the total sample taken from the field introduces a new source of error, which depends on the degree of homogeneity in the total sample when the subsamples are obtained and the size of the subsample. Assuming that the natural heterogeneity of the field plot is much greater than that of the homogenized sample, the method should be able to increase the precision of root studies considerably for an equal input of labour.

An evident disadvantage of the method is that it does not yield information about small-scale variability in root parameters within a plot. If this information is desired, the soil cores, or at least a part of them, have to be treated separately. Alternatively, the method described may be combined with a more descriptive technique, like the profile wall method (Mackie-Dawson and Atkinson 1991; Vogt and Persson 1991).

Another case where the homogenization and subsampling of the soil cores is not appropriate is in studies which require a statistical comparison of root data measured at different times in a single plot. This statistical comparison is considered necessary for the calculation of root turnover from changes in root mass between sampling dates (Fairley and Alexander 1985). Nevertheless, the method presented can be used to compare root characteristics and root dynamics of interspersed treatments with statistically independent replications, like block designs, which is often not possible with the conventional method because of the tremendous amount of time needed to characterize a single plot. The method is particularly suitable for conditions where conservation of large quantities of unprocessed soil samples and rapid processing are both impracticable due to lack of equipment and/or qualified personnel.

An important part of this work which still remains to be done is to test the method in different plant communities and on different soil types in comparison with the established method of processing every soil core in full. Moreover, the optimum sampling intensity in the field and the optimum size ratio of subsample to total sample have not yet been established under different conditions.

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