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# Root sampling methods - applications and limitations of the minirhizotron technique

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**Abstract**

Applications and limitations of the minirhizotron technique (non-destructive) in relation to two frequently used destructive methods (soil coreing and ingrowth cores) is discussed. Sequential coreing provides data on standing crop but it is difficult to obtain data on root biomass production. Ingrowth cores can provide a quick estimate of relative fine-root growth when root growth is rapid. One limitation of the ingrowth core is that no information on the time of ingrowth and mortality is obtained.

The minirhizotron method, in contrast to the destructive methods permits simultaneous calculation of fine-root length production and mortality and turnover. The same fine-root segment in the same soil space can be monitored for its life time, and stored in a database for processing. The methodological difficulties of separating excavated fine roots into living and dead vitality classes are avoided, since it is possible to judge directly the successive ageing of individual roots from the images. It is concluded that the minirhizotron technique is capable of quantifying root dynamics (root-length production, mortality and longevity) and fine-root decomposition. Additionally, by combining soil core data (biomass, root length and nutrient content) and minirhizotron data (length production and mortality), biomass production and nutrient input into the soil via root mortality and decomposition can be estimated.

## Introduction

Fine-root systems of plants play an important part in the storage of organic matter and nutrients and in the fluxes of energy and matter in the biosphere. The amount of carbon and nitrogen cycled via fine root-decomposition may be as much as or more than that returned to the soil from above ground litter fall (Arthur and Fahey, 1992; Cox et al., 1978; Joslin and Henderson, 1987). Because of difficulties in measuring fine-root dynamics (production, mortality and longevity) by destructive sampling methods, it has not been possible to estimate nutrient inputs to the soil via fine-root decomposition.

Conventional methods (i.e. soil and ingrowth cores) are unable to measure fine-root production, death and disappearance simultaneously (Kurz and Kimmins, 1987; Santantonio and Grace, 1987). The destructive nature of repeated soil coreing makes direct measurement of these processes impossible (cf. Joslin aud Henderson, 1982). Problems with destructive sampling methods are, differentiation between temporal differences in root standing crop and separation of living and dead roots. (Singh et al., 1984; Vogt and Persson, 1991).

The development of field methods for the study of the growth dynamics of fine roots has always been hampered by the fact that they are not readily visible. This has resulted in root growth phenomena being mainly interpreted from excavated core samples. Minirhizotrons have been used as a tool for observation of roots and to study root function (Bates, 1937; Böhm, 1974; Taylor, 1970).

An international minirhizotron workshop was held in Båstad, Sweden, September 17–20, 1996 as a continuation of the LTER (Long-Term Ecological research) root workshop, Estes Park Colorado, USA. The objectives of the workshop were:

- to exchange scientific results and critical reviews on the development of minirhizotron technique,

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image processing and the analysis of minirhizotron data.
- to discuss the advantages and disadvantages of the minirhizotron technique in quantifying demographic processes underlying root dynamics (production, mortality and longevity) and fine-root decomposition.

## Fine roots - definition

The most widely used root size for studies of fine-root production and mortality is less than 1 or 2 mm in diameter (Vogt and Persson, 1991). When using minirhizotrons, roots < 2 mm in diameter are often measured (cf. Majdi and Kangas, 1996). These fine roots consist of mycorrhizal short roots that are morphologically very distinct from the rest of the root system and include both the mycorrhizal host and fungal mantle tissues.

## Sequential soil coreing

Root cores can be used to obtain estimates of root length and mass, number of root tips, mycorrhizal biomass, root distribution and nutrient contents. This method can be applied only on easily penetrable soils by using iron cylinders with a sharpened edge which are driven into the ground to the desired sampling depth (Vogt and Persson, 1991). However, few data have been reported for depths greater than 1 m. In many soil types, the occurrence of stones or boulders prevent the use of metal coreing tools for root sampling. Large soil block excavations (monolith method) may then be the only feasible way of obtaining root samples.

Root coreing has its obvious limitations, the most important being that processing is time-consuming and therefore limits the frequency of samplings during the growth period. In addition, it is difficult to obtain data on root decay (cf. Vogt and Persson, 1991).

## Ingrowth core

Ingrowth cores can be used to get a quick and less laborious estimate of relative fine root production (Flower-Ellis and Persson, 1980) when root growth is rapid. This method can be used to obtain relative growth and to observe the effects of experimental manipulations on root growth (Persson and Ahlström,1991). The principle of this method is very simple. A soil volume of varying shape is removed and the same volume is replaced with root -free soil surrounded by a mesh bag. Root-free mesh bags filled with sieved mineral soil from the study area, are placed in open pits and sequentially resampled after defined time periods. It is also possible to replace the original soil with some artificial soil substrate, such as sand or perlite. By varying the nutrient content of the bulk soil in ingrowth cores, the effect of a specific nutrient or combination of nutrients on root growth on a microsite level can be studied. A major limitation of the ingrowth core is that no information on time of root ingrowth or mortality is obtained when the amount of living and dead roots are measured. The homogenised soil habitat of the reconstructed ingrowth core will present a high-nutrient and less competitive environment, which will be recolonized at different rates from other parts of the soil.

## Minirhizotron method

Minirhizotrons are a nondestructive method which is useful in monitoring the same root over a long time period. Minirhizotrons are transparent glass tubes that are installed in the soil. Root intersections along the tubes are viewed with a miniature video-camera. In recent years the minirhizotron technique has been developed (Hendrick and Pregitzer, 1996; Smit and Zuin, 1996) to increase our understanding of fine root dynamics (production, mortality and longevity).

Minirhizotrons have been installed in different ways in different ecosystems, i.e. vertically (Majdi and Nylund, 1996), at different angles and horizontally. In conjunction with minirhizotrons, different image-processing routines and software programs have been used to analyze the recorded images. The latest example of these is an interactive PC-based software package (ROOTS) developed by Hendrick and Pregitzer (1992), which makes it possible to identify the same roots on successive dates by recalling previous images. In this context, root length production is defined as the measured length of new roots between time $t$ and time $t + 1$. Mortality is defined as measured root lengths at time $t$ that move into the dead fraction or have disappeared by time $t + 1$, plus the length of any roots that are dead when they first appear at time $t + 1$.

Other minirhizotron image processing methods have been developed (Heeraman et al., 1993; Smucker, 1990), but with less capability to measure longevi-

ty. Minirhizotrons can be used to obtain quantitative data on root length production, root length mortality, longevity, rooting density and root diameter (Hendrick and Pregitzer, 1996).

The minirhizotron technique is very useful for studying the effects of different experimental manipulations, such as fertilization regimes, on root longevity (Majdi and Kangas, 1996) and thinning on root production. Minirhizotrons can even provide qualitative information, such as frequency of mycorrhizal short roots (Majdi and Nylund, 1996), root colour, branching characteristics and root decomposition.

## Application and limitation of the minirhizotron method in relation to destructive methods

Unlike destructive fine-root sampling methods (i.e. soil and ingrowth cores), the same fine-root segment in the same soil space can be monitored for its life and stored in a data base for processing, thus minimizing the spatial component of experimental error. In contrast to the sequential sampling of soil and ingrowth cores, the minirhizotron method can provide a simultaneous calculation of fine-root length, production and mortality. The difficulties of separating excavated fine roots into living and dead vitality classes are avoided, since it is possible to judge directly from the images the successive ageing of the individual roots. In this context, minirhizotrons can be used to quantify directly the rate of fine-root length production and mortality. The rates of fine root longevity and decomposition rate can be calculated by following individual roots and by tracing the length and diameter of dead root segments.

Decomposition and nutrient release of fine roots obtained by the mesh bag technique, may give unreliable estimates because of disruption of the rhizosphere, and from 50 to more than 80 percent of fine-root mass may remain after one year (cf. Fahey, 1992; Fahey et al., 1994). In contrast to buried bags, fine roots observed by the minirhizotron method can disappear completely over time (Smit and Zuin, 1996) especially when voids occur in the tube-soil interface which cause negative impact on both water and nutrient uptake.

Longevity data obtained by the minirhizotron technique (Majdi and Kangas, 1996) show that fine roots in forest ecosystems can have a relatively long life span. These findings contradict results obtained by the soil coreing method of earlier root studies (cf. Persson, 1978). Population statistics such as birth rate, death rate and median life span can be defined for fine roots. If the potential for water or nutrient uptake is considered to be age-dependent, then it is possible to describe age cohorts of roots in relation to the environmental factors by using the minirhiztron technique. It is not clear whether fine roots have a determinate life span that is regulated by carbon allocation or whether they have an indeterminate life span and die when they have been supplied with a given amount of starch (Marshall and Waring, 1985). By combining minirhizotron data (root-length production and mortality) and data obtained by soil cores (biomass and length and nutrient content), biomass production and mortality and nutrient input into the soil can be estimated. Further information on the application of minirhizotrons can be found in Hendricks and Pregitzer (1996).

## Limitations of minirhizotron method

In contrast to sequential coreing, it is not possible to obtain any information on changes of the chemical composition of fine roots or of rhizosphere soil. Fine-root length data are not directly convertible into a dry mass. Data obtained by soil cores (standing crops) are needed to estimate biomass production and nitrogen and carbon input into the soil, via root mortality and decomposition obtained by minirhizotrons.

The other limitation of minirhizotrons is the difficulty linked to their installation in stony soils. However, by using inflatable minirhizotrons it is possible to study the root dynamics in stony soils too.

There is always a much greater proliferation of fine roots in the O-horizon than in the deeper horizons, where the mycorrhizal the-root tips proliferate vigorously due to improved moisture and nutritional conditions. There is difficulty in measuring fine root growth in the O-horizon using minirhizotrons in forest soils, partly because of shallow depth of this layer and partly because shallow roots are underestimated (cf. Majdi et al., 1992; Samson and Sinclair, 1994). In ecosystems with woody plants it takes at least one year for the roots to attain equilibrium, since the soil will be recolonized after the installation of minirhizotron tubes (cf. Majdi and Nylund, 1996.) Future development of the minirhizotron technique should focus on how to minimize artifactual effects, i.e. tube installation on the root population, or improving data storage and a minimizing the time required for analyse minirhizotron images.

## Conclusions

The minirhizotron technique, in conjunction with the recently developed image processors make it possible to quantify demographic processes underlying root dynamics (production, mortality and longevity) and fine root decomposition. By combining data (root length production) obtained by minirhizotrons with data (root biamass, nutrient concentrations) obtained by soil cores, biomas production and nitrogen and carbon input into the soil may be estimated.

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## References

Arthur M A and Fahy T J 1992 Biomass and nutrients in an Engelmann spruce subalpine fir forest in north central Colarado: pools, annual production and internal cycling. Can. J. For. Res. 22, 315–325.

Bates G H 1937 A device for the observation of root growth in the soil. Nature 139, 966–967.

Böhm W 1974 Minirhizotrons for root observations under field conditions Z. Acker Pflanzenb. 140, 282–287.

Cox T L, Hams W F, Asmus B S and Edwards N T 1978 The role of roots in biogeochemical cycles in an eastern deciduous forest. Pedobiologia 18, 264–271.

Fahey T J and Arthur M A 1994 Further studies of root decomposition following harvest of a northern hardwoods forest. For. Sci. 40, 618–629.

Fahey T J 1992 Mycorrhizae and forest ecosystems. Mycorrhiza 1, 83–89.

Flower-Ellis J G K and Persson H 1980 Investigation of structural properties and dynamics of Scots pine stands. Ecol. Bull. 32, 125–138.

Heeramam D A, Crown P H and Juma N G 1993 A color composite technique for detecting root dynamics of barley (*Hordeum vulgare* L.) from minirhizotron images. Plant and Soil 157, 275–287.

Hendrick R L and Pregitzer K S 1992 The demography of fine roots in a northern hardwood forests. Ecology 73, 1094–1104.

Hendrick R L and Pregitzer K S 1996 Applications of minirhizotrons to understand root function in forests and other natural ecosystems. Plant and Soil 185, 291–302.

Joslin J D and Henderson G S 1982 A test of the budget method: a refined approach to the measurement of the root turnover. Fourth Central Hardwood Forest Conference, 8–10 Nov. 1982, Lexington, Ky. Ed. R N Muller. pp 220–228. University of Kentucky, Lexington, USA.

Joslin J D and Henderson G S 1987 Organic matter and nutrients associated with fine root turnover in a white oak stand. For. Sci. 33, 330–346.

Kurz W A and Kimmins J P 1987 Analysis of error in methods used to determine fine root production in forest ecosystems: a simulation approach. Can. J. For. Res.17, 909–912.

Majdi H, Smucker A J M and Persson H 1992 A comparison between minirhizotron and monolith sampling methods for measuring root growth of maize (*Zea mays* L. ). Plant and Soil 147, 127–134.

Majdi H and Kangas P 1997 Demography of fine roots in response to nutrient applications in a Norway spruce stand in SW Sweden. Ecoscience (*In press*).

Majdi H and Nylund J-E 1996 Does liquid fertilization affect fine root dynamics and lifespan of mycorrhizal short roots? Plant and Soil 185, 303–307.

Marshall J D and Waring R H 1985 Predicting the root production and turnover by monitoring root starch and soil temperature. Can. J. For. Res.15, 791–800.

Persson H 1978 Root dynamics in a young Scots pine stand in Central Sweden. Oikos 30, 508–519.

Persson H and Ahlström K 1991 The effects of forest liming and fertilization on fine root growth. Water Air Soil Pollut. 54, 365–375.

Samson B K and Sinclair T R 1994 Soil core and minirhizotron comparison for the determination of root length density. Plant and Soil 161, 225–232.

Santantonio D and Grace J C 1987 Estimating fine root production and turnover from biomass and decomposition data: a compartment flow model. Can. J. For. Res. 17, 900–908.

Singh J S Lauenroth W K, Hunt H W and Swift D M 1984. Bias and random errors in estimators of net root production: a simulation approach. Ecology 65, 1760–1764.

Smit A L and Zuin A 1996 Root growth dynamics of Brussels sprouts (*Brassica olearacea* var. *gemmifera*) and leeks (*Allium porrum* L.) as reflected by root length, root colour and UV fluorescence. Plant and Soil 185, 269–278.

Smucker A J M 1990 Quantification of root dynamics in agroecological systems. Remote Sens. Rev. 5, 237–248. *In* Minirhizotron observation tubes: Methods and applications for measuring rhizosphere dynamics. American Socieity of Agronomy Special Publication Number 50. Ed. H M Taylor. pp 67–80. ASA, Madison, WI, USA.

Taylor, H M, Huck M G Klepper B and Lund Z F 1970 Measurement of soil-grown roots in a rhizotron. Agron. J. 62, 807–809.

Vogt K and Persson H 1991 Measuring growth and development of roots. *In* Techniques and approaches in forest tree ecophysiology. Eds. J P Lassoie and T Hincley. pp 477–501. CRS Press, Inc., Florida, USA.

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