# Tracer applications for assessing nutrient pathways in multi-strata agroforestry systems

J. Lehmann<sup>1\*</sup> and T. Muraoka<sup>2</sup>

#### Introduction

Nutrient cycling is difficult to assess in agroforestry systems due to their spatial and/or temporal discontinuity. Multi-strata agroforestry systems have an especially high complexity, because they may comprise several different tree species in complex geometric order with contrasting above- and belowground biomass distribution. Tracer techniques can help identify and also quantify nutrient fluxes, but have not been used to a large extent in agroforestry research (Lehmann, 1998). In this paper, I will discuss (i) the use of tracers with regard to nutrient fluxes, (ii) their applicability in multi-strata agroforestry systems and (iii) the methodological approach of existing experiments.

#### The use of tracers for assessing nutrient pathways

Types of tracers and applicability

Three conceptually different kinds of tracers can be distinguished which are interesting for multistrata agroforestry research: (i) rare elements (e.g. Sr<sup>2+</sup>, Rb<sup>+</sup>, Br<sup>-</sup>), (ii) radioisotopes (e.g. <sup>32</sup>P, <sup>33</sup>P, <sup>35</sup>S) and (iii) stable isotopes (e.g. <sup>15</sup>N, <sup>34</sup>S, <sup>18</sup>O). These types of tracers differ in their handling, mode of application, analytical procedure and accuracy, in their environmental behaviour, price and availability. (i) The advantages of the rare elements are their easy analyses procedures and relatively low price depending on the background level of the tracer in the environment. The disadvantage is that no main nutrient can be simulated within the nutrient cycle. Rare elements are more useful to study areas of nutrient uptake. Nitrate may be taken as an inert element and its behaviour can be approximated with Br. (ii) The simulation of main nutrients is more easily achieved with radioisotopes, although some are quite expensive and all are hazardous, therefore restricting their use. (iii) A very attractive method is the utilization of stable isotopes. Two principally different applications exist for stable isotope studies, first, the use of tracers applied in excess of natural concentrations in order to follow their pathways and second, the use of tracers whose concentrations are naturally different between soil, soil solution or plants, for example. The enrichment method requires the purchase of expensive tracer material, whereas the natural abundance method requires a very sensitive analyses procedure which may not be available everywhere. Examples for using the natural abundance method include the determination of biological N<sub>2</sub> fixation, where the natural difference between the <sup>15</sup>N-to-<sup>14</sup>N ratio of soil and air is used to quantify the contribution of N<sub>2</sub> fixation to plant nutrition, or of the origin of leachates, which may be a product of soil mineralisation, fertilizer or rainfall. Stable isotopes in excess of the natural abundance are useful for studying N<sub>2</sub> fixation and the fate of applied fertilizer or organic material, which was labelled prior to application.

Learning from natural and human-impacted systems

Surprisingly few experiments have been published using tracers in agroforestry systems. The reason for this scarcity may lay in the associated technical difficulties, since most studies are undertaken in regions where certain specialized equipment may not be at hand. Other reasons may include the lack of methodology for using tracers in agroforestry research. The majority of tracer studies have been

<sup>&</sup>lt;sup>1</sup> Institute of Soil Science and Soil Geography, University of Bayreuth, Germany

<sup>&</sup>lt;sup>2</sup> Centro de Energia Nuclear na Agricultura CENA, University of Sao Paulo, Piracicaba, Brazil

<sup>\*</sup> corresponding author; present address: Embrapa, Manaus, Brazil; Email: johannes.lehmann@internext.com.br

conducted in agricultural systems and more recently in natural ecosystems, but rarely consider interactions between different plant species. Agroforestry introduces a new dimension to tracer applications, as they can elucidate pathways of nutrients between plants with contrasting growth patterns in time and space. In the next sections, the tracer techniques for the investigation of some important processes in multi-strata agroforestry systems are discussed using examples from natural and human-impacted systems.

#### Biological nitrogen fixation

This technique has recieved much attention in recent years, but is not without problems. Considerable difficulties have been encountered with uniformly labeling the soil, with identifying adequate reference trees and with excluding the recycling of fixed N, which changes soil <sup>15</sup>N contents (Danso et al., 1992). The large volume of soil used by trees makes it almost impossible to provide a soil source with a homogeneous <sup>15</sup>N-to-<sup>14</sup>N ratio: for annual crops, the topsoil may be mixed after applying the isotope; this is not possible for deeper rooting trees. Even using natural abundance, a uniform  $\delta^{15}$ N with depth is rather the exception than the rule. On an oxisol in the central Amazon,  $\delta^{15}$ N values increased from around 10‰ at 10cm depth to 30‰ at 200cm (J. Lehmann, unpubl. data). The issue of uniform labeling may be overcome, if a reference tree is used which matches exactly the studied leguminous tree in its above- and belowground growth and N uptake. However, this is rarely the case. Only repeated measurements with various reference trees and acompanying studies of biomass production and root distribution can give acceptable certainty about the accuracy of this method (e.g. Lehmann et al., 1998a).

### Nutrient uptake

The measurement of the fertilizer recovery using isotopes is a well established technique (Zapata and Hera, 1995). Additionally, the uptake of mulched N in agroforestry has been successfully studied with stable isotopes (e.g. Lehmann et al., 1998b). Since a large portion of the fertilized or mulched nutrients is often not taken up by the crops (Lehmann et al., 1998a; Lehmann et al., 1998b), but is either leached or bound in soil organic matter, the use of stable isotopes is preferable. Only then it is possible to make a full assessment of the fate of the added nutrients, i.e. in the soil organic matter (Vanlauwe et al., 1998). What is lacking, however, are detailed analyses of the availability of these nutrients including organic N compounds in long-term experiments.

Investigations of the areas from which plants take up nutrients have been conducted many times, but often using the difference method by analysing soil nutrient depletion, without being able to exactly trace the fate of nutrients. A comprehensive study on the use of P radioisotopes for determining the distances and depths of nutrient uptake by several fruit trees of IAEA (1975) was not followed by further methodological work. <sup>15</sup>N has been used in a few experiments, but the technique has not been fully explored. Problems arise as the mobility of N compounds is high and the exact area of N uptake is difficult to control. Furthermore, due to the large dilution in plant and soil, highly enriched <sup>15</sup>N substances must be used, which prove very expensive. Rare elements, though seldom used in this context (e.g. Van Rees and Comerford, 1986; Lehmann et al., 1998c) deserve more attention. It must be kept in mind, however, that rare elements may not be as rare as expected: in a tropical dryland, large amounts of exchangeable Sr were found in the soil, making it difficult to measure the added Sr tracer (Weigl and Lehmann, unpubl. data).

## Nutrient fluxes

Nutrient leaching is often assessed with tracer applications. The type of tracer used will depend on the nutrient of interest. For N and S, stable isotopes exist that possess the same properties as total

soil N or S. For an approximation of nitrate movement through soil, other quasi inert tracers such as Br (e.g. Sticksel et al., 1996) or Cl can be used. This does not work, however, when plant uptake is part of the processes which are to be determined.

The source of leached nitrate in mature forest ecosystems has been investigated by using the natural abundance of  $^{15}$ N and  $^{18}$ O (Durka et al., 1994). The principle is to use the isotope ratio difference e.g. between rainwater, soil solution, soil (subsoil and topsoil), microbial biomass or plants as sources for one of the compartments. For example, the  $\delta^{15}$ N values of the soil solution can give information about the proportion of N derived from the soil and from the rain water, if the  $^{15}$ N contents of these compartments are known. The source of throughfall can also be determined with natural abundance or with labelling the tree, as shown with  $^{35}$ S by Cape (1993).

The transfer of biologically fixed N from legumes to associated non-legumes has been a focus for some time and experimental methods have been developed using <sup>15</sup>N (Chalk and Smith, 1994). These methods may have some value for intercropping with annual plants, but their applicability in agroforestry experimentation is doubtful (Lehmann et al., 1998a). Injections of <sup>15</sup>N into trees may provide a more direct solution to the question of N input to soil (Horwath et al., 1992).

#### Tracer studies in multi-strata agroforestry

Only rarely have tracer techniques been used in multi-strata agroforestry systems. The majority of the few tracer experiments that do exist in agroforestry research, have been conducted in alley cropping systems.

Lateral nutrient uptake of different tree crops in multi-strata agroforestry can be studied with tracers (e.g. Zakra et al., 1991). By applying <sup>15</sup>N enriched ammoniumsulfate at the topsoil of different areas in a multi-strata agroforestry system, it could be quantitatively determined that *Theobroma grandiflorum* took up more of its N from areas covered with *Pueraria phaseoloides* than the associated *Bactris gasipaes* (Fig.1; J. Lehmann, unpublished data). Due to the rapid nitrification in many tropical areas, however, as well as the high mobility of nitrate, the measurements of foliar <sup>15</sup>N contents should be done very rapidly.

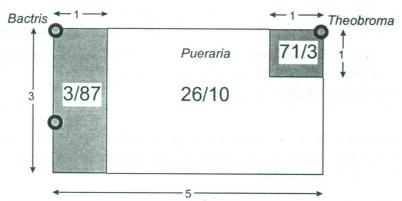


Figure 1. Relative N uptake (%) of *Theobroma grandiflorum* and *Bactris gasipaes* (devided by a slash) from areas under *Theobroma*, *Bactris* or intercropped *Pueraria* (numbers at arrows are distances in [m]); values are significantly different at p<0.05 (J. Lehmann, unpublished data).

Quantification of N pathways of biologically fixed N using natural <sup>15</sup>N abundance poses large difficulties. Thus, the natural abundance method in the preceding experiment indicated that the N nutrition of the non-leguminous trees benefited from atmospherical N<sub>2</sub> fixation of the pueraria grown between the trees (data not shown); however, it was necessary to consider the soil N uptake

compared to the control, as the <sup>15</sup>N abundance increased with depth. Using the natural abundance method for N, sources of error often are the variation of soil <sup>15</sup>N contents with depth or the <sup>15</sup>N discrimination during microbial or chemical transformation. This problem can sometimes be overcome with adequate control plots. These control plots are difficult to design, as they essentially need the same plant species and arrangement to avoid different root activity between experimental plot and control.

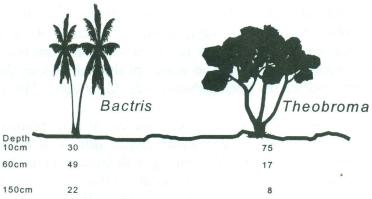


Figure 2. Tree uptake of <sup>32</sup>P from different soil depths in a multi-strata agroforestry system as proportion of total <sup>32</sup>P uptake (J. Lehmann and T. Muraoka, unpublished data).

Studies of nutrient uptake from different depths can help to understand soil nutrient dynamics. In a multi-strata system with four different tree crops and a leguminous ground cover, uptake of applied <sup>32</sup>P differed profoundly between trees (Fig.1). Total uptake from different soil volumes cannot be quantified with this approach, but the importance of different soil depths for crop nutrition can be compared.

## How to apply tracer techniques?

The presented examples illustrate the power of tracer techniques for assessing nutrient pathways in multi-strata agroforestry systems. The complexity of the addressed processes, however, require a special experimental approach. Specific questions have to be identified which can be answered using tracer techniques. These questions have to be as simple as possible to avoid uninterpretable results. With a combination of different tracer experiments it is possible to accurately follow pathways of nutrients. Robust experiments for perennial agroforestry systems are:

- Comparison of nutrient uptake from different depths and distances to the trees
- Quantification of soil nutrient use in different soil areas
- Quantification of nutrient losses by leaching

This survey emphasizes the need to exploit the potential of tracers for the advancement of agroforestry research. Further studies are necessary to develop tracer techniques which can be easily conducted and unambiguously interpreted.

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