# A litterbasket technique for measurement of nutrient dynamics in forest floors

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## ABSTRACT

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We describe a litterbasket technique for quantifying litter decomposition and changes in litter and forest floor nutrient pools over time. Litterbaskets are constructed of wire hardware cloth, $10 \times 10 \times 10$ cm. Intact cores, removed from forest floors, are separated into individual strata (litter layer, F-layer, soil) with plastic window screen. The core is reassembled in the litterbasket, which is replaced in the hole from which the core was removed. Pre-weighed aliquots of experimental substrates can be added to replace the litter layer. The advantages of the litterbasket method include: (1) reduced microclimatic effects relative to litter enclosed in litterbags; (2) allowance for input of exogenous radioactive or stable isotope tracers; (3) quantification of changes in nutrient contents in the various layers of the forest floor over time; (4) easy extraction of invertebrates and quantification of microbial populations from individual horizons; (5) the ability to quantify the movement of radioactive or stable tracers from litter through the forest floor profile over time. Preliminary results from experiments are summarized to illustrate the utility of this technique.

## INTRODUCTION

Rates of forest floor litter decomposition are integrating variables since they are the product of the interactions of resource quality, activities of the forest floor biota and microclimate. Nutrient dynamics and release during decomposition are system-level processes which have important impacts on ecosystem nutrient cycling. Recently, considerable research has focused on nitrogen (N) dynamics, largely because of the limiting nature of N in many forests. Recent work has studied the relationship between N availability and litter production and chemistry (Vitousek, 1982; Nadelhoffer et al., 1983; Birk and Vitousek, 1986), the influence of environmental parameters and/or litter chemistry on decomposition and nutrient release (Meentemeyer, 1978; Berg and Staaf, 1981; Melillo et al., 1982), or the effects of forest disturbance on

N cycling (Aber et al., 1978; Vitousek et al., 1982; Swank, 1986). Fewer studies have examined the role of litter and soil animals on the cycling of N in the litter–soil–plant subsystem (Blair and Crossley, 1988).

In this paper, we describe a litterbasket technique which we have recently begun using to investigate the interactions of fauna, microbes and litter quality in affecting litter decomposition and forest floor N dynamics. Adapted from a design for a substrate box used with litterbags (Olson and Crossley, 1963) and a litter basket used by Stevenson and Dindal (1981), the litterbasket technique has several advantages over litterbags. The technique has improved our measurements of the distribution of microflora and fauna in different forest floor strata, and has provided a means for experimental manipulation of these organisms. Furthermore, using this technique in conjunction with $^{15}$N as a tracer, we have been able to quantify the release of endogenous litter N and its subsequent movement through selected forest floor pools, as well as the immobilization and release of exogenous N inputs. Some preliminary work with radioactive tracers has also proved successful in tracing elemental movement through forest floor strata. The technique seems well suited to experimental applications of biocides, such as insecticides or fungicides, for analysis of the interactions in forest floor food webs affecting nutrient dynamics. Finally, litterbasket techniques offer an alternative to the commonly used litterbag method for measuring decomposition and mineralization. Although widely used (Wieder and Lang, 1982), litterbags have been criticized for alteration of microclimates and fungal colonization (St. John, 1980) and exclusion of larger soil invertebrates. Litterbaskets represent an intermediate step between litterbags and unconfined leaf litter.

## ASSEMBLY AND ANALYSIS OF LITTERBASKETS

Litterbaskets ($10 \times 10 \times 10$ cm) were constructed of wire hardware cloth with a 6-mm mesh and were coated with polyurethane prior to placement in the field to prevent leaching of zinc from the galvanized metal wire mesh. Intact cores, $\sim 8.5$ cm deep, were removed from the forest floor using a square coring tool ($10 \times 10$ cm). The individual horizons of the forest floor profile (L-layer, F-layer and soil) were then carefully separated, plastic window screen (mesh $1.5 \times 1.8$ mm) was inserted between the layers, and the core was reassembled in the litterbasket and inserted into the hole from which the original core was removed. Figure 1 shows an expanded view of the relationship between substrates in the litterbasket. When installed in the forest floor, individual layers are contiguous with the horizons from which they came.

In some experiments, a pre-weighed substrate (dogwood, *Cornus florida* L. leaves, Fig. 1) was inserted between the surficial litter layer (water oak, *Quercus nigra* L. leaves) and the F-layer; in other experiments, a pre-weighed substrate replaced the existing L-layer. Collection of replicate litterbaskets over
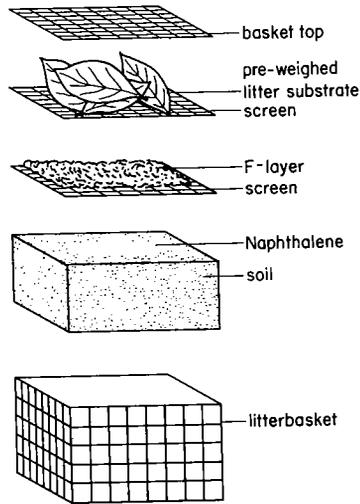
Fig. 1. Design of litterbaskets and arrangement of substrates. When the basket is returned to the hole from which the core was taken, the litter layer will be flush with the rest of the forest floor.

time allowed for quantification of litter decay rates and patterns of nutrient immobilization or mineralization.

Microarthropods were excluded from substrates in some baskets by using a small (1–2 g) addition of naphthalene to the soil surface beneath the F-layer. Although only applied to the soil surface, naphthalene effectively reduced microarthropod populations in all horizons, including surficial litter.

After the litterbaskets had been in the field for 30 days, we added $^{15}$N-enriched ammonium sulfate (5 ml) or a radioactive tracer (1 ml of strontium-85 ($^{85}$Sr)) in liquid form to surficial litter. Additions of $^{15}$N were made once and a set of litterbaskets was collected bi-monthly for a year. Additions of radioactive tracer were made seasonally and a set of litterbaskets was sampled for 4 weeks following each addition.

Litterbaskets were collected into individual plastic bags and returned to the laboratory for processing. Intact litter, experimental substrates and F-layer horizons were carefully separated from the baskets by lifting the underlying screen. Microarthropods were extracted from a subset of these samples using Tullgren funnels. A 5-cm core was taken from the soil portion of the litterbaskets and microarthropods were removed from it using high-gradient extraction (see Crossley and Blair, 1991). Following microarthropod extraction, the substrates were analyzed for nutrient content, isotope ratios or radioactivity, depending upon the experiment. Subsamples of litter F-layer and soil horizons from replicate litterbaskets were used for the enumeration of bacteria, fungi and nematodes. Nematodes were extracted on Baermann funnels and microbial populations were estimated using direct count techniques.

RESULTS OF ANALYSES

Decomposition rates of experimental substrates (dogwood leaves) measured in monthly collections are shown in Table 1. Experiments were conducted at the University of Georgia's Horseshoe Bend facility at Athens, Georgia, in a floodplain forest containing mixed hardwood species dominated by water oak (*Q. nigra*). Rates reported in Table 1 are similar to those obtained for dogwood litter using litterbag methods in the southern Appalachians (Blair and Crossley, 1988). Applications of naphthalene to the soil surface resulted in reduced litter decomposition rates in litterbaskets, and again rates were similar to those obtained with litterbag biocide experiments (Table 1). Naphthalene is frequently used as a biocide in field experiments to reduce soil arthropod populations (Seastedt and Crossley, 1983), but like all biocides may have non-target effects. Newell et al. (1987) reported that naphthalene vapors reduced the radial growth of fungal cultures in sealed containers. Naphthalene vapors alone were effective in excluding soil microarthropods in these litterbasket experiments. We did not measure fungal populations in this particular experiment, but in other experiments we found that naphthalene did not significantly reduce the abundance of total or FDA-active fungi on litter substrates confined in litterbaskets (Callaham et al., unpublished data). In any case, decomposition measurements made on litterbasket substrates are comparable to the more widely used litterbag technique.

Downward movement of radioactive tracers through the layered substrates in litterbaskets was readily measured. Applications of $^{85}$Sr were made to sur-

TABLE 1

Decomposition rates for dogwood (*C. florida* L.) leaves in litterbaskets and confined in litterbags. The decomposition constant, $k$, was estimated from regression of log percent mass remaining on time (days) (after Olson, 1963)

| Treatment | Regression coefficient ($k$, per day) | $r^2$ | Annual $k$ (per year) | Percent remaining (1 year) |
|---|---|---|---|---|
| Control litterbaskets | | | | |
| 1983–84 | −0.00305 | 0.920 | −1.11 | 32.9 |
| 1984–85 | −0.00383 | 0.832 | −1.00 | 24.7 |
| Naphthalene litterbaskets | | | | |
| 1983–84 | −0.00093 | 0.562 | −0.34 | 71.2 |
| 1984–85 | −0.00075 | 0.545 | −0.27 | 76.1 |
| Control litterbags | | | | |
| 1984–85 | −0.00270 | 0.935 | −0.99 | 37.3 |
| Naphthalene litterbags | | | | |
| 1984–85 | −0.00130 | 0.838 | −0.47 | 62.2 |

face litter seasonally (in January, April, July and October) to measure immobilization in forest floor strata in litterbaskets. Half of the litterbaskets were treated with naphthalene to examine the effects of microarthropod reduction on tracer movement. The expected patterns of movement are illustrated in Fig. 2. If microarthropod activity decreased microbial immobilization, a fairly rapid downward movement was anticipated (Pattern "A", Fig. 2). Conversely, slower downward movement in the presence of microarthropods would indicate that microbial immobilization was stimulated by microarthropod activity (Pattern "B", Fig. 2). Our experimental results did not consistently distinguish between these two patterns and we obtained conflicting results regarding microarthropod effects. Leaf litter usually retained 25–50% of the radioactive tracer after 4 weeks. Litterbaskets treated with naphthalene generally showed more rapid downward movement of tracer, suggesting that microarthropod activity stimulated microbial immobilization of tracer in the untreated baskets. Summertime measurements, however, showed decreased downward movement of tracer in naphthalene-treated litterbaskets. This suggests that microarthropods increase tracer movement during the summer season, when their populations and activity are highest, perhaps by comminution of litter or consumption of microbes. In general, over the four seasons, high populations of microarthropods were correlated with more rapid loss of tracer from the litter; slower losses occurred when microarthropod populations were low. These results resemble those reported by Witkamp and
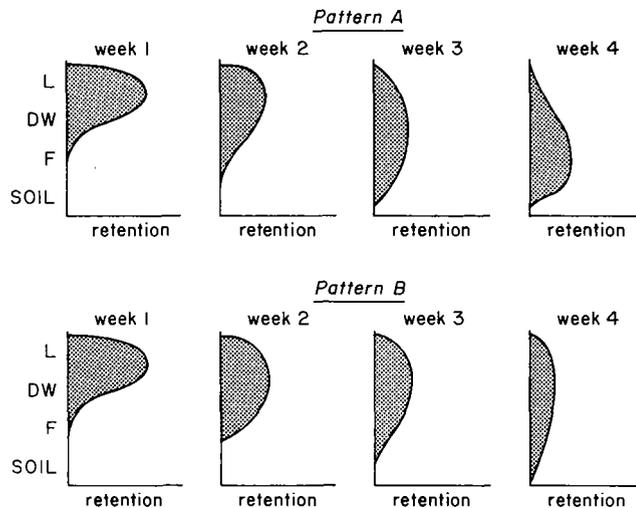


Fig. 2. Expected patterns of downward movement of isotopic tracers applied to surficial litter in litterbaskets. Pattern "A" represents low microbial immobilization in litter and dogwood leaves; Pattern "B" represents higher immobilization in those layers. L = surficial litter; DW = pre-weighed dogwood (*C. florida* L.) leaves; F = F-layer.

Crossley (1966), who found that microarthropods increased leaching of radioactive cesium-134 from white oak leaf litter.

Experiments using surface additions of $^{15}$N to litterbaskets showed a rapid turnover of nitrogen pools in decomposing leaf litter. Nitrogen immobilized from various exogenous sources appears to enter into rapidly metabolized pools. The amount of added $^{15}$N recovered in litter layers declined over time until ~30% remained in the litter after an 8-month period. However, during this same period, absolute amounts of total N in the litter increased, implying simultaneous immobilization of N from other exogenous sources while the labeled N input was being lost from the litter. Measurements of $^{15}$N in the F-layer and soil strata showed a significant downward movement. Tracer accumulated rapidly in the soil horizons of litterbaskets containing chestnut oak (*Quercus prinus* L.) litter, but was slower to do so in dogwood litterbaskets. We attribute this result to (1) a more abundant decomposer community in dogwood litter and (2) a greater C:N ratio in dogwood litter than in chestnut oak, which would favor microbial immobilization. A complete analysis of our experiments with tracers is beyond the scope of this paper on techniques, but the results demonstrate the potential utility of using the litterbasket technique, in conjunction with radioactive or stable tracers, to examine factors affecting forest floor nutrient dynamics.

CONCLUSIONS

The litterbasket technique described here has promise as a means for developing in situ experiments in forest floors. Separating forest floor strata, so important in tracer studies, is less arbitrary because layers are initially separated with plastic window screen. Results summarized here show that litterbaskets are an acceptable technique for studying forest floor nutrient dynamics using both stable and radioactive tracers. Litter decomposition rates measured in litterbaskets were similar to those obtained with litterbags. Mesh size is adequate to admit larger invertebrates such as earthworms. The technique offers an alternative to litterbag methods, while retaining more experimental control than is possible in experiments with unconfined litter.

ACKNOWLEDGMENTS

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