



The fate of glucose, a low molecular weight compound of root exudates, in the belowground foodweb of forests and pastures

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ABSTRACT

Increasing evidence suggests that much of belowground, heterotrophic activity in terrestrial ecosystems is fueled by inputs of low molecular weight carbon compounds (LMWCCs). Root exudation (rhizodeposition) is a primary source of these inputs and will likely increase with rising atmospheric CO₂. Yet the fates of these compounds belowground, as well as the environmental factors that influence them, are relatively unexplored. Using stable isotopes we track the fate of one dominant LMWCC, glucose, in three pasture and three forest sites located in South Carolina, USA. We resolve glucose-derived C in CO₂, dissolved and soil organic C (DOC, SOC), microbial biomass, and microarthropods (Collembola, oribatid and mesostigmatid mites). After 72 h, the greatest proportions of glucose-C are in microbial biomass and SOC, followed by CO₂, DOC, and microarthropods. Within this short time frame, glucose-C propagates through the foodweb to the highest trophic level, predatory mesostigmatid mites. The biomass of these predators is the only variable that explains the relative partitioning across sites of glucose-C, with higher biomass associated with reduced partitioning of glucose-C to respiration and hence greater retention belowground. Our results suggest that LMWCCs entering belowground systems may propagate through soil foodwebs rapidly, and that their partitioning belowground may potentially be determined by higher trophic levels.

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1. Introduction

Low molecular weight carbon compounds (LMWCCs), composed of sugars, organic acids, and amino acids, are some of the most reactive forms of carbon (C) entering the belowground in terrestrial ecosystems (Yang and Janssen 2002, van Hees et al., 2005; Boddy et al., 2007). Indeed, they may fuel the bulk of microbial growth and activity and account for as much as 30% of total soil respiration (van Hees et al., 2005). In doing so, they play both a direct and indirect role in the formation, decomposition and stabilization of soil C stores, the regulation of nutrient cycling, and the provision of energy to belowground foodwebs (Dakora and Phillips 2002; van Hees et al., 2005; De Graff et al., 2010). They are primarily derived from recent photoassimilate that enters the belowground in dissolved form through root exudation (van Hees et al., 2005; De Graff et al., 2010; Strickland et al., 2010; Phillips et al., 2011). Their initial residence in the dissolved organic C (DOC) pool is on the order of hours, and from there they are either sorbed to soil surfaces or

assimilated by the microbial biomass (Saggar et al., 1999; van Hees et al., 2005; Boddy et al., 2007; Fischer et al., 2010). Given this rapid removal of LMWCCs from the DOC pool, their fate is typically accounted for in respired C, microbial biomass, DOC, and soil organic C (SOC), and more often as only what is respired versus what remains in the soil (Fig. 1) (Fischer et al., 2010). This accounting does not resolve the potential for trophic interactions such as microbivory and predation to influence the fate and partitioning of LMWCCs belowground, despite the potential role of such interactions in regulating above and belowground foodweb and C dynamics (Bonkowski, 2004; Bradford et al., 2007; Hawlena and Schmitz, 2010a,b; Wickings and Grandy, 2011).

Studies tracking the fate of photoassimilate show that soil animals, specifically microarthropods, derive a substantial proportion of their C from recently, photosynthetically-fixed C (Pollierer et al., 2007; Bradford et al., 2007; Högberg et al., 2010). Undoubtedly, some of this C enters the belowground as LMWCCs through root exudation (Dakora and Phillips 2002; De Graff et al., 2010; Pollierer et al., 2007), but much of the C may also enter through other pathways (e.g., herbivory) and in other forms (e.g., frass). This means that to understand the specific fate of LMWCCs requires studies that track individual LMWCCs belowground. Glucose, a dominant

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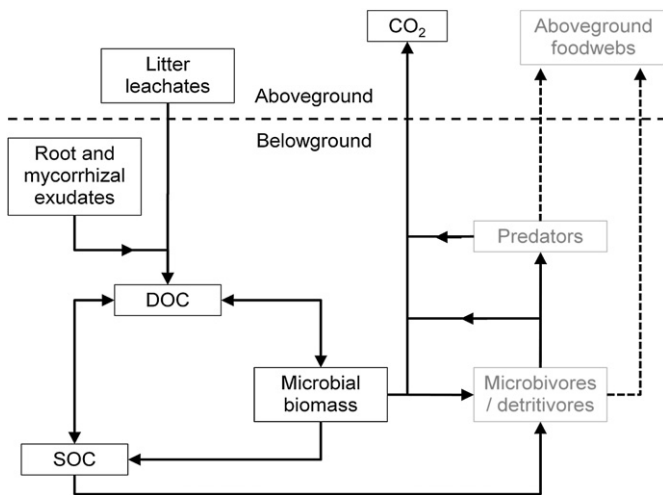


Fig. 1. Conceptual diagram showing the fate and expected pathways of LMWCCs belowground. Generally, these compounds have been tracked in respiration, DOC, SOC and microbial biomass C. However, as these compounds likely fuel a significant proportion of microbial activity, it is also likely that they will enter higher trophic levels in soils and ultimately aboveground systems (indicated in gray). Although these trophic pathways have been explored for C fixed via photosynthesis, they have not been explored for specific LMWCCs.

LMWCC in DOC and exudates (van Hees et al., 2005; De Graff et al., 2010), is commonly used for such tracking experiments but its partitioning in belowground foodwebs is largely unknown. Some common expectations are that the bulk of C derived from LMWCCs will be respired through microbial activity, incorporated into microbial biomass, or sorbed to soil surfaces (Saggar et al., 1999; van Hees et al., 2005; Fischer et al., 2010). Given that most LMWCCs can be assimilated by the microbial biomass, variation in partitioning is often considered to be driven primarily by characteristics of the microbial community (i.e., size, activity, and composition) (van Hees et al., 2005; De Graff et al., 2010; Strickland et al., 2010). Yet research on isotopically-labeled photoassimilates indicates the potential for LMWCCs to rapidly (i.e., days) enter a range of belowground pools from sporocarps to microarthropods (Pollierer et al., 2007; Högberg and Read 2006; Högberg et al., 2008, 2010; Wu et al., 2009). There is also some indication that factors besides those directly related to the microbial community, such as soil nutrient status and land management, influence the partitioning of LMWCCs belowground (Strickland et al., 2010). To better understand the partitioning of LMWCC belowground, and the environmental factors that influence it, requires field investigations that track inputs of specific LMWCCs into belowground C pools and foodwebs.

The objective of this study was to resolve the fate of glucose-C, a common LMWCC often found in root exudates, in the belowground C pools of forests and pastures. We achieve this objective by tracking ^{13}C labeled glucose into soil C (DOC and SOC) and belowground foodwebs as well as soil respiration (CO_2). We also sought to better understand what soil chemical, physical, and biological factors might influence or are at least related to the partitioning of this addition of glucose-C belowground.

2. Methods

2.1. Site description

In June 2007, we conducted a ^{13}C -glucose pulse-chase experiment in three pasture (P1–P3) and forest (F1–F3) sites near the

Calhoun Experimental Forest, South Carolina, USA (approx. 34.5°N , 82°W). Pastures are fertilized, limed, cattle-grazed and rye (*Lolium* sp.) and Bermuda (*Panicum* sp.) grasses are the dominant plant cover. Forests are ~ 75 y old oak-hickory (*Quercus* sp. and *Carya* sp.) stands with minimal understory species composed in part of American Holly (*Ilex opaca*) and Virginia creeper (*Parthenocissus quinquefolia*). One forest site (F2) is grazed by cattle. Soils at these sites are acidic Ultisols classed as fine, kaolinitic, thermic Typic Kanhapludults of the Appling, Cecil, Hiwassee, and Madison series (Callahan et al., 2006). Average bulk densities (for depth 0–7.5 cm) ± 1 S.E. are 1.66 ± 0.10 and 1.16 ± 0.17 g cm^{-3} for pasture and forest sites, respectively. Average soil pH ± 1 S.E. are 5.13 ± 0.09 and 5.17 ± 0.28 for pasture and forest sites, respectively. Mean SOC content (g m^{-2} to 7.5 cm depth) ± 1 S.E. for forests are 1670.5 ± 317.7 and pastures are 1221.5 ± 29.5 . All forest soils and P1 are sandy loams and the other pasture soils are loamy sands (Strickland et al., 2010). All sites are within 12 km of each other and are located on uplands with minimal slope and on interfluvies from similar bedrock (i.e., granitic-gneiss). Thus, we attempted to control for native soil characteristics, geomorphology, and geology. See Strickland et al. (2010) and Table 1 for further site details.

The pasture and forested sites represented two distinct land cover types within the Calhoun Experimental Forest, specifically, and the Southeastern United States, in general, that differ both in their contemporary and historic management regimes (Richter et al., 1999; Richter and Markewitz, 2001). Due to these contemporary and historic management regimes these sites are representative of the variation in soil chemical, physical, and biological characteristics associated with soils in the Southeast and for this reason it should be noted that this was not a controlled study solely aimed at examining differences between land management regimes. It was an observational study aimed at understanding the potential characteristics of a site, including the contemporary management regime, that may influence the partitioning of low molecular weight C compounds belowground and more often than not pastures and forests represented the extremes in many of these characteristics.

2.2. Tracking ^{13}C -glucose

To track ^{13}C -glucose we used the approach described in Strickland et al. (2010). Briefly, two PVC collars (15.4 cm dia., inserted 5 cm into the soil) were placed at each site (i.e., analytical replicates) and, the day prior to glucose additions, water (1 L) was added to each collar to minimize differences in water potentials across collars. Then, 99-atom% ^{13}C -glucose (1 L of 2.5 mM solution) was added to each collar. The amount of C added was small (<25 $\mu\text{g C g}$ dry weight soil $^{-1}$ in the top 7.5 cm), representing $<0.0001\%$ of total soil C. The glucose addition drained for 2 h and then soil respiration was tracked across 72 h. Soil respiration was measured using a closed-chamber technique, and initial samples (i.e., prior to glucose addition) gave natural abundance values for CO_2 produced. A gas sample was taken immediately after sealing each PVC collar with a cap fitted with a septa, and 45 min post sealing, using a 20 mL SGE gas syringe. Samples were transported back to the lab (located in Athens, GA, USA) in evacuated 12 mL Exetainers where total CO_2 concentration was determined on an infrared gas analyzer (IRGA; Li-Cor Biosciences, Lincoln, NE, USA, Model LI-7000) and $^{13}\text{CO}_2$ was determined using isotope-ratio mass spectrometry (IRMS; see below).

After 72 h soil contained within the entire PVC collar was harvested to 7.5 cm depth, divided into halves, with one half sieved (4 mm) before DOC, SOC, and microbial biomass C determinations, and the other half lightly crumbled by hand, to ensure even drying in the Tullgren funnels, for microarthropod extractions. Microbial biomass C and DOC were determined using the modified

Table 1
Soil and community characteristics for study sites. P1–P3 are pastures and F1–F3 are forests. Means for pastures ($n = 3$) and forests ($n = 3$) are shown in the last two rows of the table. Standard errors are shown in parentheses. All values are reported as g m^{-2} to 7.5-cm depth, except fungal:bacterial ratio (unitless), pH, SIR (reported as $\text{g m}^{-2} \text{h}^{-1}$), mineralizable C (reported as $\text{g m}^{-2} \text{h}^{-1}$), soil temperature (reported as $^{\circ}\text{C}$), and soil moisture (reported as % volumetric moisture).

Plot ID	Fungal:bacterial	SOC	SON	P	pH	SIR	Mineralizable C	Microbial C	Microbial N	DOC	DON	NO ₃	NH ₄ ⁺	Soil temp.	Soil moisture	Roots	Collembola	Oribatid mites	Mesostigmatid mites
P1	0.004	1263.7	94.7	1.4	5.1	0.3	76.5	15.5	1.7	21.3	2.5	0.34	1.8	24.1	14.6	1201.5	0.04	0.30	0.02
P2	0.003	1119.8	77.2	2.3	5.3	0.2	54.6	4.9	0.8	14.4	1.9	0.51	10.4	24.3	18.3	514.7	0.03	0.14	0.01
P3	0.005	1280.9	88.5	2.2	5.0	0.2	50.8	6.0	0.3	8.1	1.6	2.23	1.4	24.4	13.0	401.5	0.02	0.07	0.02
F1	0.010	941.5	52.1	0.1	5.5	0.1	62.7	3.8	0.7	11.7	1.5	0.03	2.3	20.0	15.0	252.2	0.03	0.10	0.02
F2	0.003	2748.9	99.5	0.4	4.6	0.2	67.5	21.5	1.2	26.0	1.7	0.04	2.1	20.5	14.5	677.8	0.02	0.35	0.10
F3	0.006	1321.1	67.3	0.1	5.4	0.1	59.0	1.2	0.9	11.5	1.2	0.10	1.5	21.5	21.6	282.8	0.03	0.36	0.08
Pasture	0.004	1221.5	86.8	1.9	5.2	0.22	60.6	8.8	0.9	14.6	2.0	1.0	4.5	24.3	15.3	705.9	0.03	0.17	0.02
	(0.001)	(51.1)	(5.1)	(0.3)	(0.1)	(0.02)	(8.0)	(3.4)	(0.4)	(3.8)	(0.3)	(0.6)	(2.9)	(0.1)	(1.6)	(249.9)	(0.01)	(0.07)	(0.01)
Forest	0.006	1670.5	73.0	0.2	5.1	0.16	63.1	8.8	0.9	16.4	1.4	0.1	2.0	20.7	17.0	404.3	0.03	0.27	0.07
	(0.002)	(550.2)	(14.0)	(0.1)	(0.3)	(0.03)	(2.5)	(6.4)	(0.1)	(4.8)	(0.2)	(0.02)	(0.3)	(0.4)	(2.3)	(137.1)	(0.01)	(0.08)	(0.02)

chloroform fumigation extraction (CFE) procedure as described in Fierer and Schimel (2002, 2003) using a Total Organic Carbon Analyzer (Shimadzu, Columbia, MD, USA, Model TOC 5000) and, for biomass, an extraction efficiency correction of 0.45. Dried (105°C), ball-milled soil was used for SOC determinations on an NA1500 CHN Analyzer (Carlo Erba Strumentazione, Milan, Italy). Microarthropods were extracted using Tullgren funnels into salt water for the first 24 h and then into ethanol for the remainder of the extraction and enumerated under stereomicroscopes. Those organisms collected in the first 24 h were dried at 65°C , weighed, and analyzed for total C. Extractions were continued for 7–10 days until the soils dried and then the additional organisms were counted and total abundance was corrected for the mass of soil used in the extraction. We present data for Collembola, oribatid mites, and mesostigmatid mites as these were the most abundant groups across all samples and were present in samples from all sites after the initial 24 h extraction into salt water. The first two groups are microbivores/detritivores, and the mesostigmatids are generalist predators (Pollierer et al., 2009; Staddon et al., 2010; Crotty et al., 2011).

To determine the amount of glucose-derived ^{13}C in each of the C pools, the atom% ^{13}C was determined using IRMS. The atom% ^{13}C of CO_2 was determined following hand-injection of 5 mL of sample gas into a continuous flow IRMS (CF-IRMS; Thermo, San Jose, CA, USA, Model ThermoFinnigan Delta^{Plus}); the IRMS was coupled to a TOC analyzer for determination of the atom% ^{13}C of DOC and microbial biomass; and solid samples were introduced to the IRMS via an elemental analyzer. The amount of C derived from the ^{13}C -glucose was calculated using the following isotope mixing equation (sensu Ineson et al., 1996): $C_{\text{glucose-derived}} = C_{\text{pool}} \times (\text{atom}\% ^{13}\text{C}_{\text{pool}} - \text{atom}\% ^{13}\text{C}_{\text{nat. abn.}}) / (\text{atom}\% ^{13}\text{C}_{\text{glucose}} - \text{atom}\% ^{13}\text{C}_{\text{nat. abn.}})$, where C_{pool} is the measured size of the pool (cumulative CO_2 , POM C, mineral-associated C, DOC, microbial biomass C, and microarthropod C), $\text{atom}\% ^{13}\text{C}_{\text{pool}}$ is the atom% ^{13}C value of the respective pool in collars where glucose was added, $\text{atom}\% ^{13}\text{C}_{\text{nat. abn.}}$ is the atom% ^{13}C value of the respective pools from additional unlabeled plots (or, for CO_2 , the respective soil collar prior to ^{13}C -glucose addition), and $\text{atom}\% ^{13}\text{C}_{\text{glucose}}$ is the value of the glucose itself. We had one unlabeled PVC collar in each land cover to derive natural abundance values. Glucose-derived C in SOC was calculated by subtracting the contribution to this pool of labeled C in microbial biomass and DOC.

Site-specific soil and community characteristics were determined from 10 soil cores collected using a stratified random approach from 10×10 m plots located at each site. Soil cores (8 cm dia., 0–7.5 cm depth) were sieved (4 mm) and homogenized by site. The characteristics determined included total soil organic C and N (SOC and SON), microbial biomass C and N, dissolved organic C and N (DOC and DON), extractable NO_3^- and NH_4^+ , mineralizable C, soil phosphorus (P), soil pH, soil temperature (via a digital probe thermometer), gravimetric soil moisture, substrate-induced respiration (SIR), fungal:bacterial ratio, total Collembola biomass, total oribatid mite biomass, and total mesostigmatid mite biomass. SOC and SON, microbial biomass C, and DOC were all determined as described above. Microbial biomass N was determined using the same approach used for microbial biomass C. DON and inorganic N pools were determined on a Lachat autoanalyzer (Milwaukee, WI, USA, Model FIA 8000) after extraction in 0.5 M K_2SO_4 . Mineralizable C was determined using a 60-day soil incubation (65% water holding capacity and 20°C) where headspace CO_2 concentrations were monitored 8 times and mineralized C at the end of 60 days was estimated via integration (i.e., mineralizable C was estimated as the area under the curve after plotting CO_2 production against time). Soil P was extracted with Mehlich I double acid and measured on an Alpkem analyzer (OI Analytical, College Station, TX,

Model 3550). Soil pH (1:1 soil:H₂O by volume) was determined using a bench-top pH meter (VWR International LLC., Radnor, PA, USA, Model symphony). Soil temperature and moisture were determined in the field using handheld probes (VWR International LLC., Radnor, PA, USA, Model T-Shaped thermometer; Campbell Scientific, North Logan, UT, USA, Model Hydrosense™, probes for temperature and moisture, respectively) inserted into the soil to 7.5 cm depth. For SIR, soils slurries were incubated for 4 h (20 °C) – after a 1 h pre-incubation – with excess substrate and then head-space CO₂ concentrations were determined using an infrared gas analyzer. Fungal:bacterial ratio was determined using quantitative PCR (qPCR) after DNA extraction from the soil following Fierer et al. (2005). Collembola, oribatid mite, and mesostigmatid mite biomass were determined after Tullgren extraction into ethanol. Organisms were then sorted, counted, and estimates of biomass per individual, determined from the ¹³C tracking experiment, were used to determine total biomass per group. Bulk density at each site was determined after subtracting roots and stones from whole, unsieved, soil cores (8 cm dia., 0–7.5 cm depth) by hand. Note: we were also able to calculate total root biomass from bulk density determinations as well. Using bulk density we calculated, when appropriate, total mass in g m⁻² to 7.5 cm depth.

2.3. Data analysis

The effects of land cover, belowground pool identity, and their interaction on the absolute amount of glucose-C recovered were tested with a linear mixed effects model with site as a random effect. To investigate the relative partitioning of glucose-C, we standardized the amount recovered in each pool to the total recovered and assessed patterns with NMDS (nonmetric multidimensional scaling; Kruskal and Wish, 1978), an ordination technique, after converting data to a Euclidean distance matrix. The NMDS with the lowest stress value after 1000 random starts was used to assess relationships between environmental variables and the partitioning of glucose-C amongst sites by vector fitting (*P*-values based on 9999 permutations). Note, NMDS is

arbitrarily scaled with sites plotted so that they best maintain their rank order (i.e., sites that have similar partitioning of glucose-C will tend to be closer to each other on the NMDS). To further examine our main findings we conducted linear regression analyses and divergence analysis (Weber and Legge, 2009). The latter allowed us to reduce the similarity or dissimilarity in the partitioning of glucose-C into a single metric. This metric was derived by dividing the Euclidean distance of each site – from the mean of all sites – by the maximum distance giving values that range from 0 (100% similarity to the mean) to 1 (the most dissimilar from the mean). Regression analysis was then used to examine how dissimilarity was related to any significant vectors identified via vector fitting. All analyses were conducted using the freeware statistical package R (<http://cran.r-project.org/>) and results are considered significant at *P* < 0.05 and marginally significant at *P* < 0.10.

3. Results

Across both land covers most of the glucose-C was recovered in microbial biomass or SOC, and the least in microarthropods (Fig. 2). Overall, 8 to 21% of the glucose-C added was recovered with the remainder either below the 7.5 cm sampling depth or lost via lateral flow. Regardless, there was a significant interaction between land cover and pool identity on the absolute amount of glucose-C found in each belowground pool ($F_{6,24} = 2.63$; *P* < 0.05). This interaction seemed driven by significantly more glucose-C respired ($F_{1,4} = 10.28$; *P* < 0.05) – and marginally greater amounts in oribatid mites ($F_{1,4} = 6.08$; *P* = 0.07) – in pastures, with other pools showing no significant land cover effects (*P* > 0.27 in all cases).

Given that the absolute amount of glucose-C recovered across sites differed and was less than 100%, we also analyzed the proportion of glucose-C recovered in each pool as a way to standardize our investigation of the partitioning of the glucose-C within a site. Land cover (*P* = 0.49) and its interaction with pool identity (*P* = 0.95) was not significant but pool identity was (Fig. 3; $F_{6,24} = 64.13$; *P* < 0.0001). In line with this, NMDS did not suggest

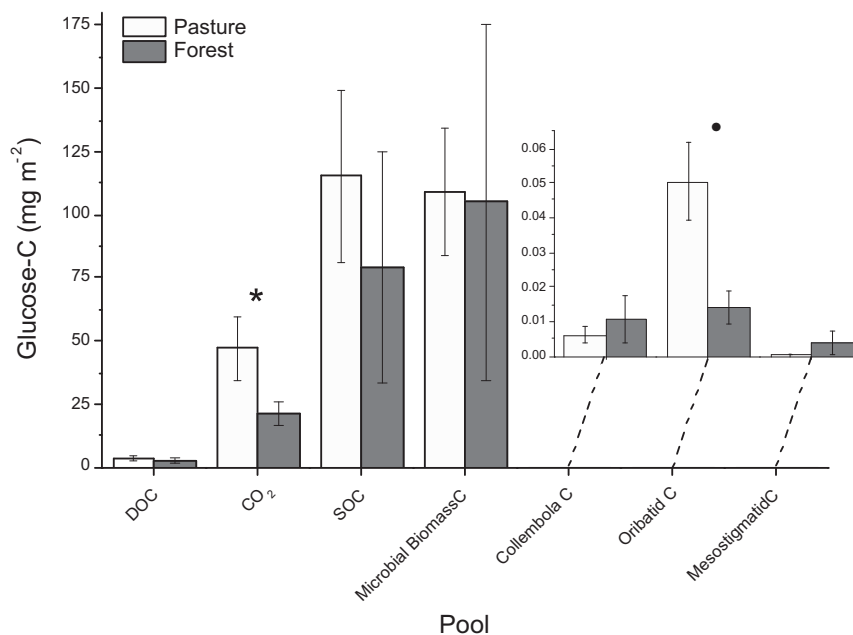


Fig. 2. The absolute amount (mg m⁻²) to a depth of 7.5 cm of glucose-derived ¹³C recovered (means ± s.e) in different C pools by land cover (pasture [*n* = 3] and forest [*n* = 3]). There was a significant interaction between land cover and pool identity ($F_{6,24} = 2.63$; *P* < 0.05). Within pools, significant (*P* < 0.05; denoted by *) and marginally significant (*P* < 0.10; denoted by •) land cover effects were observed for absolute amounts of glucose-C found in CO₂ ($F_{1,4} = 10.28$; *P* < 0.05) and oribatid mites ($F_{1,4} = 6.08$; *P* = 0.07), respectively.

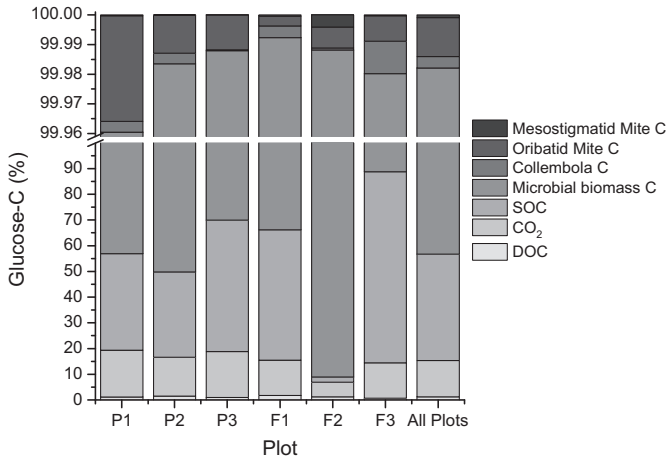


Fig. 3. The proportion of recovered glucose-derived ¹³C in belowground pools for each site. Proportions for microarthropods are magnified to ease interpretation. Land cover and its interaction with pool identity were not significant but pool identity was ($F_{6,24} = 64.13$; $P < 0.0001$). P1–P3 are pastures and F1–F3 are forests. Also shown is the mean proportion of recovered glucose-derived ¹³C in belowground pools across all sites (i.e., All Plots).

a clear land cover effect (Fig. 4). However, pastures did tend to cluster more closely than forests. Indeed, forest F1 was more similar to pastures, likely due to similar proportions of glucose-C found as CO₂, SOC and microbial biomass C between it and the pastures (Figs. 3 and 4). The other two forests were distinct, likely due to the large proportions of glucose-C found in the microbial biomass (F2) or SOC (F3; Figs. 3 and 4). To understand better what might be driving this variation across sites, we used vector analysis to look for relationships between the NMDS ordination and several soil physical, chemical, and biological properties.

The vector analysis of the NMDS ordination suggested that only mesostigmatid biomass was significantly related to the partitioning of glucose-C (Fig. 4; Table 2). Specifically, pastures and F1 tended to have lower mesostigmatid biomass than sites F3 and F2, and a greater proportion of glucose-C in DOC and CO₂. Further exploration revealed a marginally significant, positive relationship between mesostigmatid biomass and the proportion of glucose-C in mesostigmatids ($F_{1,4} = 5.57$; $P = 0.08$; $r^2 = 0.48$); and a marginally significant, negative relationship with the proportion respired ($F_{1,4} = 6.89$; $P = 0.06$; $r^2 = 0.54$). Also, the relative divergence in the partitioning of glucose-C amongst sites from the mean of all sites was positively related to total mesostigmatid mite biomass ($F_{1,4} = 108.12$; $P < 0.0001$; $r^2 = 0.96$). That is, sites with greater mesostigmatid biomass also tended to be more dissimilar in how glucose-C was partitioned (Fig. 4, inset).

4. Discussion

We sought to understand the short-term fate of glucose inputs belowground, and its potential controls. After only 72 h, we observed glucose-C in all of the measured belowground pools. In terms of absolute amount of glucose-C recovered, we found large amounts in the microbial biomass and SOC pools, intermediate amounts respired as CO₂, and small amounts in the DOC and microarthropods (Fig. 2). We also noted that the absolute amount of glucose-C found in these various pools was dependent on the interaction between land cover and the C pool.

Specifically, greater amounts of glucose-C were respired as CO₂ and recovered as oribatid mite biomass in the pasture sites, but amounts of glucose-C found in the other pools did not differ between forests and pastures. The increased respiration of glucose-C for these pasture sites versus the hardwood sites has already been reported (Strickland et al., 2010), and was attributed to higher levels of soil P, which when limiting can reduce microbial activity (Cleveland and Townsend, 2006). The marginally greater absolute amount of glucose-C recovered in oribatid mites in pastures may indicate that C flow is more rapid in this system. Lending support to this possibility is the observation that more glucose-C was respired in the pastures compared to the forests, yet glucose-C recovered in microbial biomass was not significantly different between land

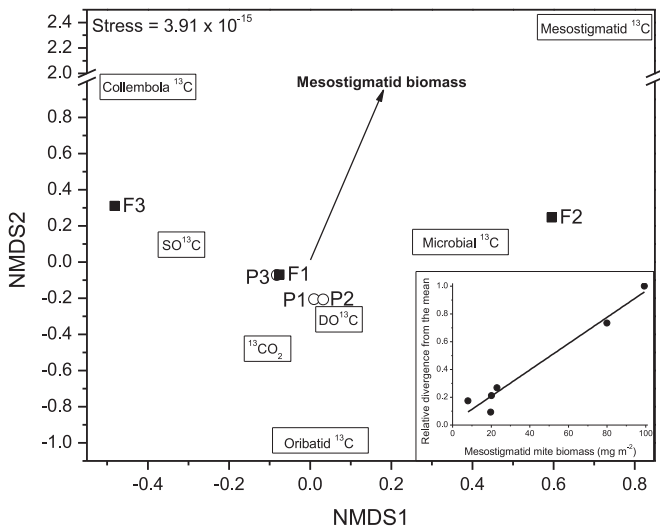


Fig. 4. NMDS ordination examining the partitioning of glucose-C across sites. Pools of glucose-C are represented by boxed labels. The closer a site is to the label for a specific pool (i.e., ¹³CO₂, SO¹³C, DO¹³C, Microbial biomass ¹³C, Oribatid ¹³C, Collembola ¹³C, or Mesostigmatid ¹³C) the more likely it is to have a higher proportion of ¹³C in that pool than are other sites. The arrow represents the only significant vector ($P < 0.001$; $r^2 = 0.97$) – mesostigmatid mite biomass (g m^{-2}) – related to the ordination. The inset shows the relationship ($F_{1,4} = 108.12$; $r^2 = 0.96$; $P < 0.0001$) between relative divergence in the partitioning of glucose-C from the mean of all sites and mesostigmatid mite biomass (mg m^{-2}). The closer a site value is to zero the more similar it is to the mean, with a value of one denoting the site most dissimilar from the mean.

Table 2

Results of vector analysis examining the relationship between environmental factors and NMDS results (Fig. 4) for the partitioning of glucose-C. Units for all environmental variables except fungal:bacterial ratio (unitless), pH, SIR (reported as $\text{g m}^{-2} \text{h}^{-1}$), mineralizable C (reported as $\text{g m}^{-2} \text{h}^{-1}$), soil temperature (reported as °C), and soil moisture (reported as %) are in g m^{-2} . Only mesostigmatid mite biomass (shown in bold) was significant.

Vector	r ²	P-value
Fungal:bacterial ratio	0.22	0.57
Total SOC	0.95	0.18
Total SON	0.38	0.48
P	0.49	0.26
pH	0.72	0.23
SIR	0.41	0.46
Mineralizable C	0.12	0.85
Microbial biomass C	0.75	0.12
Microbial biomass N	0.10	0.76
DOC	0.62	0.20
DON	0.61	0.32
NO ₃	0.12	0.65
NH ₄	0.22	0.46
Soil temperature	0.45	0.36
Soil moisture	0.64	0.18
Roots	0.26	0.53
Collembola biomass	0.44	0.45
Oribatid mite biomass	0.43	0.41
Mesostigmatid mite biomass	0.97	<0.001

covers. If there is more rapid C flow through the microbes in the pasture sites, there may be a number of possible explanations as to why more C was recovered in the oribatid mites. First, a more active microbial biomass – given higher P in the pastures – might stimulate microarthropod feeding (Mamilov et al., 2001; Lennon and Jones, 2011). Alternatively or in addition, higher feeding rates by oribatids might stimulate turnover of the microbes and hence their respiration (Coleman et al., 1977; Seastedt, 1984). Lastly, the lower predatory mite biomass in pastures might permit the glucose-derived C to accumulate in oribatid mites feeding on the microbes (Hunt et al., 1987; Wardle, 2002; Schmitz, 2010). Indeed, marginally lower ($P < 0.10$) glucose-C was recovered in the oribatid mites of the forest sites while, on average, a greater amount was recovered in the mesostigmatid mites for those same sites.

None of the proposed mechanisms above are mutually exclusive, and clearly there needs to be more work on trophic interactions within belowground foodwebs to explain differences in the absolute amounts of glucose-C recovered in the different pools we resolved. Given that glucose is a dominant component of rhizodeposits (Grayston et al., 1996; van Hees et al., 2005), that rhizodeposition may fuel a substantial fraction of soil foodweb C demands (van Hees et al., 2005), and that rhizodeposition rates are expected to increase with elevated CO_2 (Phillips et al., 2011), this is an important research need if we are to understand C dynamics in soils.

Given that the total amount of glucose-C recovered varied across sites, we examined the proportion of glucose-C found in the pools to understand partitioning belowground independent of absolute mass recovered (Fig. 3). As previously observed (Boddy et al., 2007; Fischer et al., 2010), a large proportion (~86%) of the recovered LMWCC was found belowground, with ~14% respired. Only ~1% was recovered in DOC, which agrees with the expectation that LMWCCs are rapidly removed from this pool by microorganisms (Saggar et al., 1999; van Hees et al., 2005; Boddy et al., 2007; Högberg et al., 2008, 2010). The large proportion (~43%) of glucose-C recovered in microbial biomass supports this expectation. A large proportion (~41%) of glucose-C was also associated with SOC. This may have been driven by sorption of glucose (a polar molecule) to soil surfaces or sorption of microbial-derived metabolites, fates that again are congruent with rapid removal of LMWCCs from DOC (Saggar et al., 1999; Fischer et al., 2010).

Most surprising, given the 72-h timescale of our study, was the recovery of glucose-C in higher trophic levels (i.e., microarthropods). Glucose-C most likely flowed to higher trophic levels through consumption of microbial biomass by microbivores (i.e., Collembola and oribatids), and subsequent predation on microbivores by mesostigmatid mites (Fig. 1). Although a small proportion (~0.02%) of glucose-C was recovered in the animals, our work demonstrates the potential for LMWCCs to be a pathway through which recent photosynthate propagates rapidly through belowground foodwebs (Högberg and Read, 2006; Wu et al., 2009; Högberg et al., 2010). Given that microarthropods can be an important food source for aboveground predators such as spiders and salamanders, this pathway may also provide a bridge for rapid return of recent photosynthate to aboveground foodwebs, but its significance for aboveground energy flow remains to be evaluated. Notably, there was marked variation across sites in the partitioning of glucose-C. To understand what drove this variation we examined the relationship between the partitioning of glucose-C and soil chemical, physical, and biological factors across sites.

The only variable significantly related to glucose-C partitioning across sites was the biomass of predatory mites (Table 2). The relationship likely occurs because of greater similarity in the partitioning of glucose-C for sites that had lower mesostigmatid mite biomass (i.e., P1–P3, and F1), when compared to sites with more mesostigmatid biomass (i.e., F2 and F3) (Fig. 4). Divergence analysis

supported this inference, given the positive relationship between divergence (i.e., the difference between a given site and the mean of all sites) in the partitioning of glucose-C and mesostigmatid biomass (Fig. 4 inset). Notably, higher mesostigmatid biomass was associated with lower proportions of glucose-C respired and greater proportions remaining belowground.

The potential mechanisms that might explain this result can likely be derived from the wealth of research examining the role of belowground communities in regulating ecosystem processes (Seastedt, 1984; Hunt and Wall, 2002; Coleman, 2008). For example, the potential influence of microarthropods on litter decomposition rates has been well established (Seastedt, 1984), although it is often context dependent being influenced by factors such as climate and land management (e.g., Wall et al., 2008; Ayres et al., 2010; Wickings and Grandy, 2011). The most plausible explanation in our system is that higher predatory mite biomass limits the oribatid mites and hence the accumulation of the glucose-C in these microbivores. If microbivores stimulate microbial activity (Coleman et al., 1977; Petersen and Luxton, 1982; Seastedt, 1984) and hence respiration, then where there is lower oribatid activity then there is also a reduction in glucose-C loss as CO_2 . This differs from a regular trophic cascade, where suppression of the second trophic level by the third, permits higher activity of the basal trophic level (Oksanen et al., 1981; Wardle and Yeates, 1993) but microbivore feeding is well known to stimulate microbial activity (Petersen and Luxton, 1982). Together these possibilities (in addition to many others) may indicate why trophic cascades in soils are typically weak or nonexistent (Wardle and Yeates, 1993; Wardle, 2002). Further research is required to test this explanation, but what is well known is that predators play an integrative role in highly diverse, belowground foodwebs, with multiple organisms being consumed by them (Hunt et al., 1987; Nielsen et al., 2011). This means that increases in predator abundance or biomass, and the concomitant effects on prey density and prey species identity (and any potential ensuing trophic cascade), may lead to different outcomes (Pace et al. 1999; Fox, 2007). These effects – as well as those that might arise through prey behavioral and physiological changes in the presence of a predator (Hawlena and Schmitz, 2010a,b; Schmitz, 2010) – can certainly influence rates of C and N cycling. Of course, the alternative is that the mesostigmatid mites are simply responding to multiple environmental factors and are not the drivers of change in the partitioning of glucose-C. To clarify the role of predators on the flux and partitioning of LMWCCs in belowground foodwebs, more work is necessary that empirically examines the effects of trophic structure in belowground communities.

In conclusion, we have shown that a representative LMWCC is partitioned across multiple belowground C pools in a relatively short period of time and that the bulk of C derived from this compound remained belowground. Furthermore, we show that LMWCCs have the potential to propagate through soil foodwebs rapidly, and that the partitioning belowground of these compounds may be determined by higher trophic levels, such as predatory mites. Yet, a better understanding of the role of trophic structure in determining ecosystem processes, such as the partitioning of LMWCCs, is needed. Additionally a fundamental understanding of the interactions amongst different exudates and soil organisms is unknown but may be of great importance especially given that LMWCC inputs may fuel much of belowground heterotrophic activity, and are likely to change in magnitude, composition, and quality with environmental change (Chapin et al., 2009; Ostle et al., 2009).

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