Geoderma 148 (2008) 113–119

Contents lists available at ScienceDirect

Geoderma

journal homepage: www.elsevier.com/locate/geoderma

Preferential sequestration of microbial carbon in subsoils of a glacial-landscape toposequence, Dane County, WI, USA

Chao Liang [a,b,*], Teri C. Balser [a]

[a] Department of Soil Science, University of Wisconsin-Madison, WI, 53706, USA
[b] Great Lakes Bioenergy Research Center (GLBRC), Madison, WI, 53706, USA

ARTICLE INFO

Article history:
Received 19 August 2008
Accepted 15 September 2008
Available online 22 October 2008

Keywords:
Amino sugar
Microbial residue
Soil organic carbon
Carbon storage
Soil profile

ABSTRACT

Microorganisms participate in soil carbon storage by contributing biomass in the form of refractory microbial cell components. However, despite the important contribution of microbial biomass residues to the stable carbon pool, little is known about how the contribution of these residues to soil carbon storage varies as a function of depth. In this study, we evaluated microbial residue biomarkers (amino sugars) in varied pedogenic horizons from six soil profiles of two geographic sites on a glacial-landscape toposequence in Dane County, WI. We found that the amino sugars appeared to preferentially accumulate in subsoil. Specifically, although total amounts of amino sugars decreased downward through the profile as even as total organic carbon did, the rate of decrease was significantly lower, suggesting that these compounds are more refractory than general soil organic carbon. The proportion of amino sugars to soil organic carbon increased along the depth gradient (from top to bottom), with the exception of Bg horizons associated with high water tables. We also observed that microbial residue patterns measured by amino sugar ratio (e.g., glucosamine to muramic acid) showed different dynamic tendencies in the two different geographic sites, suggesting that residue carbon contribution by fungi and bacteria is likely site-specific and complex. In summary, regardless of the redox microenvironment created by groundwater dynamics in a given soil, our study supports the hypothesis that microbial residues are refractory and that they contribute to terrestrial carbon sequestration.

1. Introduction

Interest in the transformation and sequestration of soil organic carbon (SOC) has increased substantially in recent years due to its importance in global carbon (C) cycling. Soil carbon is the major reservoir of C in terrestrial ecosystems, storing approximately 1600 Pg (1 Pg=10^{15} g) of organic C (Eswaran et al., 1993)—slightly more than twice as much C as is stored as CO_2 in the atmosphere. Dynamics of the terrestrial C pool are heavily influenced by the catabolic and anabolic activity of microbial communities (Balser, 2005; Kandeler et al., 2005), and synthesized microbial metabolites have further been suggested as part of the stable C pool (Kiem and Kögel-Knabner, 2003; Kindler et al., 2006; von Lützow et al., 2006; Simpson et al., 2007). Because the stable C pool is highly relevant for the role of soils as a terrestrial C sink within the global C cycle (Falloon and Smith, 2000), the long-term sequestration of C in soil may thus be related to senesced microbial residues.

The depth distribution of both microbial biomass and SOC is asymmetric. Batjes (1996) estimated that more than 50% of the organic C contained in a 1-meter soil profile was in deeper (>25 cm) soil horizons. Despite this confirmed C sink and thus correspondingly large C stabilization potential (Lorenz and Lal, 2005), detailed understanding of C dynamics in subsoils has eluded researchers. In the study of terrestrial C dynamics, the soil C pool is typically divided into two fractions based on different turnover rates: the labile pool and the stable pool. In the deeper layer, the stable C pool of progressively more recalcitrant compounds is enriched due to utilization of the labile C pool by microorganisms as well as the rare replacement of fresh organic matter (Fontaine et al., 2007). Therefore, the SOC pool of the subsoil is relatively enriched in stable compounds compared with the surface soil. To understand the nature and function of SOC, it is important to determine whether it is plant- or microbial-derived, especially in subsoils containing large amounts of stabilized organic C of as-yet-unknown origin.

Research on pedogenesis and variation in soil C has been conducted through the soil depth profile (Almond and Tonkin, 1999; Jobbagy and Jackson, 2000), with more recent focus on variation in microbial biomass and communities with depth (Fritze

* Corresponding author. Department of Soil Science, University of Wisconsin-Madison, WI, 53706, USA. Tel.: +1 608 265 4850; fax: +1 608 265 2595.
E-mail address: chaoliang@wisc.edu (C. Liang).

0016-7061/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.geoderma.2008.09.012

et al., 2000; Ekelund et al., 2001; Taylor et al., 2002; Fierer et al., 2003; Allison et al., 2007). Barring few exceptions (e.g. spodosols), organic C (and correspondingly, the biomass of its decomposers) generally decreases with depth in mineral soil (Jobbagy and Jackson, 2000). The change in C substrate induces different abundance of specific microbial groups in different strata (Fierer et al., 2003). However, little is known about the contribution of microbial residues to SOC storage within and between individual mineral horizons of soil depth profiles.

Studies of biomarker molecules may be most appropriate to determine the microbial origin of SOC. Soil monosaccharide ratios have traditionally been used to assess the relative contribution of microbes or plants to carbohydrate composition of SOC, since microorganisms primarily synthesize hexoses whereas pentoses are important components in plant cells (Murayama, 1984; Oades, 1984). However, this has led to varied sugar-ratio choices among research groups (Puget et al., 1998; Angers and Mehuys, 1990; Jolivet et al., 2006). Further, all of these ratios assess relative contribution by microbes over plant-derived sources, rather than quantify the specific microbial contribution to SOC. Additionally, hexose/pentose ratios can't separate compounds originating from different microbial groups. Alternatively, amino sugars can be used for this purpose. The concentration of amino sugars has often been used as an indicator for microbially sequestered C (Zhang et al., 1998; Rodionov et al., 2001; Solomon et al., 2001; Amelung et al., 2002; Liang et al., 2007a,c). Amino sugars are useful this way because of their absence in plants (Parsons, 1981), and their stability against fluctuations in enzyme activities and living microbial biomass (Nannipieri et al., 1979; Guggenberger et al., 1999). It has been shown that amino sugars are highly stabilized in soils and persist after cell death (Glaser et al., 2004; Niggemann and Schubert, 2006). In addition, different amino sugars are derived from different groups of soil microorganisms (Sowden and Ivarson, 1974; Parsons, 1981; Amelung, 2001), so the ratios of glucosamine to galactosamine and glucosamine to muramic acid can, for instance, serve as useful indicators for relative fungal and bacterial contribution to SOC (Zhang et al., 1998; Solomon et al., 2001; Liang et al., 2007a). A recent study by Liang et al. (2007b) showed that the dynamics of galactosamine are different from muramic acid, and thus suggested that both ratios (glucosamine to galactosamine and glucosamine to muramic acid) be recognized as to belonging to two evaluation systems and serving different goals. In the meantime, some reports suggested that fungi contribute larger percentages of galactosamine than bacteria, contradicting the traditional consensus stated in several publications (Engelking et al., 2007).

In this study, we investigated microbial residue (amino sugars) distribution through six soil profiles of two varied geographic sites on a glacial-landscape toposequence in Dane County, WI. To characterize the vertical distribution of amino sugars, different pedogenic horizons from soil profiles were selected. Our objective was to assess whether long-term pedogenic processes led to

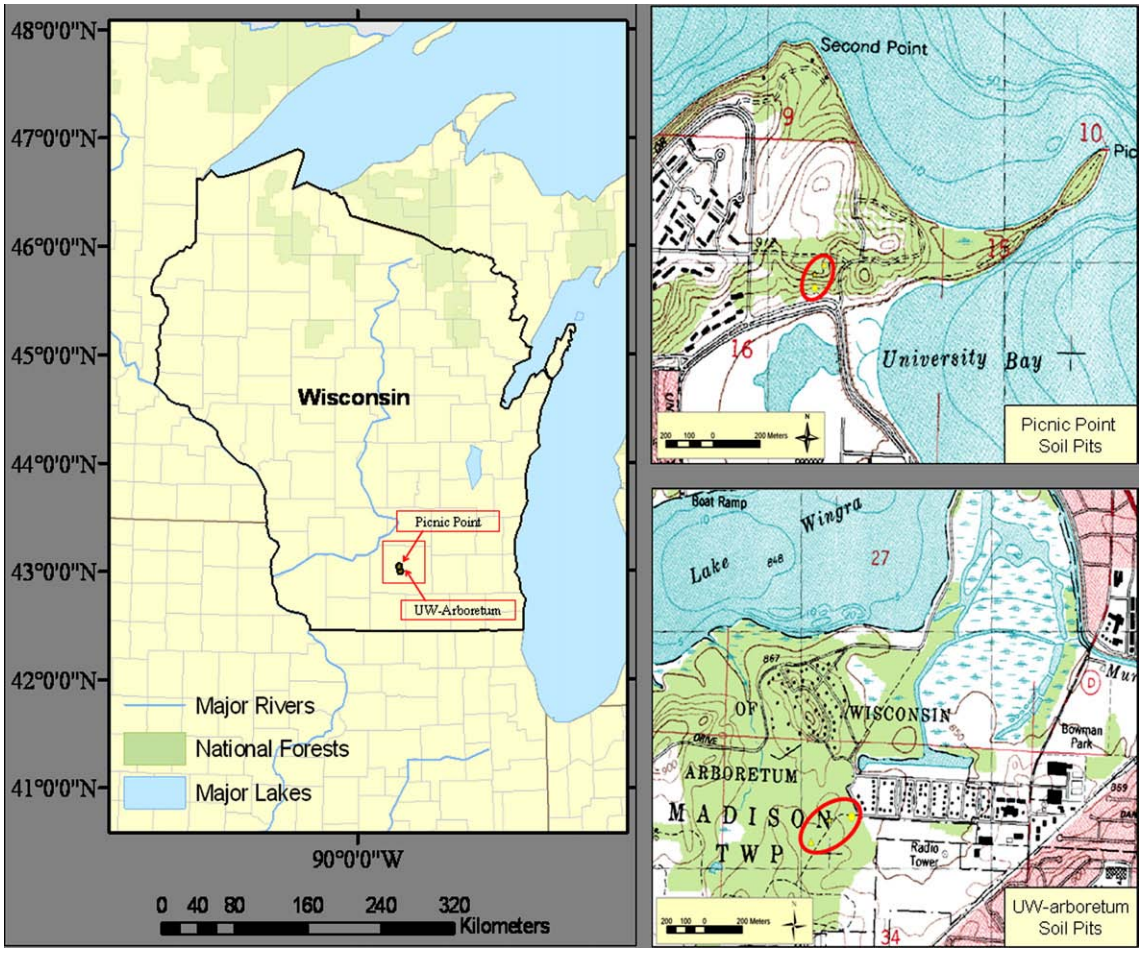

Fig. 1. Location map of Wisconsin (USA) and contour map of two geographic study sites in its Dane County. Yellow points within red circles represent the soil pits. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

different amounts of microbially sequestered C in the surface soil and subsoil.

2. Materials and methods

2.1. Description of area studied

We sampled soils from two geographic areas near the campus of the University of Wisconsin, Madison: the UW-Arboretum (Arboretum of University of Wisconsin, Madison) and Picnic Point (Fig. 1). Both are located in Madison Valley of Dane County, WI, U.S.A., within Township 7 North, Range 9 East of the 4th Principal Meridian (T7 N, R9E, 4th PM). The bedrock geology of the UW-Arboretum and Picnic Point is primarily Precambrian igneous and metamorphic rock overlain by Paleozoic sedimentary rocks (Clayton and Attig, 1997).

2.1.1. UW-Arboretum site

The UW-Arboretum site is known as the Lost City Forest, named for residential development that was planned and commenced in the early 1900s but never completed. It is dominated by native oaks (*Quercus* spp.) and overgrown with exotic species such as honeysuckle (*Lonicera* spp.) and common buckthorn (*Rhamnus cathartica*).

Three pits (Pit 1, Pit 2 and Pit 3) at approximately 262 m elevation and 240 meter apart (ca. 43°02′ N, 89°25′ W) were chosen for sampling. Soils from all three pits had a mesic temperature regime, while Pit 1 had an aquic and Pits 2 and 3 an udic moisture regime due to different flow regimes of Lake Wingra at different depths. Before 1900, Lake Wingra was approximately 0.3 m deeper than it is today (Kline, 1992). Pit 1 is formed from the historic lacustrine basin sediments of Lake Wingra, with a seasonal high water table indicated by the existence of gleyed horizons and redoximorphic features. Pit 2 is formed from loess layered over ice-contact stratified material and is free of influence from the water table in most years. Pit 3 is formed by ice-contact stratified materials but positioned in a micro-topographic low. The apparent differences among these three soil profiles are due to the parent materials deposited by the different flow regimes of the lake at different depth levels and their impact on topographic development of the area.

2.1.2. Picnic Point site

Following the most recent Wisconsin glaciation (14,000–8500 BC), the Picnic Point site was an oak savanna (100 cm annual precipitation, 16–24 °C). Beginning with European settlement a large portion of this site was logged, used for livestock grazing, and eventually made into a protected natural area. Presently, Picnic Point vegetation can be described as a secondary regrowth deciduous forest, dominated by hackberry (*Celtis occidentalis*), cherry (*Prunus* spp.), hickory (*Carya* spp.), and oak (*Quercus* spp.) along with a number of invasive species such as honeysuckle (*Lonicera* spp.) and buckthorn (*Rhamnus* spp.). Understory species include trout lily (*Erythronium americanum*), wild geranium (*Geranium merculatum*), Virginia creeper (*Parthenocissus quinquefolia*), white snakeroot (*Eupatorium rugosum*), Solomon's seal (*Polygonatum* spp.), false Solomon's seal (*Smilacina racemosa*), and enchanter's nightshade (*Circaea lutetiana*).

Three pits (here designated as Pits 4, 5 and 6) were chosen from west of this site through a slope that rises to the Eagle Heights campus residential area. The pits were located in the lower, upper and middle slope, respectively, approximately 84 meter apart (ca. 43°05′ N, 89°25′ W), and all formed from loess deposited over glacial till due to ground moraine deposition. Soils from three pits of Picnic Point have an udic moisture regime and a mesic temperature regime. Pit 4 is on a 6% foot-slope of the hillside and has redoximorphic features within the deeper Bt horizon. Pit 5 displays the shallowest soil profile and is located at the highest elevation, with a 20% steep shoulder grade. The loess layer of Pit 5 shows significant signs of erosion, which result in an absence of E horizon development. Pit 6 is located on the northeast slope with an 11% gradient back-slope. The soil profile of Pit 6 can be considered stable, with relatively higher soil organic matter (SOM) accumulation compared with pits on the south-facing slope. The development of these three soil profiles is heavily influenced by topographic factors including slope shape, aspect and position in the landscape.

2.2. Soil sampling

The six pits were originally excavated and maintained as models for pedology education at the Department of Soil Science, University of Wisconsin-Madison. An evaluation of the soil profiles at these two study areas (the UW-Arboretum and Picnic Point) illustrates the great impact which soil-forming factors can have on soils located even within close geographic proximity. Due to the impact of soil-forming factors even at small scales and the threat of overly disturbing the sites, we did not dig replicate pits.

Soil was sampled in October, 2004. Firstly, the pits were redug inward >30 cm, then the trench wall of every pit was scraped inward at least 5 cm using a clean knife. Sampling was based on pedogenetic horizons. Three replicates of each horizon were collected from each profile by digging vertically along different lines within the same trench wall. After sampling, soil specimens were immediately transported back to the laboratory. We mixed the three replicates without sieving, removed visible root and stones, freeze-dried and homogenized the samples, and then stored them at −20 °C until analysis. Descriptive properties of the soil horizons are shown in Appendix A.

2.3. Laboratory analysis

The determination of four amino sugars was conducted by GC after their conversion to aldononitrile acetates according to Guerrant and Moss (1984). The method is described in detail by Zhang and Amelung (1996). Briefly, samples were hydrolyzed

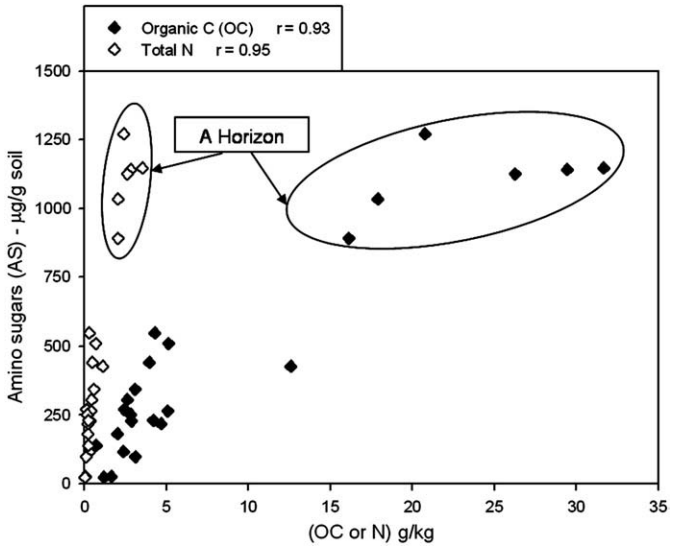

Fig. 2. Relationship between amino sugar contents in soils and organic C or total N. Data from all horizons.

with 6 M HCl at 105 °C for 8 h, and the solution was filtered and purified by neutralization. After drying of the supernatant, methanol was used to wash amino sugars out from the residues, and amino sugars were then transformed into aldononitril derivatives. Excess anhydride was destroyed with water and 1 M HCl before the amino sugar derivatives were extracted from the aqueous solution with dichloromethane. GC separation of the amino sugar derivatives was carried out on an Agilent 6890A (Agilent Tech. Co., USA) equipped with an HP-5 (25 m by 0.33 mm by 0.25 μm) fused silica column and FID. The individual amino sugar derivatives were identified by comparing their retention time with those of authentic standards. Quantification was gained relative to the internal standard myo-inositol, which was added to the samples prior to purification, and the recovery standard methyl-glucamine was added before derivatization. In addition to the individual amino sugar contents, we calculated the total amino sugar contents as the sum of four amino sugars determined, glucosamine (GluN), galactosamine (GalN), muramic acid (MurA) and mannosamine (ManN). Since MurA was used as the biomarker for bacterial cell wall residues, GluN approximately for fungal cell wall residues, in this study the GluN/MurA ratio was used to indicate relative fungal and bacterial contributions to soil SOC.

Carbon and nitrogen concentration were analyzed after dry combustion with a LECO total CNS analyzer (LECO Corporation, St. Joseph, MI).

2.4. Statistical analyses

Linear regressions were employed to model relationships between total amino sugar amounts in soils and either organic C or total N, with correlation coefficients (R^2) indicating goodness of fit. Statistical analyses were performed with the SPSS (SYSTAT Software, Inc.) software for Windows, and regression analyses and figure preparations were accomplished using Sigma Plot (SYSTAT Software, Inc.).

3. Results and discussion

Total amino sugar contents had a positive correlation with SOC content (r=0.93), as well as with total soil N (r=0.95). As shown in Fig. 2, amino sugar amounts are enriched when sufficient organic C or total N is available. In comparison with their respective sub-horizons, the A horizons of 6 pits all contained markedly more organic C and total N. This is not surprising, as the A horizon is continuously

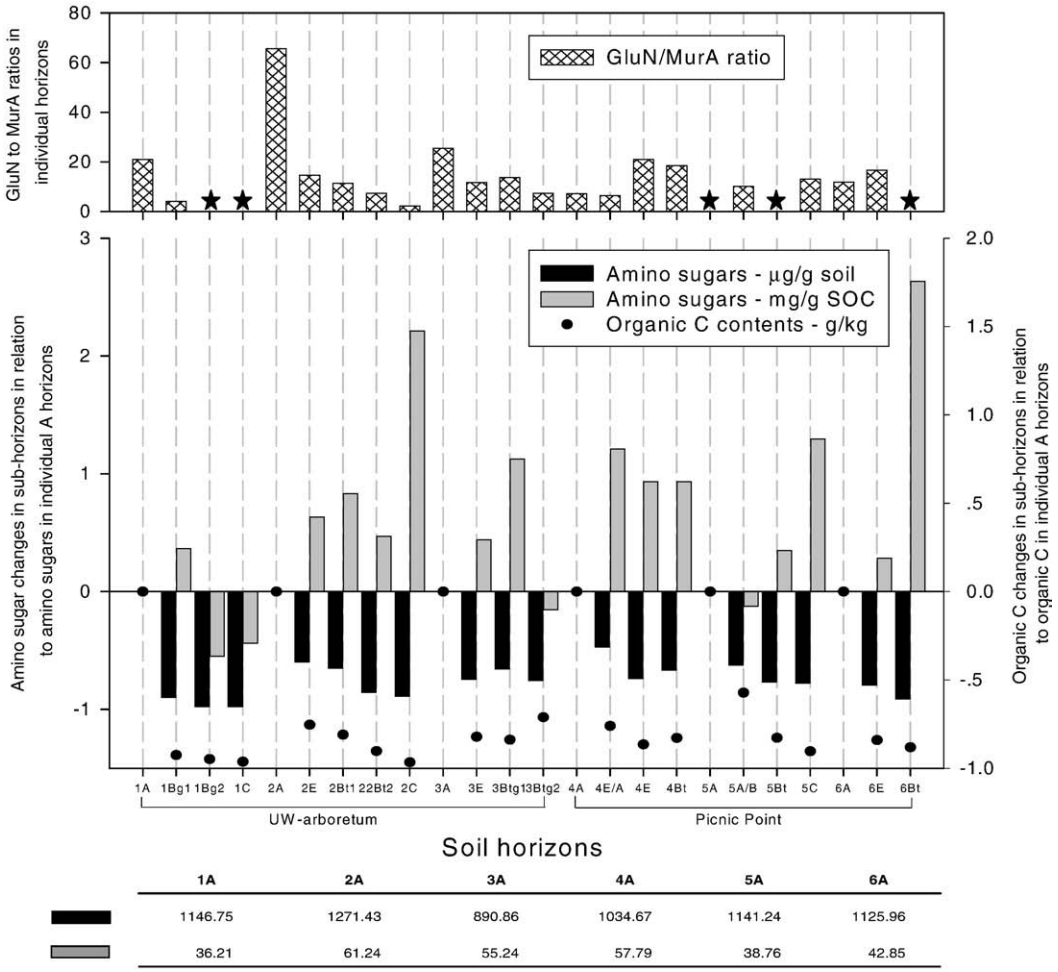

Fig. 3. GluN/MurA ratios in each horizons and changes in amino sugars and organic C in sub-horizons compared to those in respective A horizons of six pits in the UW-arboretum and Picnic Point, Wisconsin, USA. The pit numbers was put in front of pedogenic letter. Star symbols represented MurA contents below the detection limit (cannot be calculated).

fed with the organic inputs from plant litter and exudates from the dense root system. Differences in nutrient availability cause varied microbial biomass in the soil profile, and thus varied microbial residues (amino sugars). In this study, organic C amounts were significantly correlated (R^2 = 0.97) with total N. Although soil total N is more strongly correlated to amino sugar amounts than organic C is, we pay less attention to N due to the more complex mechanisms of N dynamics in soils, as well as inorganic N that exists without quantitatively distinguishing from organic N type in this study. Our interest here is more on how to assess the contribution of microbial amino sugars to SOC storage.

In order to compare the depth distribution of both SOC and amino sugars independently of total SOC and amino sugar contents, we calculated the relative proportions for each horizon by dividing changes in SOC and amino sugars between each individual horizon and the A horizon by SOC and amino sugar contents of the A horizon. Total amino sugar and organic C contents in soils generally decreased with depth in each site (Fig. 3). Exceptions to the reported trend occurred at 3Bt and 4Bt, where the total amino sugar and organic C contents both were higher than that of their respective E horizons. We think the explanation lies in noting the proximity of these horizons to the water table; the B horizons of Pit 3 and Pit 4 contain prominent redoximorphic features, indicating seasonal water table fluctuations (Appendix A). The level of the water table can affect the distribution of aerobic and anaerobic microorganisms (Sundh et al., 1997). Organic matter decomposition would be hampered by anaerobic conditions when the soil is saturated. Past researchers have found that microbial biomass generally declines with increasing depth (Fritze et al., 2000; Taylor et al., 2002; Fierer et al., 2003; Allison et al., 2007), while total microbial biomass C could increase with increasing water table levels (Morris et al., 2004). We suspect that, in regions with fluctuating water tables, microbial communities might shift to dominant species with thicker cell walls able to withstand changing water potentials (such as Gram-positive bacteria), or with reduced utilization of their metabolically produced biomass components (amino sugars). Alternately, the enriched SOC in the B horizon of Pit 4 may also have been caused by clay translocation downward (which commonly occurs at the lower slope positions). In this study, the pattern of the total amino sugars was strikingly similar to the organic C content in each profile (Fig. 3). This supports our previous finding that the ability of SOC to sustain amino sugars was relatively constant (Liang et al., 2007c). Nevertheless, 3Bt showed a contrasting dynamic regarding total amino sugar and organic C content. This might be explained by the root size distribution; in comparison with 3Btg1 horizon, 3Btg2 was enriched with very fine roots which could not be fully separated out, thus contributing to organic C in our analysis.

The lower microbial biomass in subsoils might result in a generally decreasing accumulation of absolute amino sugars with increasing depth. But when considering the proportion of amino sugars within soil organic C, a different picture is obtained. Compared with the A horizon, the proportions of amino sugars in subsoils *increased* on the whole (with the exception of 1Bg2 and 3Btg2) (Fig. 3). Pits 1 and 3 are both located in the UW-Arboretum and spend time seasonally below the water table. Despite this, we see that amino sugar proportions in SOC generally increased with soil depth, likely indicating increasing microbial contribution to organic C storage. This phenomenon almost certainly results from the loss of non-microbial-derived organic C more rapidly than that of microbe-derived amino sugars with depth, and can be assumed due to progressive decay of plant-derived organic materials or conservative accumulation of amino sugars by the degrading microbes, or both. From combined structural and isotopic analyses of microbial lipids, there has been an evidence that not all available carbon in soil depth profiles is equally utilized by soil microbes (Kramer and Gleixner, 2008). In addition, preferential leaching of microbial cell materials compared with other soluble organic compounds might be another explanation. For this, there are dissenting opinions: Lowe (1983) asserted that amino and amide-N group are bonded in clay-organic complexes, and therefore accumulate in soils through stabilization by clay particles, whereas Kaiser et al. (2004) found that amino sugars in forest sites are inclined to be transported downwards via precipitation. In the study by Kaiser et al. (2004), the in situ hydrable amino sugars were determined by collecting 7 or 14-day discharge water. However, their sampling strategy doesn't preclude newly-bio-synthesized cell walls during the collection interval, and may not accurately reflect in situ amino sugars contents. In contrast, we have found that the concentrations of dissolved amino sugars in agricultural soils are below detection limits using the protocol of Zhang and Amelung (1996), which might indicate that amino sugars do not exist in a free, water-soluble form in these soils owing to their general incorporation into insoluble structural polymer matrices within a conglomeration (per Zhang, personal communication).

While the interpretation of amino sugar ratios is limited by potentially different turnover times and ambiguous origins, GluN/GalN and GluN/MurA have been widely used to trace how fungal and bacterial groups contribute to soil organic matter. In view of the unique origin of bacterial MurA, we used the GluN/MurA ratio in this study to quantify the relative contribution of fungal to bacterial residues to SOC with depth (Fig. 3). At the UW-Arboretum site, GluN/MurA ratios decreased as a function of depth, indicating bacterial contribution to deeper SOC relatively more than to surface SOC in forest soils. In contrast, the GluN/MurA ratio trend at the Picnic Point site was distinct from that in the UW-Arboretum. In particular, MurA of 5A was not detected in contrast to other surface A horizons. It is likely that Pit 5 may have had an E horizon before erosional events that stripped away the upper layers, and ultimately led to a missing E horizon and shallow Bt horizon. The slope steepness at this pit is in fact conducive to surficial erosion of the original loess cover. Such intense weathering could significantly redistribute or inhibit the growth of microbial communities. The distinct depth trends of GluN/MurA in the two different geographic sites suggest microbial contribution by fungi and bacteria is likely site-specific and complex.

In conclusion, we analyzed 6 pits located in 2 geographic sites containing of pedogenetic gradients in amount and pattern of microbial residues (amino sugars) with depth, and reached three main conclusions. First, each pit had an A horizon enriched with abundant amino sugars and SOM, associated with the highest organic input from plant litters and root exudates, in contrast to the deeper portions of the profiles. Second, in spite of the redox microenvironment created by the water table, microbial amino sugars preferentially accumulated in subsoils compared to general SOC. Third, amino sugar ratios alone are not sufficient to elucidate the mechanisms through which bacteria and fungi contribute to C storage, as the contribution is site-specific and subject to history factors influencing the trajectory of microbial community development.

Acknowledgements

This project was funded by USDA-CSREES Hatch Act. The authors thank Ben LaBarre, Miguel Morales, Michael Upthegrove and Sheri Young for assistance with field work, Harry Read and Ibrahim Saeed for assistance with instrumentation, Cynthia Stiles, Krista Stensvold, Zhuo Zhang and Ryosuke Fujinuma for helpful discussions, and Meredith Schuman for editorial proofreading on this manuscript.

Appendix A. Selected properties of the six studied soil profiles according to the Field Book for Describing and Sampling Soils (Version 2.0, 2002)

Horizons		Depth (cm)	Matrix: moist	Mottles: size-contrast-color	Texture	Structure: grade-size-type	Consistence moist-wet	pH	Root: number/size	Boundary
PIT-1										
1A	A	0–30	10YR 2/1		SICL	2-M-ABK	SH/SS-MS/MP	6.6	3F/3M/ 2C	AS
1Bg1	Bg1	30–50	(95%)G1 5/1 Y (5%)10YR2/1	1-P-5YR 5/8 (5% F3 M)	SIC	2-CO-ABK	SH/SS/SP	8.7	1F/2M	CS
	Bg2	50–70	G1 5/1 Y	1-D-10YR 7/8 (5% FMM)	SIC	2-VC-ABK	SH/SS/MP	8.6	1M	GW
1Bg2	Bg3	70–90	(95%)G1 7/5GY (5%)10 YR 8/2		SIC 2″S-lens	3-VC-SBK	SH/SS/MP	8.6	1F	CS
	Bg4	90–120	(90%)10 YR 8/2 (10%)G1 4/10GY		LS	3-VC-SBK	FR/SS/PO	8.5		AS
1C	C	120+	10 YR 5/8		S	0-VF-SGR	LS/SO/PO	8.5		AS
PIT-2										
2A	A	0–10	10 YR 2/2		SIL	1-F/M-GR	FR/SS/PO	6.4	3VF/2F	AW
2E	E	10–22	(90%)10 YR 4/4 (10%)10 YR 3/1		SICL	2-F-ABK	S/VFR/SS/SP	5.6	2M/1F	GS
2Bt1	Bt1	22–42	(70%)10 YR 3/4 (30%)10 YR 3/4		SIC	1-VF/F-SBK	S-FR/SS/MP	5.6	2M/1F	GS
	Bt2	42–60	10 YR 5/4		SIC	1-M-GR	S-FR/SS/MP	5.5	1F/2M/3C	GS
22Bt2	2Bt3	60–80	10 YR 4/6		SC	2-F/M-ABK	MH-FI/SS/MP	5.8	1VF	GS
	2Bt4	80–115	10 YR 4/6		SCL	2-F-ABK	SH-FI/SS/MP	6.1	2M	GS
2C	C	115–150	10 YR 4/6		LS	SG	S-VFR/SO/PO	8.3		
PIT-3										
3A	A	0–20	10 YR 2/1		SCL	1-M-SBK	FI/SS/MP	4.86	1VF/2M	GW
3E	E	20–40	10 YR 4/4		SC	1-M/CO-ABK	FI/MS/MP	5.12	2F/2M-CO	AW
3Btg1	Btg1	40–50	10 YR 3/4	1-D 10YR 2/1 (MNM) 1-P 2.5 YR 7/8	SIC	2-F-SBK	VFI/MS/VP	5.02	2F/1M	AS
	Btg2	50–70	7.5 YR 4/3	1-P 2.5YR 4/8 (FE)	SIC	1-F/M-SBK	VFI/VS/VP	5.02	2F/1M	AS
3Btg2	Btg3	70–80	7.5 YR 3/3	1-P 10YR 2/1 (MNM) 1-P 7.5YR 5/8 (FE) 3-P 10YR 2/1 (MNM)	SIC	1-M/CO-SBK/MA	VFI/VS/VP	5.02	1VF/1F	AS
	Btg4	80–100	7.5 YR 3/4	2-P 10YR 2/1 (MNM) 2-P 2.5YR 4/8 (FE)	SIC	2-F/CO-ABK/MA	VFI/VS/VP	5.02	1VF	SG
	Btg5	100+	10 YR 5/6	3-P 10YR2/1 (MNM) 2-P 7.5YR5/8 (FE)	SIC	2-M-SBK	FI/VS/VP	5.02	1VF	
PIT-4										
4A	A	0–20	10 YR 2/2		SIL	2-M-GR	S/SH/MP	5.9	2M/2F/3VF	CS
4E/A	E/A	20–40	10 YR 3/6		L	1-VF/F-SBK	S/SS/MP	6.3	2M/2F/3VF	CS
4E	E	40–60	10 YR 4/4		SICL	2-M/F-ABK	HA/SS/VP	6.2	1M/2F	GS
4Bt	Bt1	60–80	10 YR 4/4		SICL	3-M/F-ABK	VH/SS/VP	6.2	1VC/2CO/1VF	AW
	Btg2	80–110	10 YR 3/4	1-D 10YR 2/1(2% MNF) 1-D 10YR 6/8 (2% FEF)	SICL	3-M/F-ABK	HA/MS/MP	6	1F/1M	SG
	Btg3	110–150	10 YR 4/4	1-D 10YR 2/1 (2% MNF) 1-D 10YR 6/8 (1% FEF)	SICL	2-M/F-SBK	S/SS/MP	5.8	1VF	
PIT-5										
5A	A	0–6	10 YR 2/1		L	1-M-GR	S/SO/PO	7.4	2F/2M/1VC	CS
5A/B	A/B	6–13	(60%)10 YR 2/1 (40%)10 YR 4/4		L	1-M-GR parting to 1-F-SBK	S/SS/SP	7.5	2F/1M/1VC	CW
5Bt	Bt1	13–42	10 YR 3/2		SCL	2-F/M-ABK	SH/SS/PS	7.6	2M	CS
	Bt2	42–60	(85%)7.5 YR 4/6 (15%)10 YR 3/1		SCL	1-F/CO-SBK	SH/SO/SP	7.7	2VF/1M/1VC	CS
5C	C	60–120	7.5 YR 6/8		LS	1-M/F/-SBK parting to 1-VF-SBK	S/SO/PO	8.3	1F/1M/3VF	
PIT-6										
6A	A	0–17	10 YR 2/2		SL	2-F/M-SBK parting to 2-F/VF-SBK	SO/SS/SP	6	3VF/3F/2M/1C	AW
6E	E	17–40	(80%)10 YR3/4 (20%)10 YR2/2		SL	2-F/M-ABK	SH/MS/MP	6.2	2VF/2F/1M	CS
6Bt	Bt1	40–56	7.5 4/4	1-D-7.5YR 5/1(2% FMM)	SCL	2-F-SBK	SH/MS/MP	5.7	1VF/1F/1M/1C	CS
	Bt2	56–80	7.5 YR 4/4		SCL	3-F-SBK	H/VS/VP	5.8	2M/2VC	AS
	2Bt3	80–98	7.5 YR 3/4		SL	1-F-SBK	VH/SS/VP	5.8	1F/2M	AS
	2Bt4	98–107	(60%)7.5 YR4/6 (30%)7.5 YR2.5/2 (10%)10 YR3/4		SL	3-M/CO-ABK	VH/SO/PO	6.8	1F/1M	CS
	2Bt5	107–121	(70%)7.5 YR3/4 (30%)7.5 YR2.5/2		SL	2-M/CO-SBK	SH/SS/MP	6.7	1M	CW

References

Allison, V.J., Yermakov, Z., Miller, R.M., Jastrow, J.D., Matamala, R., 2007. Using landscape and depth gradients to decouple the impact of correlated environmental variables on soil microbial community composition. Soil Biology and Biochemistry 39 (2), 505–516.

Almond, P.C., Tonkin, P.J., 1999. Pedogenesis by upbuilding in an extreme leaching and weathering environment, and slow loess accretion, south Westland, New Zealand. Geoderma 92 (1–2), 1–36.

Amelung, W., 2001. Methods using amino sugars as markers for microbial residues in soil. In: Lal, R., Kimble, J.M., Follett, R.F., Stewart, B.A. (Eds.), Assessment Methods for Soil Carbon. CRC/Lewis Publishers, Boca Raton, FL, pp. 233–270.

Amelung, W., Lobe, I., Du Preez, C.C., 2002. Fate of microbial residues in sandy soils of the South African Highveld as influenced by prolonged arable cropping. European Journal of Soil Science 53 (1), 29–35.

Angers, D.A., Mehuys, G.R., 1990. Barley and alfalfa cropping effects on carbohydrate contents of a clay soil and its size fractions. Soil Biology and Biochemistry 22 (3), 285–288.

Balser, T.C., 2005. Humification. In: Hillel, D. (Ed.), Encyclopedia of Soils in the Environment. Elsevier, Oxford, UK., pp. 195–207.

Batjes, N.H., 1996. Total carbon and nitrogen in the soils of the world. European Journal of Soil Science 47 (2), 151–163.

Clayton, L., Attig, J., 1997. Pleistocene Geology of Dane County, Wisconsin, University of Wisconsin-Extension, Geological and Natural History Survey, Madison.

Ekelund, F., Ronn, R., Christensen, S., 2001. Distribution with depth of protozoa, bacteria and fungi in soil profiles from three Danish forest sites. Soil Biology and Biochemistry 33 (4–5), 475–481.

Engelking, B., Flessa, H., Joergensen, R.G., 2007. Shifts in amino sugar and ergosterol contents after addition of sucrose and cellulose to soil. Soil Biology and Biochemistry 39 (8), 2111–2118.

Eswaran, H., Vandenberg, E., Reich, P., 1993. Organic-carbon in soils of the world. Soil Science Society of America Journal 57 (1), 192–194.

Falloon, P.D., Smith, P., 2000. Modelling refractory soil organic matter. Biology and Fertility of Soils 30 (5–6), 388–398.

Fierer, N., Schimel, J.P., Holden, P.A., 2003. Variations in microbial community composition through two soil depth profiles. Soil Biology & Biochemistry 35 (1), 167–176.

Fontaine, S., et al., 2007. Stability of organic carbon in deep soil layers controlled by fresh carbon supply. Nature 450 (7167), 277–280.

Fritze, H., Pietikainen, J., Pennanen, T., 2000. Distribution of microbial biomass and phospholipid fatty acids in Podzol profiles under coniferous forest. European Journal of Soil Science 51 (4), 565–573.

Glaser, B., Turrion, M.B., Alef, K., 2004. Amino sugars and muramic acid—biomarkers for soil microbial community structure analysis. Soil Biology & Biochemistry 36 (3), 399–407.

Guerrant, G.O., Moss, C.W., 1984. Determination of monosaccharides as aldononitrile, O-methyloxime, alditol, and cyclitol acetate derivatives by gas-chromatography. Analytical Chemistry 56 (4), 633–638.

Guggenberger, G., Frey, S.D., Six, J., Paustian, K., Elliott, E.T., 1999. Bacterial and fungal cell-wall residues in conventional and no-tillage agroecosystems. Soil Science Society of America Journal 63 (5), 1188–1198.

Jobbagy, E.G., Jackson, R.B., 2000. The vertical distribution of soil organic carbon and its relation to climate and vegetation. Ecological Applications 10 (2), 423–436.

Jolivet, C., Angers, D.A., Chantigny, M.H., Andreux, F., Arrouays, D., 2006. Carbohydrate dynamics in particle-size fractions of sandy spodosols following forest conversion to maize cropping. Soil Biology and Biochemistry 38 (9), 2834–2842.

Kaiser, K., Guggenberger, G., Haumaier, L., 2004. Changes in dissolved lignin-derived phenols, neutral sugars, uronic acids, and amino sugars with depth in forested Haplic Arenosols and Rendzic Leptosols. Biogeochemistry 70 (1), 135–151.

Kandeler, E., Stemmer, M., Gerzabek, M.H., 2005. Role of microorganisms in carbon cycling in soils. In: Buscot, F.V., Varma, A. (Eds.), Microorganisms in Soils: Roles in Genesis and Functions. Springer-Verlag, Berlin, Heidelberg, pp. 139–157.

Kiem, R., Kögel-Knabner, I., 2003. Contribution of lignin and polysaccharides to the refractory carbon pool in C-depleted arable soils. Soil Biology & Biochemistry 35 (1), 101–118.

Kindler, R., Miltner, A., Richnow, H.-H., Kastner, M., 2006. Fate of gram-negative bacterial biomass in soil—mineralization and contribution to SOM. Soil Biology and Biochemistry 38 (9), 2860–2870.

Kline, V.A., 1992. Long Range Management Plan for Arboretum Ecological Communities. University of Wisconsin-Madison Arboretum, Madison, Wisconsin.

Kramer, C., Gleixner, G., 2008. Soil organic matter in soil depth profiles: distinct carbon preferences of microbial groups during carbon transformation. Soil Biology & Biochemistry 40 (2), 425–433.

Liang, C., Fujinuma, R., Wei, L., Balser, T.C., 2007a. Tree species-specific effects on soil microbial residues in an upper Michigan old-growth forest system. Forestry 80 (1), 65–72.

Liang, C., Zhang, X., Balser, T.C., 2007b. Net microbial amino sugar accumulation process in soil as influenced by different plant material inputs. Biology and Fertility of Soils 44 (1), 1–7.

Liang, C., Zhang, X., Rubert, K.F., Balser, T.C., 2007c. Effect of plant materials on microbial transformation of amino sugars in three soil microcosms. Biology and Fertility of Soils 43 (6), 631–639.

Lorenz, K. and Lal, R., 2005. The depth distribution of soil organic carbon in relation to land use and management and the potential of carbon sequestration in subsoil horizons, Advances in Agronomy, Vol 88. Advances in Agronomy, pp. 35–66.

Lowe, L.E., 1983. Carbohydrates in soil. In: Schnitzer, M., Khan, S.U. (Eds.), Soil Organic Matter. Elsevier, New York, pp. 65–93.

Morris, D.R., Glaz, B., Daroub, S.H., 2004. Organic matter oxidation potential determination in a periodically flooded histosol under sugarcane. Soil Science Society of America Journal 68 (3), 994–1001.

Murayama, S., 1984. Changes in the monosaccharide composition during the decomposition of straws under field conditions. Soil Science and Plant Nutrition 30 (3), 367–381.

Nannipieri, P., Pedrazzini, F., Arcara, P.G., Piovanelli, C., 1979. Changes in amino-acids, enzyme-activities, and biomasses during soil microbial-growth. Soil Science 127 (1), 26–34.

Niggemann, J., Schubert, C.J., 2006. Sources and fate of amino sugars in coastal Peruvian sediments. Geochimica Et Cosmochimica Acta 70 (9), 2229–2237.

Oades, J.M., 1984. Soil organic-matter and structural stability— mechanisms and implications for management. Plant and Soil 76 (1–3), 319–337.

Parsons, J.W., 1981. Chemistry and distribution of amino sugars in soils and soil organisms. In: Paul, E.A., Ladd, J.N. (Eds.), Soil Biochemistry. Marcel Dekker, New York, pp. 197–227.

Puget, P., Angers, D.A., Chenu, C., 1998. Nature of carbohydrates associated with water-stable aggregates of two cultivated soils. Soil Biology and Biochemistry 31 (1), 55–63.

Rodionov, A., Amelung, W., Urusevskaja, I., Zech, W., 2001. Origin of the enriched labile fraction (ELF) in Russian chernozems with different site history. Geoderma 102 (3–4), 299–315.

Simpson, A., Simpson, M., Smith, E., Kelleher, B., 2007. Microbially derived inputs to soil organic matter: are current estimates too low? Environ. Science and Technology 41 (23), 8070–8076.

Solomon, D., Lehmann, J., Zech, W., 2001. Land use effects on amino sugar signature of chromic Luvisol in the semi-arid part of northern Tanzania. Biology and Fertility of Soils 33 (1), 33–40.

Sowden, F.J., Ivarson, K.C., 1974. Effects of temperature on changes in nitrogenous constituents of mixed forest litters during decomposition after inoculation with various microbial cultures. Canadian Journal of Soil Science 54 (4), 387–394.

Sundh, I., Nilsson, M., Borga, P., 1997. Variation in microbial community structure in two boreal peatlands as determined by analysis of phospholipid fatty acid profiles. Applied and Environmental Microbiology 63 (4), 1476–1482.

Taylor, J.P., Wilson, B., Mills, M.S., Burns, R.G., 2002. Comparison of microbial numbers and enzymatic activities in surface soils and subsoils using various techniques. Soil Biology and Biochemistry 34 (3), 387–401.

von Lützow, M., et al., 2006. Stabilization of organic matter in temperate soils: mechanisms and their relevance under different soil conditions—a review. European Journal of Soil Science 57 (4), 426–445.

Zhang, X., Amelung, W., 1996. Gas chromatographic determination of muramic acid, glucosamine, mannosamine, and galactosamine in soils. Soil Biology and Biochemistry 28 (9), 1201–1206.

Zhang, X., Amelung, W., Yuan, Y., Zech, W., 1998. Amino sugar signature of particle-size fractions in soils of the native prairie as affected by climate. Soil Science 163 (3), 220–229.