

Greater Soil Carbon Sequestration under Nitrogen-fixing Trees Compared with *Eucalyptus* Species

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ABSTRACT

Forests with nitrogen-fixing trees (N-fixers) typically accumulate more carbon (C) in soils than similar forests without N-fixing trees. This difference may develop from fundamentally different processes, with either greater accumulation of recently fixed C or reduced decomposition of older soil C. We compared the soil C pools under N-fixers with *Eucalyptus* (non-N-fixers) at four tropical sites: two sites on Andisol soils in Hawaii and two sites on Vertisol and Entisol soils in Puerto Rico. Using stable carbon isotope techniques, we tracked the loss of the old soil organic C from the previous C₄ land use (SOC₄) and the gain of new soil organic C from the C₃, N-fixer, and non-N-fixer plantations (SOC₃). Soils beneath N-fixing trees sequestered $0.11 \pm 0.07 \text{ kg m}^{-2} \text{ y}^{-1}$ (mean $\pm$ one standard

error) of total soil organic carbon (SOC_T) compared with no change under *Eucalyptus* ($0.00 \pm 0.07 \text{ kg m}^{-2} \text{ y}^{-1}$; $P = 0.02$). About 55% of the greater SOC_T sequestration under the N-fixers resulted from greater retention of old SOC₄, and 45% resulted from greater accretion of new SOC₃. Soil N accretion under the N-fixers explained 62% of the variability of the greater retention of old SOC₄ under the N-fixers. The greater retention of older soil C under N-fixing trees is a novel finding and may be important for strategies that use reforestation or afforestation to offset C emissions.

Key words: carbon accretion; carbon retention; carbon sequestration; *Eucalyptus*; Hawaii; isotopes; nitrogen-fixing trees; Puerto Rico; soil organic carbon.

INTRODUCTION

Soils store $1.5 \times 10^{18} \text{ g}$ of organic carbon (C) globally—about twice as much C as is found in the atmosphere and three times the quantity contained in terrestrial vegetation (Schlesinger 1997). Soil organic C is intricately linked to the cycling of soil nutrients that influence ecosystem productivity (Van Cleve and Powers 1995). Thus, changes in this huge pool of C could mitigate or exacerbate the rise

in atmospheric carbon dioxide (CO₂) (Schlesinger 1990, 1999; Nadelhoffer and others 1999).

Deforestation may contribute to the loss of soil C by changing the balance between biomass production and decomposition. Tropical deforestation may be a net source of $0.2 \times 10^{15} \text{ g C y}^{-1}$, with up to 25% coming from soils (Houghton 1994, 2000). Intensive cultivation can also decrease soil C, contributing to terrestrial net fluxes of C to the atmosphere and decreased net primary productivity (Burke and others 1989; Johnson 1992). Some land-use practices (such as low-tillage, legume-based, or manure application agriculture) can increase soil C storage relative to conventional agri-

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cultural systems (Paustian and others 1997; Drinkwater and others 1998), but reforestation and afforestation may have the greatest potential for changes in land use to offset increasing CO₂ emissions (Vitousek 1991; Brown and others 1992; Mof-fat 1997; Bruce and others 1999).

Reforestation and afforestation of agricultural land increase C storage in plant biomass relative to pasture or annual-crop vegetation, but it is still unclear what differences they exert on the storage of soil C (Bashkin and Binkley 1998; Binkley and Resh 1999; Richter and others 1999; Post and Kwon 2000). Moreover, the effect may differ among tree species (Lugo and others 1990). Tree species differ in biomass production and tissue nutrient concentrations and in their effects on soil properties such as pH, nutrient cycling, and soil biota (Binkley 1996; Binkley and Giardina 1998). All comparisons of N-fixers and non-N-fixers have found 20%–100% more soil C under N-fixers (Johnson 1992; Cole and others 1995; Rhoades and others 1998); this would equate to 0.05–0.12 kg m⁻² y⁻¹ greater soil C accumulation under N-fixer forests than under comparable non-N-fixer forests (Tarrant and Miller 1963; Binkley and others 1982; Binkley 1983; Binkley and Sollins 1990; Cole and others 1995; Rhoades and others 1998; Kaye and others 2000). Despite the very consistent and large effect of N-fixing trees on the storage of soil C, the fundamental processes that yield the higher C storage remain unexplained; we do not even know if the higher C storage derives from greater C inputs or reduced C outputs.

We used a change in land use from C₄ agriculture to C₃ forest plantations to determine how N-fixers increase total soil C storage relative to non-N-fixers. Stable carbon isotope techniques allowed us to separate the effects of N-fixers on the storage of older soil C derived from C₄ vegetation (SOC₄) from the accretion of newer soil C derived from the C₃ trees (SOC₃). We compared the soil C dynamics under N-fixers with those of non-N-fixing species of *Eucalyptus* across four sites in Hawaii and Puerto Rico with differing precipitation regimes and soils. We also conducted long-term laboratory incubations of the surface soils from the N-fixer and non-N-fixer stands from three of our study sites to determine whether recently added SOC₃ was more or less labile in soils beneath N-fixing trees.

METHODS

Study Sites

The study sites consisted of 7- to 16-year-old plantations of N-fixers and *Eucalyptus* species (non-N-

fixers) with three to six replicated plots of each species at each site (Table 1). At the Hawaii sites, the plantation species were *Albizia falcataria* (L.) Fosberg (also known as *Paraserianthes falcataria* (L.) Nielsen and *Falcataria moluccana* (Miquel) Barneby & Grimes) and *Eucalyptus saligna* (Sm.). At the Puerto Rico sites, the plantation species were *Casuarina equisetifolia* (L.), *Leucaena leucocephala* (Lam.) de Wit variety K636, and *Eucalyptus robusta* J.E. Smith. *Albizia* and *Leucaena* belong to the Fabaceae, subfamily Mimosoideae, and *Casuarina* belongs to the Casuarinaceae.

Albizia is nodulated by the *Rhizobia* and *Bradyrhizobia* bacteria and also hosts vesicular-arbuscular mycorrhizae (VAM) (Turk and Keyser 1992). *Albizia* is native to the eastern islands of the Indonesian archipelago and is widely grown in tropical plantations for fuel, pulp, and light construction (Binkley and Giardina 1997). *Albizia* trees grow very rapidly to 25–30 m tall and up to 1 m in diameter at rates of 6–18 Mg ha⁻¹ y⁻¹ of stem increment with N-fixing rates of 65–140 kg N ha⁻¹ y⁻¹ (Little and Skolmen 1989; Binkley and Giardina 1997).

Leucaena is nodulated by *Rhizobia* bacteria and hosts VAM (Binkley and Giardina 1997). *Leucaena* is native to Central America, and *Leucaena leucocephala* is planted extensively in tropical plantations for fuel, roundwood, pulp and paper, and soil improvement (Parrotta 1992; Zabala 1997). *Leucaena* trees grow rapidly up to 20 m in height and 0.5 m in diameter with aboveground biomass production of 5–55 Mg ha⁻¹ y⁻¹ (Parrotta 1992) and N-fixation rates of 70–150 kg N ha⁻¹ y⁻¹ (Little and Skolmen 1989; Parrotta and others 1996; Binkley and Giardina 1997).

Casuarina is nodulated by *Frankia* actinomycetes and also hosts VAM and ectomycorrhizae (Binkley and Giardina 1997). *Casuarina* is native to Australia, Malaysia, and Polynesia (Subbarao and Rodríguez-Barrueco 1995). It is grown throughout the tropics for land reclamation, sand dune stabilization, and wind breaks, as well as timber (Parrotta 1993; Binkley and Giardina 1997). *Casuarina* grows to 30–40 m tall and 0.5 m in diameter with wood increments on good sites of 10–15 Mg ha⁻¹ y⁻¹ and N-fixation rates of 60–95 kg N ha⁻¹ y⁻¹ in pure stands (Parrotta 1993; Parrotta and others 1994, 1996; Subbarao and Rodríguez-Barrueco 1995; Binkley and Giardina 1997).

The two species of *Eucalyptus* are native to south-eastern Australia. Both species may grow to 40–60 or more m in height and diameters of 1.2 m or more. *E. robusta* grows well in Puerto Rico and was also planted extensively in Hawaii until the 1960s, when planting shifted to *E. saligna* and *E. grandis*.

Table 1. Description of the Four Study Sites

Characteristic	Kamac, HI 20°N, 155°W	Chinchuck, HI 20°N, 155°W	Lajas, PR 18°N, 76°W	Toa Baja, PR 18°N, 66°W
Plantation establishment	September 1982	September 1982	February 1980	September 1989
Age at sampling	15 yr	16 yr	16 yr	7 yr
Land use at time of planting	Fallow sugarcane	Fallow sugarcane	Fallow sugarcane	Pasture
Species				
N-fixer(s)	<i>Albizia</i>	<i>Albizia</i>	<i>Castuarina, Leucaena</i>	<i>Castuarina, Leucaena</i>
non-N-fixer	<i>Eucalyptus saligna</i>	<i>Eucalyptus saligna</i>	<i>Eucalyptus robusta</i>	<i>Eucalyptus robusta</i>
Climate				
Precipitation	4000 mm y ⁻¹	4000 mm y ⁻¹	1100 mm y ⁻¹	1600 mm y ⁻¹
Temperature	21°C	21°C	25°C	27°C
Soils				
Classification	Typic Hydrudands	Typic Hydrudands	Typic Haplusterts	Typic Troposammments
Surface soil pH (CaCl ₂)	4.6 ^a	5.0 ^a	5.6	7.0
Fertilization regimes	11 and 5 g/m ² N and P at planting	4, 1.8, and 3.3 g/m ² N, P, and K, respectively, at planting, 4 and 8 mo (and at 12, 18, 24, and 36 mo for <i>Eucalyptus</i> seedlings)	11.2 g/m ² N before planting and 6-mo intervals for initial 2 yr	1.0 g/m ² N at 6-mo intervals from March 1990 to 1992

^aGarcía-Montiel and Binkley (1998)

See text for primary references for each site

Eucalyptus wood is used for housing, pulp, fuel, charcoal, and pallets (Little and Skolmen 1989).

The Hawaii study sites are located on the north-east coast of Hawaii (Table 1). These sites are described in greater detail as "Upper Kamae" and "Chinchuck" in Binkley (1997) and Garcia-Montiel and Binkley (1998). In the 1920s, both sites were converted from wildland forest (C_3 species) to sugarcane (*Saccharum officinarum*) (C_4 species). Sugarcane production at the Kamae site was abandoned in 1960 due to low production. Sugarcane production at the Chinchuck site continued until 1980. In 1981, old vegetation was cleared on both sites, soils were plowed, and new growth was treated with herbicide. In 1982, pure species trials of *Albizia* and *Eucalyptus* were planted in a randomized complete block design with four blocks (eight plots total). The trees were planted at a spacing of 1.5×1.5 m in 12×18 m plots for both *Albizia* and *Eucalyptus* at the Kamae site and at a spacing of 2×2 m in 15 by $30 \times$ plots for *Albizia* and 30×30 m plots for *Eucalyptus* at the Chinchuck site, with no buffers between plots for either site. These sites are within 1 km on similar soils, but they differed substantially in tree biomass and total soil N (Garcia-Montiel and Binkley 1998). Based on these differences, we treated the sites as independent units for statistical purposes. At the time of soil sampling, the Kamae and Chinchuck tree plantations were 15 and 16 years old, respectively.

One Puerto Rico site is located in the Lajas Valley in southwest Puerto Rico (Table 1) (Lugo and others 1990; Wang and others 1991). The site was plowed and treated with herbicide before planting. In February 1980, seedlings of six taxa were planted in pure species plots; we used three of these for this study. Weeding and fertilization were used for the first 6 months, and irrigation was applied when necessary to ensure initial survival. Pure plots of *Casuarina*, *Leucaena*, and *Eucalyptus* were planted in a complete block design with six blocks. The trees were planted at an original spacing of 1×1 m which was thinned to 2×2 m by age 1.5 years. Plot size was 10×10 m with a 3-m buffer between plots and a 5-m buffer between blocks. At the time of planting, the site was fallow following long-term sugarcane management. When we sampled the plantation in December 1996, the area outside the plantation was dominated by a C_4 grass (*Panicum maximum*) with some herbaceous C_3 species. Woody species have not dominated this site for the past 100 years prior to these tree plantations (A. E. Lugo, personal communication). Trees were 16 years old at the time of sampling.

The second Puerto Rico site is located on the

northern coast at the University of Puerto Rico's Toa Baja experimental farm (Parrotta and others 1996; Parrotta 1999). The site has had many disturbances that included leveling of the original forested dunes, sand extraction, and cultivation. Pasture grasses, mostly *Panicum maximum*, were the dominant vegetation at plantation establishment. Site preparation consisted of disking the soil to a depth of 20 cm. The control plots, which remained in pasture grass, were also disked. Pure and mixed plots of *Casuarina*, *Leucaena*, and *Eucalyptus* were established in a randomized complete block design with three blocks. For this current study, only the pure species plots were used. Trees were planted at a spacing of 1×1 m in 16×16 m plots. There was a 5-m buffer between plots and blocks. Recent samples (December 1996) and archived soils from just prior to tree plantation establishment were analyzed from this site. Trees were 7 years old at the time of sampling.

Sample Collection and Processing

We collected soil samples in January 1997 from Kamae, in January 1998 from Chinchuck, and in December 1996 from the Puerto Rico sites. We used a 6-cm-diameter soil core lined with a butyrate plastic liner for soil sampling at the Hawaii and Lajas sites. For soil sampling at the Toa Baja site, we used a 5.1-cm-diameter soil corer. In each of the eight plots at the Kamae site, we collected one soil sample to a depth of 40 cm and two samples to a depth of 20 cm near the plot center in a triangular pattern 1 m apart. Three 40-cm cores were collected from neighboring sugarcane fields to be analyzed as a proxy for pre-forest plantation conditions. In each of the eight plots at the Chinchuck site, we collected two soil samples to a depth of 50 cm. Data for neighboring sugarcane soil carbon was provided by M. Bashkin (unpublished). In each of the 18 plots at the Lajas site, we collected one 40-cm soil sample near each plot center. Control plots were located at the end of each of the six blocks, and a 40-cm soil sample was collected at least 5 m from the plantation edge to use as a proxy for pre-plantation conditions for this site. The same sampling design used at Kamae was used for the Toa Baja plantation in Puerto Rico. However, at this site, control plots of pre-plantation vegetation were included in the experimental design and were sampled in the same way as the forest soils; one 40-cm and two 20-cm samples were taken from each of the three control plots.

Upon collection, we immediately processed the Kamae, Chinchuck, and Toa Baja soils at a field lab. Lajas soils were stored in the butyrate soil core

liners for less than 5 days for transport back to Colorado State University (CSU) before processing. In the lab, we divided all soils into 10-cm segments, with the exception of the Chinchuck soils, which we divided into 0–20- and 20–50-cm segments. Where we had more than one sample per plot, we composited all soil samples into one sample per plot. Roots larger than 2 mm were removed by hand before any analyses were done. Bulk density (Table 2) was determined for each of the soil segments by oven-drying a known fresh mass to a constant dry mass at 105°C; rock mass was negligible.

We treated the soils from Kamae and the two Puerto Rico sites with a 0.2N HCl wash to remove CaCO_3 that would affect $\delta^{13}\text{C}$ values (Ehleringer 1991). There was no evidence of CaCO_3 in the Chinchuck soils when 0.2N HCl solution was applied to 10 subsamples. We oven-dried the soils at 105°C and ground them to flour texture for analysis of total C, N, and C isotopes. Soil organic C and N concentrations (Table 2) were determined by dry combustion (LECO-1000; LECO Corporation, St. Joseph, MI, USA). We corrected the C and N concentrations to account for the added salt mass from the acidification process. The $\delta^{13}\text{C}$ values of the soils were measured using a VG isochrom-NA stable isotopic ratio mass spectrometer (VG, Middlewich, UK). The mass spectrometer is operated in automatic continuous flow mode using helium as the carrier gas after combustion. The $\delta^{13}\text{C}$ values, measured in ‰ (parts per thousand), were calculated as:

$$\delta^{13}\text{C} = [(R_{\text{sample}}/R_{\text{PDB}}) - 1] \times 10^3 \quad (1)$$

where $R = {}^{13}\text{C}/{}^{12}\text{C}$ and PDB = Pee Dee Belemnite standard for carbon.

We used archived soils taken at plantation establishment (1989) from the Toa Baja site as pre-plantation estimates of soil $\delta^{13}\text{C}$. The archived soil samples were air-dried at the time of sampling, stored in plastic containers, and then analyzed concurrently with the fresh soil samples for the procedures described above.

Soil Incubations

We conducted 4- and 8-month soil incubations to determine whether SOC_3 derived from N-fixing trees decomposed more rapidly than SOC_3 derived from *Eucalyptus*. Surface soils (0–10-cm) from each tree plot from Kamae, Lajas, and Toa Baja were coarsely sieved (6-mm). We incubated fresh soil samples equivalent to 15 g of oven-dried soil. Each soil sample was brought to -0.01 MPa water potential using a pressure plate; the samples were

weighed and checked twice weekly and brought back to the initial -0.01 MPa mass. Soils were incubated at 35°C inside loosely covered 1-L glass jars, which also contained a vessel of water to maintain a high humidity environment. The jars were opened to ambient atmosphere twice weekly to ensure adequate availability of oxygen for decomposition.

To determine relative loss of SOC_3 , pre- and postincubation SOC concentration and $\delta^{13}\text{C}$ values were measured for each sample, and Eqs. (2) and (3) were used to determine the % C_3 and mass of the pre- and postincubation SOC_3 . The SOC_3 change over time was determined by subtracting the postincubation values from the preincubation values and dividing by the preincubation values to get a relative C_3 loss comparison (mg C_3 loss/initial C_3). We analyzed for the effect of species (that is, N-fixer vs non-N-fixer) on the relative loss of SOC_3 using a split-plot statistical design.

Corrections for Bulk Density

Bulk density did not differ between species for the Hawaii sites (Table 2A and B). However, bulk density differed for soils under the pasture and forest at Lajas and over time at Toa Baja (Table 2C and D). This means that a 0–10-cm sample taken at one place or time represented a different mass of soil at another place or time. To correct for the bulk density differences, Lajas forest soil mass was adjusted to an equivalent pasture soil mass and Toa Baja 1996 soil masses were adjusted to 1989 equivalent masses to reflect equivalent soil mass comparisons. Concentrations of C and N and $\delta^{13}\text{C}$ values were all corrected for the depth adjustments. For simplicity, depths will be addressed as 0–10, 10–20, 20–30, and 30–40 cm. The bulk density adjustments for Lajas increased the average SOC_T (kg/m^2) under the trees by 4%.

Calculations

The percent of total soil organic carbon (SOC_T) coming from pasture grasses or sugarcane (C_4) was calculated as:

$$\% \text{C}_4 = [(\delta^{13}\text{C}_{\text{sample}} - \delta^{13}\text{C}_3) / (\delta^{13}\text{C}_4 - \delta^{13}\text{C}_3)] \times 100 \quad (\text{Cerri and others 1985}) \quad (2)$$

where $\delta^{13}\text{C}_3$ is the $\delta^{13}\text{C}$ of a composite sample of pure forest litterfall and roots and $\delta^{13}\text{C}_4$ is the $\delta^{13}\text{C}$ of a composite of pure C_4 grass or sugarcane litter, leaves, and roots. The C_3 and C_4 endpoints were different for each site and for each tree species within the Toa Baja and Lajas sites (Table 3). The

Table 2. Soil Bulk Density, pH, Effective Cation Exchange Capacity (ECEC), Base Saturation (BS), N, C, and ^{13}C Concentrations for **A** the Kamae Site, **B** the Chinchuck Site, **C** the Lajas Site, and **D** the Toa Baja Site

A Kamae							
Depth (cm)	Bulk Density (kg L ⁻¹)	pH ^a (in Water)	ECEC ^a (mmol _c kg ⁻¹)	BS ^a (%)	N (g kg ⁻¹)	C (g kg ⁻¹)	δ ¹³ C (‰)
<i>Eucalyptus</i> (<i>n</i> = 3)							
0–10	0.51 ± 0.08	4.8 ± 0.1	40.4 ± 3.3	22.3 ± 4.3	8.2 ± 0.3	133.4 ± 10.5	-22.8 ± 1.2
10–20	0.37 ± 0.13				6.4 ± 1.4	102.5 ± 15.4	-22.5 ± 1.1
20–30	0.24 ± 0.04				4.5 ± 0.2	81.7 ± 5.4	-24.1 ± 0.3
30–40	0.27 ± 0.03				4.6 ± 0.2	82.4 ± 3.1	-23.9 ± 0.7
<i>Albizia</i> (<i>n</i> = 4)							
0–10	0.49 ± 0.06	4.7 ± 0.04	39.4 ± 2.2	20.0 ± 4.7	9.4 ± 0.4	142.4 ± 5.0	-22.9 ± 0.5
10–20	0.40 ± 0.14				7.5 ± 0.7	113.8 ± 7.0	-23.1 ± 0.8
20–30	0.28 ± 0.06				5.3 ± 0.6	85.5 ± 9.7	-23.5 ± 0.6
30–40	0.26 ± 0.07				4.6 ± 0.4	77.5 ± 12.5	-23.8 ± 0.6
Cane (<i>n</i> = 3)							
0–10	0.44 ± 0.10	no data	no data	no data	6.9 ± 0.3	103.7 ± 7.0	-20.4 ± 0.7
10–20	0.43 ± 0.12				5.3 ± 0.9	85.9 ± 11.5	-22.2 ± 1.3
20–30	0.32 ± 0.04				4.9 ± 0.8	82.4 ± 15.7	-23.7 ± 0.7
30–40	0.34 ± 0.07				3.2 ± 1.3	57.0 ± 21.2	-24.0 ± 0.3
B Chinchuck							
Depth (cm)	Bulk Density (kg L ⁻¹)	pH ^a (in Water)	ECEC ^a (mmol _c kg ⁻¹)	BS ^a (%)	N (g kg ⁻¹)	C (g kg ⁻¹)	δ ¹³ C (‰)
<i>Eucalyptus</i> (<i>n</i> = 4)							
0–20	0.34 ± 0.07	5.2 ± 0.2	19.5 ± 3.9	52.3 ± 18.6	6.2 ± 0.8	112.0 ± 12.8	-24.3 ± 0.4
20–50	0.28 ± 0.10				3.7 ± 1.1	69.6 ± 19.7	-24.7 ± 0.3
<i>Albizia</i> (<i>n</i> = 4)							
0–20	0.39 ± 0.07	5.0 ± 0.1	24.3 ± 3.3	36.3 ± 3.1	7.4 ± 1.1	114.6 ± 16.6	-23.9 ± 0.7
20–50	0.27 ± 0.04				4.4 ± 0.6	77.3 ± 7.5	-24.9 ± 0.4
C Lajas							
Depth (cm)	Bulk Density (kg L ⁻¹)	pH ^a (in Water)	ECEC ^a (mmol _c kg ⁻¹)	BS ^a (%)	N (g kg ⁻¹)	C (g kg ⁻¹)	δ ¹³ C (‰)
<i>Eucalyptus</i> (<i>n</i> = 6)							
0–10	1.09 ± 0.11	no data	460 ± 10	14.7	1.6 ± 0.2	21.1 ± 2.7	-18.7 ± 1.1
10–20	1.24 ± 0.10				1.1 ± 0.2	14.8 ± 1.5	-16.3 ± 0.7
20–30	1.26 ± 0.09				0.9 ± 0.2	10.2 ± 2.8	-17.5 ± 0.8
30–40	1.36 ± 0.16				0.6 ± 0.3	6.8 ± 2.9	-18.0 ± 1.1
<i>Casuarina</i> (<i>n</i> = 6)							
0–10	0.96 ± 0.12	no data	490 ± 30	14.1	2.1 ± 0.5	29.9 ± 7.0	-20.0 ± 1.7
10–20	1.27 ± 0.08				1.1 ± 0.2	15.7 ± 3.4	-17.1 ± 0.7
20–30	1.29 ± 0.10				0.9 ± 0.2	12.6 ± 4.1	-17.3 ± 1.1
30–40	1.40 ± 0.12				0.7 ± 0.4	9.4 ± 5.9	-17.3 ± 1.4
<i>Leucaena</i> (<i>n</i> = 6)							
0–10	1.00 ± 0.06	no data	500 ± 20	14.4	2.9 ± 1.3	34.2 ± 13.7	-20.7 ± 1.3
10–20	1.21 ± 0.11				1.2 ± 0.3	15.9 ± 3.4	-16.2 ± 2.0
20–30	1.33 ± 0.06				1.0 ± 0.3	12.6 ± 4.0	-17.2 ± 1.5
30–40	1.33 ± 0.07				0.8 ± 0.3	10.0 ± 5.4	-18.3 ± 3.4

Table 2. (Continued)

C Lajas							
Depth (cm)	Bulk Density (kg L ⁻¹)	pH (in Water)	ECEC ^b (mmol _c kg ⁻¹)	BS ^{b,c} (%)	N (g kg ⁻¹)	C (g kg ⁻¹)	δ ¹³ C (‰)
Pasture (<i>n</i> = 6)							
0–10	1.27 ± 0.11	no data	no data	no data	1.4 ± 0.1	18.0 ± 1.5	–16.5 ± 0.8
10–20	1.35 ± 0.07				1.2 ± 0.1	16.3 ± 1.6	–16.1 ± 0.4
20–30	1.37 ± 0.05				1.0 ± 0.2	13.5 ± 2.4	–16.2 ± 0.8
30–40	1.39 ± 0.06				0.8 ± 0.1	10.5 ± 2.6	–17.2 ± 1.1
D Toa Baja							
Depth (cm)	Bulk Density (kg L ⁻¹)	pH ^d (in Water)	ECEC ^d (mmol _c kg ⁻¹)	BS ^e (%)	N (g kg ⁻¹)	C (g kg ⁻¹)	δ ¹³ C (‰)
<i>Eucalyptus</i> (<i>n</i> = 3)							
0–10	1.21 ± 0.19	7.77	68.6	98.7	1.1 ± 0.4	11.4 ± 2.7	–23.6 ± 0.6
10–20	1.28 ± 0.10			99.0	0.9 ± 0.4	7.7 ± 2.4	–21.9 ± 1.6
20–30	1.24 ± 0.13			98.8	0.6 ± 0.3	5.6 ± 3.3	–22.0 ± 1.0
30–40	1.41 ± 0.19			99.2	0.5 ± 0.2	4.3 ± 2.6	–21.6 ± 0.7
<i>Casuarina</i> (<i>n</i> = 3)							
0–10	1.16 ± 0.05	7.93	83.4	98.3	1.1 ± 0.3	12.6 ± 5.0	–24.8 ± 1.3
10–20	1.29 ± 0.06			99.0	0.9 ± 0.3	7.9 ± 2.9	–21.6 ± 0.9
20–30	1.49 ± 0.16			99.0	0.9 ± 0.4	7.0 ± 3.4	–20.4 ± 1.0
30–40	1.50 ± 0.13			99.1	0.5 ± 0.1	3.6 ± 0.1	–21.0 ± 0.4
<i>Leucaena</i> (<i>n</i> = 3)							
0–10	1.10 ± 0.08	7.95	93.0	98.5	1.5 ± 0.2	13.9 ± 1.8	–24.3 ± 1.4
10–20	1.23 ± 0.12			98.7	1.0 ± 0.4	8.1 ± 3.0	–21.5 ± 1.3
20–30	1.40 ± 0.26			98.8	0.8 ± 0.4	6.8 ± 2.7	–20.3 ± 1.1
30–40	1.42 ± 0.24			99.4	0.5 ± 0.3	4.1 ± 2.0	–20.6 ± 0.6
Pasture (<i>n</i> = 3)							
0–10	1.16 ± 0.09	7.70	97.3	98.9	1.4 ± 0.6	13.7 ± 5.7	–19.2 ± 1.2
10–20	1.27 ± 0.10			99.4	1.3 ± 0.3	11.7 ± 4.0	–20.3 ± 0.8
20–30	1.42 ± 0.16			98.7	0.9 ± 0.6	7.6 ± 4.7	–20.2 ± 1.7
30–40	1.45 ± 0.19			99.2	0.6 ± 0.7	5.9 ± 5.1	–23.1 ± 2.9

Standard deviations are provided where applicable.

At the Kamae site, one *Eucalyptus* plot is excluded from analysis due to the predominance of rock in the last 20 cm of the 0–40-cm soil core.

^aData from García-Montiel and Binkley (1998). Data were measured from the surface 20 cm of soil when the plantation was 12 years old.

^bData from Wang and others (1991). Data were measured from the top 3 cm of soil when the plantation was 5.5 years old.

^cBase saturation was calculated as the sum of the basic cations Ca²⁺, Mg²⁺, K⁺, and Na⁺ divided by the CEC.

^dData from Parrotta (1999). Data were measured from the surface 20 cm of soil when the plantation was 7.5 years old.

^eBase saturation was calculated as the sum of the basic cations Ca²⁺, Mg²⁺, K⁺, and Na⁺ divided by the CEC from Parrotta (1999 and unpublished). Data were measured when the plantation was 7.5 years old.

δ¹³C values for Hawaii are from Bashkin and Binkley (1998). Root δ¹³C values are from roots collected throughout all 0–40-cm soil cores and composited for each species.

Total SOC in kg C/m² was calculated based on the soil bulk density and C concentration for each sample depth. The equation for SOC₄ (kg C/m²) was:

$$\text{SOC}_4 = (\%C_4/100) \times \text{SOC}_T \quad (3)$$

Soil C derived from C₃ vegetation was calculated by subtracting SOC₄ from SOC_T.

We estimated the net changes in the SOC pools by using initial soil conditions at the time of planting, developed either from pre-plantation sampling (at Toa Baja) or from adjacent areas where land use remained in the prior condition of pasture or sugarcane (other sites). We also evaluated the divergence in SOC and soil N between the N-fixers and

Table 3. Site and Species C₃ and C₄ δ¹³C Endpoints used for Equation (2)

Site	Species	C ₃ Endpoint ^a	C ₄ Endpoint
Hawaii sites ^b	<i>Eucalyptus</i> , <i>Albizia</i> , sugarcane	-25.11	-11.52
Lajas (<i>n</i> = 6)	<i>Eucalyptus</i>	-27.01 ± 0.53	
	<i>Casuarina</i> ^c	-26.30 ± 0.17	
	<i>Leucaena</i>	-26.94 ± 0.50	
	Pasture	-26.60 ± 1.13	-13.80 ± 0.38
Toa Baja (<i>n</i> = 3)	<i>Eucalyptus</i>	-28.33 ± 0.10	
	<i>Casuarina</i> ^c	-29.41 ± 0.59	
	<i>Leucaena</i>	-28.64 ± 0.27	
	Pasture	-28.78 ^d	-14.52 ± 0.31

Endpoints are composites of litterfall and roots from each species at each site.

^a ± are standard deviation where applicable

^b Bashkin and Binkley (1998)

^c *Casuarina* differed significantly from both *Eucalyptus* and *Leucaena* (*P* < 0.1 for both sites).

^d Average of the three tree species endpoints

non-N-fixers by contrasting the soils at the time of sampling. Annual changes (kg m⁻² y⁻¹) in SOC_T, SOC₄, and SOC₃ since plantation establishment were estimated using the following equation example:

$$\text{SOC}_3 \text{ change} = (\text{current SOC}_3 - \text{initial SOC}_3) / \text{plantation age} \quad (4)$$

Separating Pre-Sugarcane/Pasture SOC₃ from Contemporary SOC₃

The SOC₃ pools for each site included both pre-C₄ and contemporary C₃ components. We assumed no change in the C₃ components that predated the establishment of C₄ vegetation for two reasons. Because disturbances to the original forest SOC₃ occurred several decades ago for all sites, any remaining SOC₃ from this period must be very recalcitrant to have remained in the soil for this long. For the Toa Baja site, we also had archived soil samples from before the establishment of the tree and pasture plantations, and there was no discernible loss of this old SOC₃ in the plots that were planted with C₄ grasses (Resh 1999; S. Resh unpublished). Because the Toa Baja soil is extremely sandy and would be predicted to have the highest SOC turnover rates compared with the other three sites with clay soils (Burke and others 1989; Schimel and others 1994), the Hawaii and Lajas clays also should have no measurable loss of older SOC₃. Thus, we attributed any changes in SOC₃ (Eq. [4]) since forest plantation establishment to additions of contemporary SOC₃. Furthermore, given that the pre-sugarcane/pasture SOC₃ turnover would be the same within any one site, SOC₃ comparisons between

current plantation species at the site would be affected equally by the older SOC₃ turnover. The resulting bias could possibly be an overprediction of the magnitude of contemporary SOC₃ pools and changes in those pools for the site by no more than 10% (Schimel and others 1994), but it would not change the relative differences between species presented in this paper.

Our estimates of the magnitude of change in the pools of organic matter derived from C₃ and C₄ sources depend in part on our expectation that the δ¹³C of organic matter from prior land use was the same as the values we used for the current C₃ (trees) and C₄ (grasses) vegetation. We are confident that for the Hawaiian sites the historic SOC₃ was similar in δ¹³C to SOC derived from *Eucalyptus*, based on comparisons of native C₃ forests and *Eucalyptus* plantations that had no C₄ influence (Bashkin and Binkley 1998). In Puerto Rico, the prior soil organic matter derived from C₃ forests may have differed somewhat in δ¹³C from the endpoint we used, but any such difference should be minor (less than 2 per mil); it would provide small biases in our estimate SOC₄ and SOC₃ pool sizes (10%–20%); and it would not affect the test of the differing effects of tree species.

Statistical Analysis

All analyses were conducted using SYSTAT version 7.0 (SYSTAT 1997). For each analysis, residual plots of the raw data were inspected visually for nonnormality and heteroscedasticity violations of analysis of variance (ANOVA) assumptions.

Species effects on SOC and N pools within site. We used an ANOVA for a general two-factor com-

pletely randomized block design with two to three levels of species and three to six levels of block (the number of species and block depends on site). To test for the tree species effects on the SOC pools and soil N within a given site, we used species as the main effect with species by block interaction as the error term. For the Puerto Rico sites with more than two tree species, we used Tukey's multiple comparison for determination of significant differences.

Species effects on changes in SOC pools across the four sites. To test for N-fixer vs non-N-fixer differences across sites, annual SOC changes were used (Eq. [4]). For the Lajas and Toa Baja sites, where two N-fixer species were sampled, the species were averaged together into one N-fixer category to test the difference between N-fixing and non-N-fixing species. For each site, the research plantations used a completely randomized block design. However, combining the data into a single analysis is no longer a completely randomized block design due to the species-within-blocks restriction. An ANOVA was used with site as the main effect, blocks as plot effects, and species-within-blocks as the split-plot effects. With this model, site is treated as a fixed effect, and species and block are random effects. The error terms were plot-within-site sum of squares for the main effect and species by plot-within-site sums of squares for the species effect and the site by species interaction effect (Binkley 1997).

RESULTS

Across the four sites, N-fixers sequestered significantly more SOC_T than non-N-fixers ($0.11 \text{ kg m}^{-2} \text{ y}^{-1}$; $P = 0.02$) (Figure 1). This SOC_T difference between the N-fixers and the non-N-fixers combines a relatively small loss of old SOC₄ ($0.05 \pm 0.05 \text{ kg C}_4 \text{ m}^{-2} \text{ y}^{-1}$) with a large gain of new SOC₃ ($0.16 \pm 0.05 \text{ kg C}_3 \text{ m}^{-2} \text{ y}^{-1}$) under the N-fixers. In comparison, the non-N-fixers lost significantly much more old SOC₄ ($0.11 \pm 0.05 \text{ kg C}_4 \text{ m}^{-2} \text{ y}^{-1}$; $P = 0.03$) and accumulated less new SOC₃ ($0.11 \pm 0.05 \text{ kg C}_3 \text{ m}^{-2} \text{ y}^{-1}$; $P = 0.07$). No interactions between site and species were significant ($P > 0.1$).

SOC_T pools in the surface 40 cm ranged from 3 to 16 kg/m²; Toa Baja had the smallest and Hawaii had the largest SOC_T pools. Although only statistically significant for *Leucaena* at the Lajas site, the SOC_T and soil N pools under the N-fixers tended to be greater relative to the non-N-fixers for each of the four sites (Figures 2 and 3). At Kamae and Chinchuck in Hawaii, *Albizia* had 1.16 ($P = 0.97$) and 2.1 kg/m² ($P = 0.28$), respectively, more SOC_T than *Eucalyptus*. Soil N was 0.14 ($P = 0.94$) and

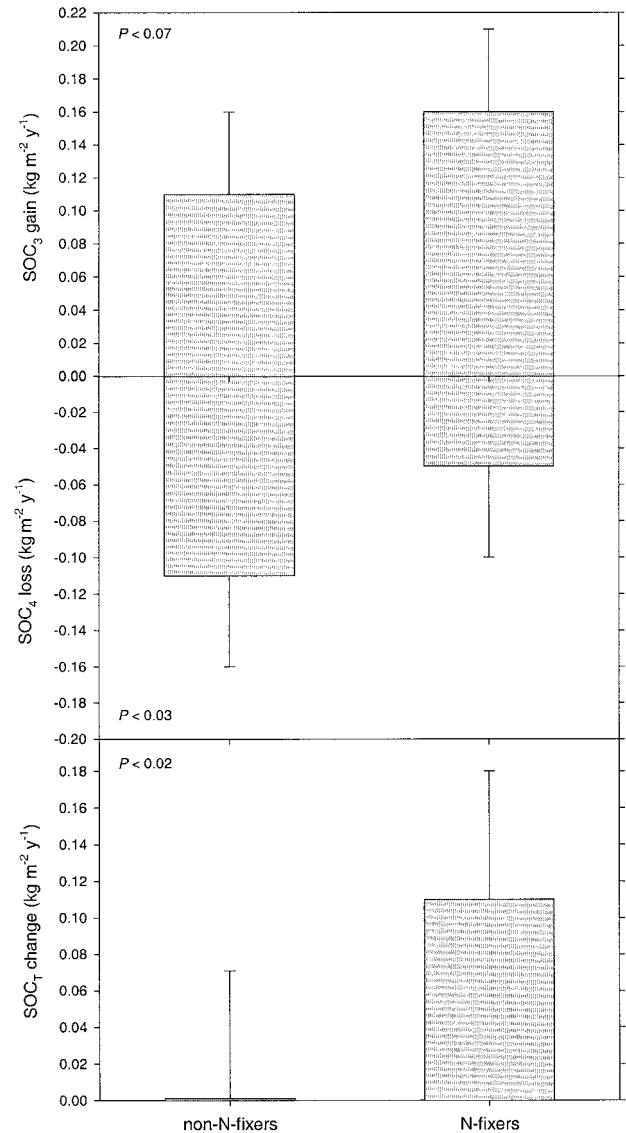


Figure 1. Aggregated SOC changes ($\text{kg C m}^{-2} \text{ y}^{-1}$) since plantation establishment across the four study sites. The error bars are standard errors of the mean using $n = 4$ for non-N-fixer species and $n = 6$ for the N-fixer species. The P values are for the test of the species-within-block split-plot effect.

0.23 kg/m^2 ($P = 0.13$) greater under *Albizia* than under *Eucalyptus* at Kamae and Chinchuck. *Leucaena* consistently had the highest SOC_T and soil N in Puerto Rico. At Lajas, *Leucaena* had 2.6 kg/m^2 more SOC_T ($P = 0.09$) and 0.22 kg/m^2 more soil N ($P = 0.05$) than *Eucalyptus*. For Toa Baja, *Leucaena* had 0.5 kg/m^2 more SOC_T ($P = 0.96$) and 0.10 kg/m^2 more soil N ($P = 0.87$) than *Eucalyptus*.

Greater SOC_T under the N-fixers resulted from a combination of both larger SOC₄ pools and larger

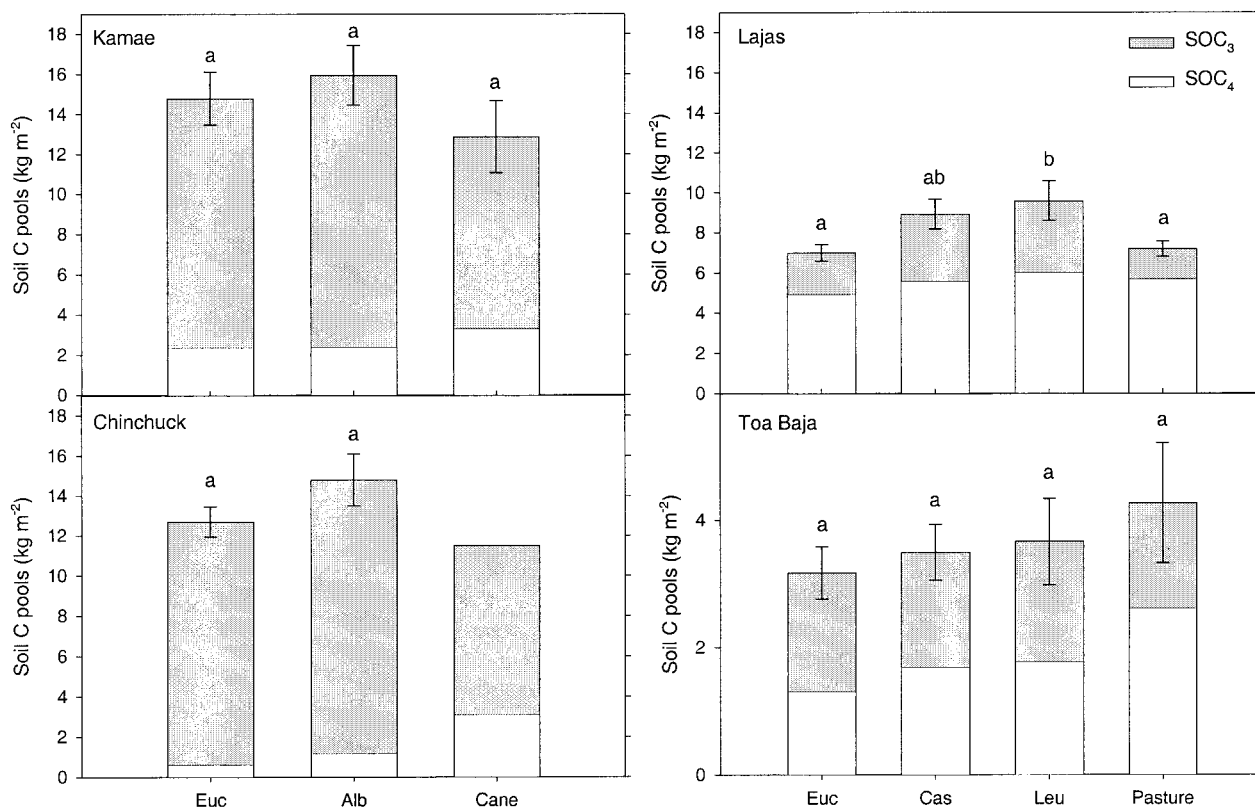


Figure 2. SOC pools to 40 cm (or 50 cm for Chinchuck) for each species at Kamae ($n = 3$ for *Eucalyptus* and sugarcane, $n = 4$ for *Albizia*), Chinchuck ($n = 4$ for *Eucalyptus* and *Albizia*), Lajas ($n = 6$ for all species), and Toa Baja ($n = 3$ for all species). The error bars are standard errors of the mean for the total SOC pool. Different letters above the bars indicate significant differences for the total SOC pool between species within a site with $P < 0.1$ (see text for specific P values). The sugarcane data for the Chinchuck site are from M. A. Bashkin (unpublished). Note the change in y-axis scale for the Toa Baja site.

SOC₃ pools, but the differences between species for these pools were not statistically significant for any of the sites. The differences in SOC₄ pools under the N-fixers compared with the *Eucalyptus* ranged from 0.04 kg/m² ($P = 1.00$) higher under *Albizia* at Kamae to 1.07 kg/m² ($P = 0.44$) higher under *Leucaena* at Lajas (Figure 2). The N-fixers, with one exception, also had greater SOC₃ pools than *Eucalyptus*, ranging from 0.04 kg/m² ($P = 1.00$) higher under *Leucaena* at Toa Baja to 1.5 kg/m² ($P = 0.18$) higher under *Leucaena* at Lajas and 1.5 kg/m² ($P = 0.27$) higher under *Albizia* at Chinchuck (Figure 2).

The SOC₃ under N-fixing trees decomposed more rapidly than the SOC₃ under *Eucalyptus*, but there were no significant changes in SOC₄ for the N-fixers or non-N-fixers during the incubation period. After 8 months of laboratory incubations, N-fixer soil lost 98 mg C₃ loss/g C₃ initial more than non-N-fixer soil, or a 68% greater loss of the SOC₃ (Figure 4). The difference in SOC₃ was significant only for the first 4 months (78 mg SOC₃ loss/g

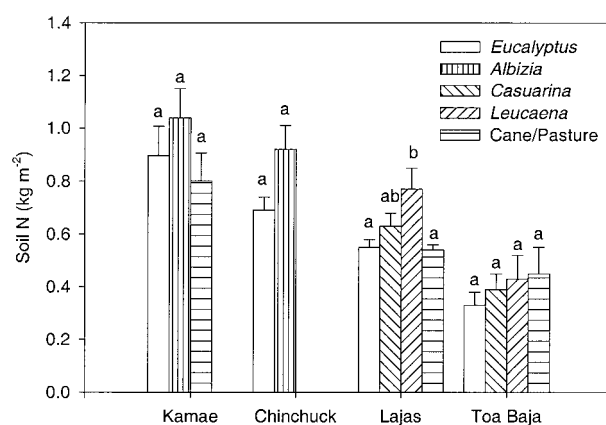


Figure 3. Soil N pools to 40 cm (or 50 cm for Chinchuck). The error bars are standard errors of the mean, sample sizes are as given in Figure 1. Different letters above the bars indicate significant differences between species within a site with $P < 0.05$ (see text for specific P values). The sugarcane data for the Chinchuck site are from M. A. Bashkin (unpublished).

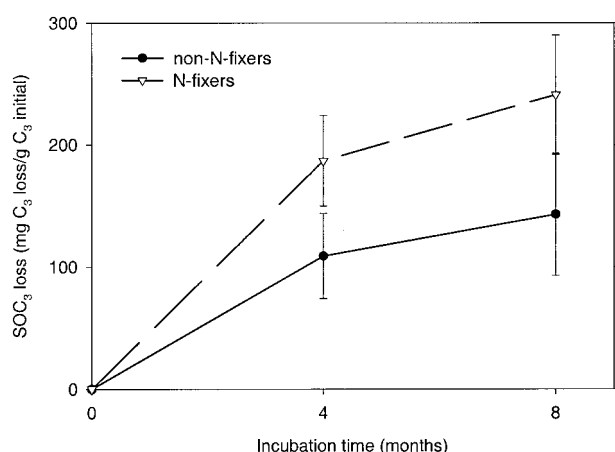


Figure 4. Cumulative loss of SOC₃ (mg SOC₃ loss/g initial SOC₃) from N-fixer and *Eucalyptus* soil during 4-month ($n = 13$) and 8-month ($n = 12$) laboratory incubations. The 4-month SOC₃ loss differed between N-fixers and *Eucalyptus* ($P = 0.05$). The error bars represent standard errors of the mean.

initial SOC₃; $P = 0.05$). Between 4 and 8 months, the loss of SOC₃ slowed for both the N-fixer and non-N-fixers soil and became more variable resulting in an overall greater loss of SOC₃ from the N-fixer soil. However, the difference was no longer significant.

DISCUSSION

SOC₄ Retention under N-fixers

Across this set of sites and species, soils beneath N-fixing trees showed greater retention of older, recalcitrant soil C (SOC₄). The higher retention of SOC₄ related well to the level of N accretion in each plot; N accretion explained 62% of the variability in SOC₄ retention under the N-fixers ($P < 0.001$) (Figure 5 top). Where the N-fixers lost soil N relative to the non-N-fixers, they also lost more SOC₄; conversely, when the N-fixers gained relatively more soil N, the N-fixers retained more SOC₄. This correlation points to a consistent C:N stoichiometry across the species and sites.

Why might retention of old carbon relate to soil N accretion? Until recently, the dominant paradigm was that N additions to soils increased the microbial decomposition of organic matter (Fog 1988). Because microbes manufacture enzymes to decompose organic substrates and enzymes require N, microbes with more N might produce more enzymes and thus speed decomposition (Aber and Melillo 1991). Studies have shown increased decomposition of litter (Harmon and others 1990; Green and

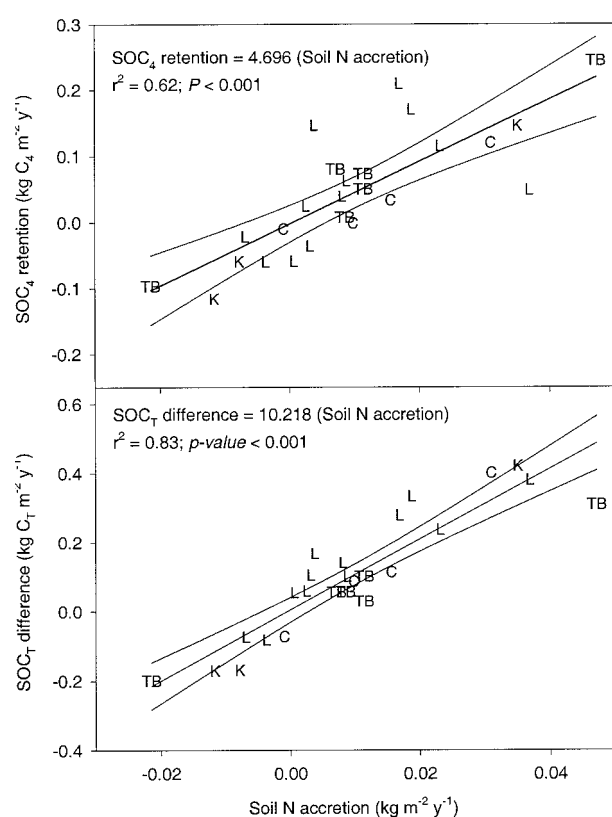


Figure 5. Soil nitrogen accretion vs SOC₄ retention (top) and SOC₄ difference under the N-fixers (calculated as the difference between N-fixer and *Eucalyptus* soil N and SOC₄ or SOC_T). Each datum is a N-fixer species within a block within a site ($n = 25$). K = Kamae site, C = Chinchuck site, L = Lajas site, and TB = Toa Baja site. Linear regression with 95% confidence intervals is shown.

others 1995) and increased microbial activity (Hart and Stark 1997; Carreiro and others 2000) following N additions (that is, urea or ammonium nitrate).

However, empirical studies have also shown that N additions can inhibit decomposition (Fog 1988; Berg and Tamm 1991; Camire and others 1991; Cole and others 1995; Prescott 1995; Arnebrant and others 1996; Carreiro and others 2000). The majority of these studies compared litter mass loss or CO₂ evolution from tissues of different species, with or without additions of exogenous N. Direct effects of high soil N supply and indirect effects of high N concentrations in litter generally increase initial decomposition rates of fresh litter (Melillo and others 1982; Berg and Matzner 1997; Berg 2000; but see Prescott 1995), but they inhibit the decomposition of humified soil C (Arnebrant and others 1996; Berg 2000). The effects of N addition on decomposition

may vary with substrate chemistry; Hobbie (2000) found that N addition speeded decomposition of low-lignin but not high-lignin leaves.

Two working hypotheses address N suppression of later phases of litter and humus decomposition (Fog 1988; Berg and Matzner 1997; Berg 1986, 2000). N additions, especially in the form of ammonium, may give a competitive edge to several cellulose decomposers (for example, bacteria) over lignocellulose decomposers (for instance, fungi), owing to the suppression of lignolytic enzyme synthesis by many species of fungi. This would favor the decomposition of labile soil organic C while inhibiting the decomposition of recalcitrant soil organic C (Carreiro and others 2000). Alternatively, low-molecular-weight N compounds (for example, amino acids, ammonium, and nitrate) may be incorporated into covalent bonds between C molecules (for example, lignin molecules and phenolics). This would create larger molecules that are resistant to microbial decomposition (Stevenson 1994). *Albizia* soils in Hawaii had lower active fungal and higher active bacterial biomass compared to *Eucalyptus* soils in a three-site comparison in Hawaii that includes the two Hawaii sites addressed in this present study (Garcia-Montiel and Binkley 1998), which lends support to the importance of changes in the microbial community.

Greater retention of old C under N-fixers has been identified in at least two other sites. Rhoades and others (1998) found 0.1 kg/m² greater old pasture C to a depth of 15 cm under individual stems and patches of an N-fixer compared with that under a non-N-fixer growing within pastures on the west slope of the Ecuadorian Andes. Kaye and others (2000) found a positive correlation ($r^2 = 0.61$; $P = 0.07$) between old soil C pools and increasing proportions of *Albizia* mixed with *Eucalyptus* in a replicated replacement series design in Hawaii. Additionally, Kaye and others (2001), using plots from the Toa Baja site with experimental additions of ¹⁵N, found a greater proportion of ¹⁵N retained in nonlabile soil pools under N-fixers compared with *Eucalyptus*, providing further evidence linking N-fixers with altered humification rates.

Across the four sites, about 55% of the SOC_T difference between the N fixer soils and non-N-fixers soils came from greater SOC₄ retention under the N-fixers. However, this proportion differed by site, ranging from 10% at Kamae to 90% at Toa Baja, with the remainder of the proportion of the SOC_T difference between the N-fixers and non-N-fixers consisting of greater SOC₃ accretion under the N-fixers. Corresponding with this shift in dom-

inance from SOC₄ retention to SOC₃ accretion, there is a site difference in average soil N content that ranges from 0.87 kg N/m² at Kamae to 0.38 kg N/m² at Toa Baja. Thus, there is a general trend across these four sites that as the base level of soil N content increases, the accretion of new C becomes the more dominant mechanism for the greater total soil C difference between N-fixers and non-N-fixers. This effect of higher base level soil N content on SOC₃ accretion is probably an increased site productivity effect.

Greater SOC₃ Accretion under N-fixers

Across the four sites, greater accretion of new SOC₃ under N-fixing trees accounted for about 45% of the SOC_T difference between N-fixer soils and non-N-fixers soils, with ranges in this proportion from 10% to 90% depending on site. Rhoades and others (1998) found that over 90% of the SOC_T difference between the N-fixer and non-N-fixer came from new N-fixer C accretion.

Greater SOC₃ accretion under the N-fixers could result from greater N-fixer C inputs to the soil, or from slower decomposition of N-fixer SOC₃ litter, or some combination of both. Our laboratory soil incubations showed equal or greater loss of SOC₃ from N-fixer soils, indicating that the larger pools of SOC₃ derived from higher inputs rather than slower decomposition of fresh litter.

There is evidence in the literature from temperate N-fixer studies for both slower long-term decomposition of N-fixer litter (Cole and others 1995; Camire and others 1991) and greater N-fixer inputs (Cole and others 1995). Binkley and Ryan (1998) found higher belowground production under *Albizia* (1.54 kg m⁻² y⁻¹) than under *Eucalyptus* (1.18 kg m⁻² y⁻¹)—although total production was similar between *Albizia* (3.88 kg m⁻² y⁻¹) and *Eucalyptus* (4.14 kg m⁻² y⁻¹)—in a three-site comparison in Hawaii that included the two sites used in the present study. These data, combined with our conclusion that C inputs explain the SOC₃ accretion show the importance of below ground inputs to soil C accretion in Hawaii. The importance of inputs from aboveground and belowground for soil C accretion may vary among sites. Parrotta (1999) measured higher aboveground net primary production for both *Leucaena* (3.09 kg m⁻² y⁻¹) and *Casuarina* (3.85 kg m⁻² y⁻¹) compared with *Eucalyptus* (2.24 kg m⁻² y⁻¹) at Toa Baja. However, without knowing belowground production for Toa Baja, we cannot speculate on the relative importance of above vs belowground control on soil C accretion.

Greater SOC_T Sequestration under N-fixers

The greater SOC_T difference under N-fixers compared with non-N-fixers resulted from the greater retention of old SOC₄ and the greater accretion of new SOC₃. The accretion of soil N explained 78% of the variability in the SOC_T difference ($P < 0.001$) (Figure 5 bottom). This study includes sites from a broad geographical area in the tropics with *Eucalyptus* as a proxy for non-N-fixers. The resulting SOC_T difference between the N-fixers and *Eucalyptus* is in agreement with N-fixer/non-N-fixer comparisons from temperate forest (Johnson 1992; Cole and others 1995) and other tropical sites (Rhoades and others 1998; Kaye and others 2000).

The varying SOC pool sizes between sites strongly reflect previous site history and probably edaphic characteristics as well (Figure 2). For example, the Hawaii sites were dominated by wild-land forests (Bashkin and Binkley 1998) before the C₄ sugarcane plantations were established in the 1920s. Thus, one would expect these Hawaii sites to have a strong influence of C₃ vegetation reflected in the proportionally larger SOC₃ to SOC₄ pools relative to the other sites. Furthermore, the magnitudes of SOC_T for these Hawaii sites are twice that of the Lajas site and four times that of the Toa Baja site. The high organic content of these Hawaii soils results from the greater ability of the amorphous clays in these Andisols to retain C than other soil types (Torn and others 1997). The higher SOC_T of the Lajas site Vertisol clays compared to the Toa Baja sandy soil probably reflects the typical pattern that soil organic C increases with increased clay content (Tiessen and others 1982; Burke and others 1989). Because this study includes multiple sites with three differing soil textures and the pattern of greater SOC_T sequestration, SOC₄ retention, and SOC₃ accretion under N-fixers remains consistent across the textural changes, we can conclude that soil texture is probably not a determinant mechanism affecting the observed changes in SOC.

While reforestation and afforestation are being considered as mechanisms to reduce C emission penalties (UNFCCC 1997), it is important to recognize that, from a soil C sequestration standpoint, tree species differ in their effects. N-fixers sequester 0.05–0.12 kg m⁻² y⁻¹ more C in their soils than non-N-fixers. Pure and mixed species forests that include N-fixers appear to be an option for soil C sequestration (Johnson 1992; Cole and others 1995; Kaye and others 2000). These findings may be relevant to countries interested in planting forests for C sequestration purposes.

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REFERENCES

- Aber JD, Melillo JM. 1991. Terrestrial ecosystems. Philadelphia: Saunders.
- Arnebrant K, Bååth E, Söderström B, Nohrsted H-Ö. 1996. Soil microbial activity in eleven Swedish coniferous forests in relation to site fertility and nitrogen fertilization. *Scand J For Res* 11:1–6.
- Bashkin MA, Binkley D. 1998. Changes in soil carbon following afforestation in Hawaii. *Ecology* 79:828–33.
- Berg B. 2000. Litter decomposition and organic matter turnover in northern forest soils. *For Ecol Manage* 133:13–22.
- Berg B. 1986. Nutrient release from litter and humus in coniferous forest soils—a mini review. *Scand J For Res* 1:359–69.
- Berg B, Matzner E. 1997. Effect of N deposition on decomposition of plant litter and soil organic matter in forest systems. *Environ Rev* 5:1–25.
- Berg B, Tamm CO. 1991. Decomposition and nutrient dynamics of litter in long-term optimum nutrition experiments. I. Organic matter decomposition in Norway spruce (*Picea abies*) needle litter. *Scand J For Res* 6:305–21.
- Binkley D. 1997. Bioassays of the influence of *Eucalyptus saligna* and *Albizia falcataria* on soil nutrient supply and limitation. *For Ecol Manage* 91:229–34.
- Binkley D. 1983. Ecosystem production in Douglas-fir plantations: interaction of red alder and site fertility. *For Ecol Manage* 5:215–27.
- Binkley D. 1996. The influence of tree species on forest soils: processes and patterns. In: Mead DJ, Cornforth IS, (editors). *Proceedings of the Trees and Soils Workshop*. Special publication no. 10. Canterbury: Agronomy Society of New Zealand. p 1–33.
- Binkley D, Cromack K Jr, Fredriksen RL. 1982. Nitrogen accretion and availability in some snowbrush ecosystems. *For Sci* 28:720–4.
- Binkley D, Giardina C. 1997. Nitrogen fixation in tropical forest plantations. In: Sadanandan Nambiar EK, Brown AG, (editors). *Management of soil, nutrients and water in tropical plantation forests*. ACIAR monograph no. 43. Canberra: CSIRO.

- Binkley D, Giardina C. 1998. Why do tree species affect soils? The warp and woof of tree-soil interactions. *Biogeochemistry* 42:89–106.
- Binkley D, Resh SC. 1999. Rapid changes in soils following *Eucalyptus* afforestation in Hawaii. *Soil Sci Soc Am J* 63:222–5.
- Binkley D, Ryan MG. 1998. Net primary production and nutrient cycling in replicated stands of *Eucalyptus saligna* and *Albizia falcataria*. *For Ecol Manage* 112:79–85.
- Binkley D, Sollins P. 1990. Factors determining differences in soil pH in adjacent conifer and alder-conifer stands. *Soil Sci Soc Am J* 54:1427–33.
- Brown S, Lugo AE, Iverson LR. 1992. Processes and lands for sequestering carbon in the tropical forest landscape. *Water Air Soil Poll* 64:139–55.
- Bruce JP, Frome M, Haites E, Janzen H, Lal R, Paustian K. 1999. Carbon sequestration in soils. *J Soil Water Conserv* 54:382–9.
- Burke IC, Yonker CM, Parton WJ, Cole CV, Flach K, Schimel DS. 1989. Texture, climate, and cultivation effects on soil organic matter content in U.S. grassland soils. *Soil Sci Soc Am J* 53:800–5.
- Camiré C, Côté B, Brulotte S. 1991. Decomposition of roots of black alder and hybrid poplar in short-rotation plantings: nitrogen and lignin control. *Plant Soil* 138:123–32.
- Carreiro MM, Sinsabaugh RL, Repert DA, Parkhurst DG. 2000. Microbial enzyme shifts explain litter decay responses to simulated nitrogen deposition. *Ecology* 81:2359–65.
- Cerri CC, Feller C, Balesdent J, Victoria R, Plenecassagne A. 1985. Application du traçage isotopique naturel en ^{13}C , à l'étude de la dynamique de la matière organique dans le sols. *Comptes Rendus Acad Sci Paris T.300, II*, 9:423–8.
- Cole DW, Compton JE, Edmonds RL, Homann PS, Van Miegroet J. 1995. Comparison of carbon accumulation in Douglas fir and red alder forests. In: McFee WW, Kelly JM, (editors). Carbon forms and functions in forest soils. Madison (WI): Soil Science Society of America. pp. 527–546.
- Drinkwater LE, Wagoner P, Sarrantonio M. 1998. Legume-based cropping systems have reduced carbon and nitrogen losses. *Nature* 396:262–5.
- Ehleringer JR. 1991. $^{13}\text{C}/^{12}\text{C}$ fractionation and its utility in terrestrial plant studies. In: Coleman DC, Fry B, editors. Carbon isotope techniques. San Diego: Academic Press. pp. 187–200.
- Fog K. 1988. The effect of added nitrogen on the rate of decomposition of organic matter. *Biol Rev* 63:433–62.
- Garcia-Montiel DC, Binkley D. 1998. Effect of *Eucalyptus saligna* and *Albizia falcataria* on soil processes and nitrogen supply in Hawaii. *Oecologia* 113:547–56.
- Green CJ, Blackmer AM, Horton R. 1995. Nitrogen effects on conservation of carbon during corn residue decomposition in soil. *Soil Sci Soc Am J* 59:453–9.
- Harmon ME, Baker GA, Spycher G, Greene SE. 1990. Leaf-litter decomposition in the *Picea/Tsuga* forests of Olympic National Park, Washington, U.S.A. *For Ecol Manage* 31:55–66.
- Hart SC, Stark JM. 1997. Nitrogen limitation of the microbial biomass in an old-growth forest soil. *Ecoscience* 4:91–8.
- Hobbie SE. 2000. Interactions between litter lignin and soil nitrogen availability during leaf litter decomposition in a Hawaiian montane forest. *Ecosystems* 3:484–94.
- Houghton RA. 2000. Annual fluxes of carbon from deforestation and regrowth in the Brazilian Amazon. *Nature* 403:301–4.
- Houghton RA. 1994. The worldwide extent of land-use change. *BioScience* 44:305–13.
- Johnson DW. 1992. Effects of forest management on soil carbon storage. *Water Air, Soil Poll* 64:83–120.
- Kaye JP, Binkley D, Zou X, Parrotta JA. 2002. Non-labile soil ^{15}N retention beneath three tree species in a tropical plantation. *Soil Sci Soc Am J* 66: in press.
- Kaye JP, Resh SC, Kaye MW, Chimner RA. 2000. Nutrient and carbon dynamics in a replacement series of *Eucalyptus* and *Albizia* trees. *Ecology* 81:3267–73.
- Little EL Jr, Skolmen RG. 1989. Common forest trees of Hawaii (native and introduced). Agriculture handbook no. 679. Washington (DC): USDA Forest Service.
- Lugo AE, Wang D, Bormann FH. 1990. A comparative analysis of biomass production in five tropical tree species. *For Ecol Manage* 31:153–66.
- Melillo JM, Aber JD, Muratore JF. 1982. Nitrogen and lignin control of hardwood leaf litter decomposition dynamics. *Ecology* 63:621–6.
- Moffat AS. 1997. Resurgent forests can be greenhouse gas sponges. *Science* 277:315–6.
- Nadelhoffer KJ, Emmett BA, Gunderson P, Koopmans OJ, Schleppi P, Tietema A, Wright RF. 1999. Nitrogen deposition makes a minor contribution to carbon sequestration in temperate forests. *Nature* 398:145–8.
- Parrotta JA. 1993. *Casuarina equisetifolia* L. ex J. R. & G. Forst. Silvics of forest trees of the American tropics. SO-ITF-SM-56. New Orleans: US Department of Agriculture, Forest Service, Inst. Trop. For., Southern Forest Exp. Stn.
- Parrotta JA. 1992. *Leucaena leucocephala* (Lam.) de Wit. Silvics of forest trees of the American tropics. SO-ITF-SM-56. New Orleans: US Department of Agriculture, Forest Service, Inst. Trop. For., Southern Forest Exp. Stn.
- Parrotta JA. 1999. Productivity, nutrient cycling, and succession in single- and mixed-species plantations of *Casuarina equisetifolia*, *Eucalyptus robusta*, and *Leucaena leucocephala* in Puerto Rico. *For Ecol Manage* 124:45–77.
- Parrotta JA, Baker DD, Fried M. 1994. Application of ^{15}N -enrichment methodologies to estimate nitrogen fixation in *Casuarina equisetifolia*. *Can J For Res* 24:201–7.
- Parrotta JA, Baker DD, Fried M. 1996. Changes in dinitrogen fixation in maturing stands of *Casuarina equisetifolia* and *Leucaena leucocephala*. *Can J For Res* 26:1684–91.
- Paustian K, Andren O, Janzen H, Lal R, Smith P, Tian G, Tiessen H, van Noordwijk M, Woomer P. 1997. Agricultural soil as a C sink to offset CO_2 emissions. *Soil Use Manage* 13:230–44.
- Post WM, Kwon KC. 2000. Soil carbon sequestration and land-use change: processes and potential. *Global Change Biol* 6:317–27.
- Prescott CE. 1995. Does nitrogen availability control rates of litter decomposition in forests? *Plant Soil* 168:83–8.
- Resh SC. 1999. Nitrogen-fixing tropical trees sequester soil carbon [dissertation]. Fort Collins (CO): Colorado State University.
- Rhoades CC, Eckert GE, Coleman DC. 1998. Effect of pasture trees on soil nitrogen and organic matter: implications for tropical montane forest restoration. *Restor Ecol* 6:262–70.
- Richter DD, Markewitz D, Trumbore SE, Wells CG. 1999. Rapid accumulation and turnover of soil carbon in an aggrading forest. *Nature* 400:56–8.
- Schimel D, Braswell BH, Holland EA, McKeown R, Ojima DS, Painter TH, Townsend AR. 1994. Climatic, edaphic, and biotic controls over storage and turnover of carbon in soils. *Global Biogeochem Cycles* 8:279–93.

- Schlesinger WH. 1997. Biogeochemistry: an analysis of global change. 2nd ed. San Diego: Academic Press. p. 156.
- Schlesinger WH. 1999. Carbon sequestration in soils. *Science* 284:2095.
- Schlesinger WH. 1990. Evidence from chronosequence studies for low carbon-storage potential of soils. *Nature* 348:232–24.
- Stevenson FJ. 1994. Humus chemistry: genesis, composition, reactions. 2nd ed. New York: Wiley.
- Subbarao NS, Rodríguez-Barrueco C. 1995. Casuarinas. Lebanon (NH): Science Publishers.
- SYSTAT. 1997. SYSTAT 7.0 for Windows. Chicago: SPSS.
- Tarrant RF, Miller RE. 1963. Accumulation of organic matter and soil nitrogen beneath a plantation of red alder and Douglas-fir. *Soil Sci Soc Am Proc* 27:231–4.
- Tiessen H, Stewart JWB, Bettany JR. 1982. Cultivation effects on the amounts and concentration of carbon, nitrogen, and phosphorus in grassland soils. *Agron J* 74:831–5.
- Torn MS, Trumbore SE, Chadwick OA, Vitousek PM, Hendricks DM. 1997. Mineral control of soil organic carbon storage and turnover. *Nature* 389:170–3.
- Turk D, Keyser HH. 1992. Rhizobia that nodulate tree legumes: specificity of the host for nodulation and effectiveness. *Can J Microbiol* 38:451–60.
- [UNFCCC] United Nations Framework Convention on Climate Change. 1997. Kyoto Protocol to the United Nations Framework Convention on Climate Change. Available on the Internet: <http://www.cop3.de/>
- Van Cleve K, Powers RF. 1995. Soil carbon, soil formation, and ecosystem development. In: McFee WW, Kelly JM, editors. Carbon forms and functions in forest soils. Madison (WI): Soil Science Society of America.
- Vitousek PM. 1991. Can planted forest counteract increasing atmospheric carbon dioxide? *J Environ Qual* 20:348–54.
- Wang D, Bormann FH, Lugo AE, Bowden RD. 1991. Comparison of nutrient-use efficiency and biomass production in five tropical tree taxa. *For Ecol Manage* 46:1–21.
- Zabala NQ, editor. 1997. International Workshop on Albizia and Paraserianthes species. Forest, farm, and community tree research reports. Special issue. Morrilton (AK): Winrock International.