### Carbon Sequestration in Response to Grassland– Shrubland–Turfgrass Conversions and a Test for Carbonate Biomineralization in Desert Soils, New Mexico, USA

#### **Jiaping Wang**

College of Agronomy Shihezi Univ. Shihezi 832003, China

State Key Laboratory of Desert and Oasis Ecology Xinjiang Institute of Ecology and Geography Chinese Academy of Sciences Urumqi 830011 China

#### **Curtis Monger\***

New Mexico State Univ. Las Cruces, NM 88001

National Soil Survey Center USDA Lincoln, NE 68508

#### Xiujun Wang

State Key Laboratory of Desert and Oasis Ecology Xinjiang Institute of Ecology and Geography Chinese Academy of Sciences Urumqi 830011 China

Earth System Sci. Interdisciplinary Center Univ. of Maryland College Park, MD 20740

#### Matteo Serena Bernhard Leinauer

New Mexico State Univ. Las Cruces, NM 88001

#### **Core Ideas**

- Soil carbonate is typically viewed as a soil mineral that requires centuries to accumulate; our study indicates that it can be generated in months to years.
- If the source of Ca is directly from silicate minerals, soil carbonate could be managed to sequester atmospheric CO<sub>2</sub>.
- Carbon-13 values can change in less than a decade for both soil organic and inorganic C.
- This study uses an experimental pedology approach using controls, treatments, and replications.

This study uses an experimental pedology approach to examine (i) how the conversion of native C<sub>4</sub> grassland to C<sub>3</sub> woody shrubs then to irrigated C<sub>4</sub> turfgrass affects both soil organic C (SOC) and soil inorganic C (SIC) and (ii) whether SIC can be enhanced by microbial biomineralization. Three sites were studied in the Chihuahuan Desert of New Mexico. At each site, SOC, SIC, and their  $\delta^{13}$ C values were measured on control soil samples and compared to samples treated with liquid growth medium and Ca(OH)2 at 10 and 40 cm. The treated samples were left to react for 1 mo in February, May, and August of 2014. Using a space-for-time substitution, soil organic C decreased when native grasslands converted to desert shrubs, then sharply increased after desert shrubs were converted to irrigated turfgrass. Most surprising, however, was the increase of SIC in the turfgrass site, which doubled in 6 yr. The  $\delta^{13}$ C values of both SOC and SIC reflected the change from  $C_4$  to  $C_3$  then back to C<sub>4</sub> vegetation and showed how rapidly SOC and SIC can change their isotopic signatures. Soil inorganic C formation was slightly higher for the liquid growth medium, but no statistically significant differences were observed between the treatments and control samples. In addition, no biomineralization was observed with microscopy, perhaps because the 1-mo reaction time was too short and the amount applied was too small. Although SIC is typically viewed as a soil mineral that requires centuries to accumulate, our study indicate that SIC can be generated in months to years if the soil environment is suitable.

Abbreviations: SIC, soil inorganic C; SOC, soil organic C.

equestering C as SOC has achieved widespread recognition at high-profile climate conferences (Koch et al., 2015). Sequestering C as SIC, however, has achieved much less attention despite the fact that SIC is the second largest terrestrial C pool, containing at least 940 Pg C (Eswaran et al., 2000) compared to SOC's > 1500 Pg C (Post et al., 1982; Schlesinger and Bernhardt, 2013). Soil inorganic C is ubiquitous in dryland soils where it occurs at roughly 10 times the amount of SOC (Schlesinger, 1982).

In addition to SIC's natural occurrence in desert soils, recent studies have measured increases in SIC as a result of agronomic management practices. In arid croplands in northwestern China, for example, it was documented that more C was sequestrated as SIC than SOC (Wu et al., 2009; Wang et al., 2014a, 2014b; Bughio et al., 2016). In Russia, chernozems used to grow crops contained more than twice the amount of SIC than chernozems still in native grass (Mikhailova and Post, 2006). In the US, SIC in the Imperial Valley increased after 85 yr of irrigated agriculture (Wu et al., 2008). Historically, however, SIC has been less thoroghtly studied than SOC, probably because of the view that SIC (also known as CaCO<sub>3</sub> or calcite) is a soil mineral that remains unchanged by agronomic, rangeland, and forestry management techniques (Lal, 2004, 2008; Monger, 2014).

Soil Sci. Soc. Am. J. 80:1591–1603 doi:10.2136/sssaj2016.03.0061 Received 3 Mar. 2016. Accepted 19 Aug. 2016.

 $*Corresponding\ author\ (\underline{curtis.monger@lin.usda.gov})$ 

© Soil Science Society of America, 5585 Guilford Rd., Madison WI 53711 USA. All Rights reserved.

Not only has SIC formation been suggested to occur more rapidly than traditionally thought, SIC is more biological than traditionally thought. Evidence to support a biological origin of SIC has come from (i) micromorphology studies of field specimens (Drees and Wilding, 1987; Klappa, 1979; Phillips et al., 1987 Verrecchia and Verrecchia, 1994), (ii) lab studies of soil microbes grown on agar (Boquet et al., 1973), (iii) soil column studies comparing soils inoculated with microorganisms versus sterile soils (Monger et al., 1991), and (iv) experimental micropedology studies using apparatus containing a liquid growth medium inserted in the soil (Khormali et al., 2014). This biological aspects of carbonate formation by microorganisms have been increasingly used for practical applications, such as the production of biocement to protect ornamental stone (Rodriguez-Navarro et al., 2003), to strengthen construction material (De Muynck et al., 2010), and to sequester atmospheric CO<sub>2</sub> (DeJong et al., 2010).

For C sequestration purposes, knowing whether SIC was formed in situ rather than being a detrital component of the parent material is important (Eshel et al., 2007). However, it is more essential to know the source of Ca in carbonates (Schlesinger, 1999). Calcium derived directly from silicate minerals (termed silicatic pedogenic carbonate) is a prerequisite for C sequestration, in contrast to Ca derived from preexisting carbonate minerals (termed calcitic pedogenic carbonate) (Monger et al., 2015).

This study uses experimental pedology to examine SOC and SIC sequestration in desert soils as a function of vegetation change and microbial biomineralization. Pedology per se is the study of soils as a natural phenomenon based on observational evidence and characterization data. Experimental pedology adds manipulative experiments with treatments, controls, and replications to test pedological hypotheses (e.g., Hallsworth and Crawford, 1965).

Specifically, our study addresses three questions: (i) Do SOC, SIC, and their  $\delta^{13}$ C values change when native  $C_4$  grass is replaced with  $C_3$  desert shrubs, which then are converted to irrigated  $C_4$  turfgrass? (ii) Can SIC be generated by microbial biomineralization when liquid growth medium is added to the soil in situ? (iii) If so, will the amount of SIC produced by biomineralization be greater than the amount of SIC produced by chemical precipitation from additions of Ca(OH)<sub>2</sub>?

## MATERIALS AND METHODS Study Site Description

The study was conducted in the southwestern US in southern New Mexico (Fig. 1). Site 1 (the native grass site) was located at the Chihuahuan Desert Rangeland Research Center Site 2 (the desert shrub site) was located in an undeveloped area with native vegetation on the New Mexico State University campus, and Site 3 (the turfgrass site) was located roughly 100 m east of the desert shrub site. The annual rainfall in the study region is 200 to 250 mm per year, with more than 50% falling in the summer monsoon season. The average air temperature is recorded as

17°C, giving rise to soils with aridic moisture and thermic temperature regimes (Gile et al., 1981).

At the native grass site, the dominant plant species is black grama [Bouteloua eriopoda (Torr.) Torr.], which uses the  $C_4$  photosynthetic pathway and has a C isotopic value of -14.0%  $\pm$  0.6 (Gallegos, 1999). This isolated grassland, which is only a few km² in size, is a vestige of more expansive grasslands that existed a century and a half go in the region (Buffington and Herbel, 1965; Gibbens et al., 2005). This site also contains sparse amounts of  $C_3$  plants, such as soaptree yucca [Yucca elata (Engelm.) Engelm.)] and Mormon tea (Ephedra torreyana S. Watson) separated from each other by tens of meters. In addition,  $C_3$  succulents appear at various times when weather conditions are favorable. Thus, some  $C_3$  C is put into the soil at this site but the vast amount comes from  $C_4$ .

At the desert shrub site, the dominant plant species are creosotebush [Larrea tridentate (Sessé & Moc. ex DC.) Coville] and mesquite (Prosopis glandulosa Torr.) separated by bare ground with sparse cacti (Opuntia spp.) and small clumps of grasses (Aristida spp. and Bouteloua spp.), which appear after prolonged rains in the summer monsoon season. Both creosotebush and mesquite use the  $C_3$  photosynthetic pathway and have isotopic values of -24.2%  $\pm$  0.7 and -25.3%  $\pm$  0.4, respectively (Gallegos, 1999). However, according to land survey notes, the study region was dominated by native grasslands until the late 1800s, when woody shrubs replaced most of the grasses as a consequence of overgrazing and climate change (Buffington and Herbel, 1965; Gile, 1966; Peters et al., 2012). Thus we make the inference that the desert shrub site was dominated by  $C_4$  grasses 150 yr ago.

At the turfgrass site, a dense stand of bermudagrass (Cynodon dactylon L.) was established in 2008 (Serena et al., 2014). Bermudagrass, like black grama, uses the C<sub>4</sub> photosynthetic pathway. The plot was irrigated daily with a pop-up sprinkler system (Hunter Rotator MP2000-90, Hunter Industries, San Marcos, CA) at 85% of reference evapotranspiration for short grass (Snyder and Eching, 2007), which amounted to 1200 mm during 2014. The irrigation water at that time had a pH of 7.6, an electrical conductivity of 0.6 dS m<sup>-1</sup>, and a Na adsorption ratio of 1.9 mmol<sup>1/2</sup> L<sup>-1</sup>. Other chemical constituents included Ca (2.6 mmol<sub>c</sub> L<sup>-1</sup>), Mg (0.8 mmol<sub>c</sub> L<sup>-1</sup>), Na (2.1 mmol<sub>c</sub> L<sup>-1</sup>), and bicarbonate (2.2 mmol<sub>c</sub> L<sup>-1</sup>). During the research period, bermudagrass was mowed weekly to 2.0 cm with clippings removed and fertilizer applied to prevent nutrient stress. Fertilization was applied granularly in the form of urea and muriate of potash by means of a Gandy walk-behind drop spreader (Model 36H13; Gandy Corp., Owatonna, MN). Fertilization consisted of an annual total of 15 g N, 0 g P, and 4.2 g K m<sup>-2</sup> applied every 6 wk at 3.75 g N m<sup>-2</sup>, 0 g P and 1.04 g K m<sup>-2</sup> from April to September.

Except for vegetation, the five soil-forming factors were held constant for the three sites as much as possible, especially soil age which has a major effect on the amount of SIC in the soil profile. The landform at the native grassland site is a fan-skirt and the geomorphic surface is the mid-Holocene age Organ surface (Gile et al., 1981). The landform at the desert shrub and adjacent turfgrass sites is a valley border terrace and the geomorphic

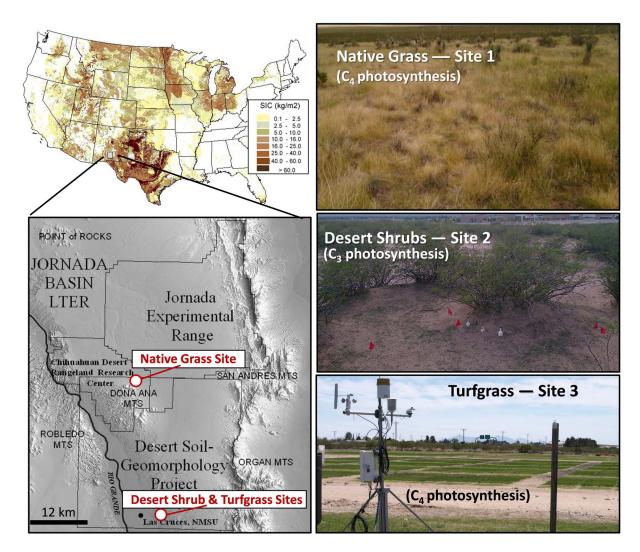


Fig. 1. Locations and photographs of the three study sites. Photosynthetic pathways of dominant vegetation: Site 1,  $C_4$ ; Site 2,  $C_3$ ; Site 3,  $C_4$ . The upper left map of soil inorganic C is from Guo et al. (2006).

surface is the mid-Holocene Fillmore surface. Both geomorphic surfaces are characterized by having soils with weakly developed profiles. The physical properties of the soils are shown in Table 1. The parent material for both sites is alluvium dominated by igneous mineralogy, mainly quartz. Limestone detritus is not a component of the parent material at either site. Soil at the native grass site is classified as a coarse-loamy, mixed, superactive,

thermic Ustic Haplocambid. Soil at both the  $\rm C_3$  desert shrub and  $\rm C_4$  turfgrass sites are classified as mixed, thermic Typic Torripsamments.

## Experimental Design, Treatments, and Sample Collection

At each study site, soil samples were collected at 10 and 40 cm from three randomly located auger holes (15 cm in diameter) (Fig. 2). As soil was removed, it was placed in labeled buckets so it could be replaced at the depths from which it had been taken. Soil samples to be treated with the liquid growth medium and Ca(OH)<sub>2</sub> were passed through a 2-mm sieve, poured into four porous

plastic cylinders (3 by 5 cm), and then wrapped with nylon mesh fabric that was stretched to make holes large enough (>4  $\mu$ m) to allow fungal hyphae to penetrate but small enough to keep soil from spilling out. Of the four samples collected at each depth, two were treated with B4 growth medium, one was treated with Ca(OH)<sub>2</sub>, and one was treated with deionized water to serve as a control.

Table 1. Basic soil properties at study sites.

Site	Depth	Horizon	Sand	Silt	Clay	<b>SOC</b> §	SIC§	рΗ
	cm				_%_			
Native grass soilt	0-17	Α	79	14.1	6.9	0.26	-	7.1
	17-45	Bw1	83.2	11.3	5.5	0.12	-	7.7
	45-68	Bw2	79.9	13.8	6.3	0.14	trace	8.2
	58-122	Bk2	79.1	14	6.9	0.03	1	8.6
Desert shrub and turfgrass soil‡	0-13	Α	90	4.9	5.1	0.14	0.1	8.6
	13-43	В	90.1	4.8	5.1	0.11	0.5	8.6
	43-64	C1ca	91.7	3.4	4.9	0.11	2.7	8.4
	64–114	C2ca	93.7	2.9	3.4	0.05	2.4	8.4

<sup>†</sup> Data for the native grass soil are courtesy of the USDA-Natural Resources Conservation Services (S04NM013–006).

<sup>‡</sup> Data for the desert shrub and turfgrass sites are from Gile et al. 1981.

<sup>§</sup> SOC, soil organic C; SIC, soil inorganic C.

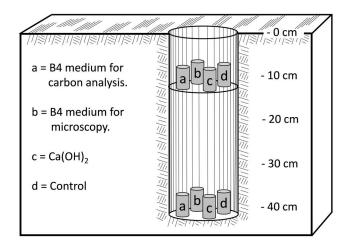


Fig. 2. Illustration showing where the porous plastic sample cylinders (3 by 5 cm) were placed in the soil. The sample cylinders were separated by at least 5 cm. The auger holes are 15 cm in diameter and cylinders were randomly distributed throughout the study sites.

The liquid B4 growth medium consisted of 15 g calcium acetate, 4.0 g yeast extract, and 10.0 g glucose per 1000 mL of distilled water, adjusted to pH 8.0 with NaOH (Boquet et al., 1973). The B4 liquid was applied aseptically by pipetting 5 mL of the sterile liquid onto the soil in the plastic container beneath the nylon mesh fabric. One of the two B4-treated samples was used for measuring SOC, SIC, and C isotopic values. The second was used for microscopy to determine if calcified microorganisms were present. For the microscopy examination, binocular microscopes was used and 1 M hydrochloric acid was added with an eye dropper to look for effervescence. Promising zones near but not in contact with the acid were then examined via scanning electron microscopy.

The  $\text{Ca}(\text{OH})_2$  treatment was also applied by pipetting a solution beneath the nylon mesh fabric. In this case, 5 mL of a  $\text{Ca}(\text{OH})_2$  suspension was added to the soil sample. The  $\text{Ca}(\text{OH})_2$  suspension was made by adding 7.4 g per 1000 mL of deionized water and was shaken to get the  $\text{Ca}(\text{OH})_2$  particles uniformly suspended before applying 5 mL to the soil sample. This method was determined empirically to supply the same amount of Ca as the B4 treatment. The control sample was treated with 5 mL of deionized water.

The rationale for using 5 mL for the B4 and  $Ca(OH)_2$  treatments consisted of the following. First, the water holding capacity of the loamy sand textures of the soil samples could retain only 5 mL. Greater amounts dripped out the bottoms of the porous plastic containers. Second, the  $Ca(OH)_2$  concentration of 7.4 g per 1000 mL of deionized water supplied the correct amount of Ca to generate approximately the same amount of  $CaCO_3$  as the B4 treatment. Under the assumption that all the Ca reacted, the  $CaCO_3$  accumulation rates of soils with these treatments would be similar: 22.06 g C m<sup>-2</sup> yr<sup>-1</sup> for the B4 liquid medium and 23.24 g C m<sup>-2</sup>yr<sup>-1</sup> for the  $Ca(OH)_2$  suspension.

The treated soils were returned to the holes at the depths where they were taken, allowed to react for 1 mo, and then harvested and taken to the lab for analysis. We assume that no cross-

contamination occurred between the samples after they were replaced in the soil, since only 5 mL of the treatment solution was used, which was readily absorbed into the soil sample, and the samples were at least 5 cm apart.

The experiment was conducted three times, each lasting 1 mo, during three seasons. The first experiment took place from 10 February to 10 March during the relatively cold dormant winter season. The second experiment took place from 10 May to 8 June during the dry, windy spring season. The third experiment took place from 4 August to 5 September during the hot, wet monsoon season.

The rationale for choosing a duration of 1 mo was based on two observations. First, carbonate crystals typically begin forming within a few days when soil microbes are cultured on solid B4 medium (Boquet et al., 1973) and within a few weeks in soil columns (Monger et al., 1991). Therefore, a 1-mo period was assumed to be ample time for carbonate to form. Second, it took about 2 mo to process and analyze the samples after each experiment.

#### Soil Organic C, SIC, and $\delta^{13}$ C Analysis

Once harvested, the soil samples used for SOC, SIC, and  $\delta^{13}$ C analysis were air-dried, weighed, and then crushed with a shatter box until the particles were smaller than 0.25 mm. Soil organic C samples were treated with 1 M HCl to remove carbonate, washed with deionized water three times, and oven-dried. The 1 M HCl was not strong enough to affect SOC (Midwood and Boutton, 1998). Samples were weighed (1–2 mg), placed into tin capsules, and combusted in a C–N analyzer (EuroVector, Redavalle, Italy) connected to a continuous-flow isotope ratio mass spectrometer (IsoPrime-EA, Micromass, Stockport, UK). The  $\delta^{13}$ C values, which record the photosynthetic pathways of major plant groups and the effects of pCO<sub>2</sub> (Schubert and Jahren, 2012), were expressed relative to the carbon isotopic composition of Vienna-PeeDee Belemnite. The standards used were the carbonate NBS-19 +1.95‰ and graphite USGS24 -16.049‰.

Other subsamples were not treated with acid, which provided a measure of total C (TC) and its  $\delta^{13}$ C values. The SOC was corrected by the weight loss from the acid treatment. Soil inorganic C was obtained from the difference between TC and SOC. The SIC  $\delta^{13}$ C values were determined via the following equation:

$$\delta^{13}C_{SIC} = \frac{TC \times \delta^{13}C_{TC} - SOC \times \delta^{13}C_{SOC}}{TC - SOC}$$
[1]

The validity of this method was tested and confirmed using known amounts of carbonate and graphite standards mixed in various proportions. The fraction of  $C_4$  carbon in SOC ( $F_{C4}$ ) was estimated by the following equation (Boutton et al., 1999; Weems and Monger, 2012):

$$F_{C4} = \frac{\delta_{\text{sample}} - \delta_{C3}}{\delta_{C4} - \delta_{C3}}, \qquad [2]$$

where  $\delta_{sample}$  is the  $\delta^{13}C$  of the bulk soil sample,  $\delta_{C3}$  is the average  $\delta^{13}C$  value of  $C_3$  plants,  $\delta_{C4}$  is the average  $\delta^{13}C$  value of the

 $C_4$  plants. Values of -25‰ were used for  $C_3$  plants and -14‰ for  $C_4$  plants. The  $C_4$  fraction based on pedogenic carbonate was estimated via Eq. [3]:

$$F_{\rm C4} = \frac{\delta_{\rm sample ped carb} - \delta_{\rm C3 ped carb}}{\delta_{\rm C4 ped carb} - \delta_{\rm C3 ped carb}} \ , \eqno [3]$$

where  $\delta_{samplepedcarb}$  is the  $\delta^{13}C$  of the carbonate in the soil sample,  $\delta_{C3ped\ carb}$  is the average  $\delta^{13}C$  value derived from  $C_3$  plants, and  $\delta_{C4pedcarb}$  is the average  $\delta^{13}C$  value derived from  $C_4$  plants. Carbonate  $\delta^{13}C$  values of -12‰ were used for  $C_3$  plants and +2‰ for  $C_4$  plants.

#### **Soil CO<sub>2</sub> and Temperature Measurements**

Soil CO2 levels were determined by using gas wells using a Vaisala CARBOCAP Hand-Held Carbon Dioxide Meter GM70 (Vaisala Oyj, Helsinki, Finland), which has a probe accuracy of  $\pm 1.5\%$ . Gas wells were composed of polyvinylchloride pipe (12.7 mm i.d.) inserted into the soil at 10 and 40 cm depths. The end of the inserted pipe was open and had several small (2 mm) drill holes spaced 6 mm apart in the side walls up to 3 cm from the bottom (Buyanovsky and Wagner, 1983). The holes were then covered with a porous nylon cloth to keep the inside of the pipe clean. The top of each well was capped and a hose-barb was screwed into the cap. A 4-cm piece of neoprene tubing (3 mm i.d.) was attached to the barb and crimped with a clip to keep the well airtight. When a reading was taken, a Nafion membrane tube (Perma Pure, Lakewood, NJ) was attached to the crimped tube, the clip was removed, and soil air was pumped into the chamber for a CO<sub>2</sub> reading. Three readings were taken and averaged for the desert shrub and turfgrass sites and two readings for the native grass site. To determine diurnal CO<sub>2</sub> fluctuations, measurements were made at 3-hr intervals for 24 h on 26 May and 15 August.

Soil temperature was measured using NexSens micro-T temperature sensors (iButton Thermochron DS1921G, Maxim Integrated, San Jose, CA). Readings were made at 1-hr increments and averaged for month-long periods for the durations when the sensors were in the ground. The sensors were placed at 10 and 40 cm depths.

#### **Calculations and Statistical Analysis**

Soil inorganic C accumulation rates ( $R_{SIC}$ , in mg C m<sup>-2</sup> d<sup>-1</sup>) were measured by the following equation:

$$R_{\text{SIC}} = \frac{\left(\text{SIC}_{i} \times E_{i} - \text{SIC}_{CK} \times E_{CK}\right) \times H \times 10,000}{D} \qquad [4]$$

where  $SIC_i$  is the SIC content of the B4 or  $Ca(OH)_2$  treatments  $(g kg^{-1})$ ,  $SIC_{CK}$  is the SIC content of the control,  $E_i$  is the soil bulk density  $(g cm^{-3})$ , H is the length (1 cm), D is the duration of the experiment (days), and 10,000 is unit conversion factor.

ANOVAs were performed to evaluate the effect of soil temperature, soil CO<sub>2</sub>, and precipitation on SIC accumulation. Following the ANOVAs, Fisher's protected LSD was used to compare SIC and isotopic values between treatments. Student's

*t*-test was applied to evaluate the differences in the SIC accumulation rate between the B4 and Ca(OH)<sub>2</sub> treatments. SPSS version 16.0 software (SPSS Inc., Chicago, IL,) was used for the statistical analyses and SigmaPlot version 12.5 (Systat Software Inc., San Jose, CA) was used to create the graphs.

# RESULTS Carbon Baseline Soil Organic C

Soil organic C at 10 cm at the native grass site was measured in February, May, and August and had an average value of 3.50 g kg<sup>-1</sup> with a SD of 0.98 (Table 2). The desert shrub site had an average value of 1.52 g kg<sup>-1</sup> with a SD of 0.29 and the turfgrass site had an average value of 2.82 g kg<sup>-1</sup> with a SD of 1.25. The SOC values at 40 cm were lower: 2.30, 1.46, 1.78 g kg<sup>-1</sup>, respectively (Table 2).

On the basis of the inference that the native grass site represents the SOC that would have existed at the desert shrub site in the mid-1800s (Buffington and Herbel, 1965; Gile, 1966; Gibbens et al., 2005), then a drop in organic C occurred when shrubs replaced the grassland, especially at the 10 cm depth (Fig. 3A,B). In contrast, converting the desert shrub site to turfgrass increased the amount of C in 6 yr, especially at the 10 cm depth.

Isotopically, SOC  $\delta^{13}$ C values confirm the change from C<sub>4</sub> grass to C<sub>3</sub> shrubs then back to C<sub>4</sub> grass. The  $\delta^{13}$ C values of SOC at the native grass site at 10 cm depth dropped from an average of -16.5‰ in the native grassland to -22.5‰ in the desert shrub site, which then rose to -17.3‰ in the turfgrass site (Fig. 3C). Using Eq. [2], this equates to a drop from 77% organic C from C<sub>4</sub> vegetation in the native grass to 22% C<sub>4</sub> at the desert shrub site, followed by a rise to 70% in the turfgrass site (Fig. 3C). A similar trend occurred at the 40 cm depth (Fig. 3D). The  $\delta^{13}$ C values at the turfgrass site demonstrate how rapidly SOC can convert from C<sub>3</sub> to C<sub>4</sub> signatures.

#### Soil Inorganic C

Soil inorganic C at 10 cm in the native grass site had an average value of 1.22 g kg<sup>-1</sup> with a SD of 0.04 (Table 2). The desert shrub site had an average value of 1.92 g kg<sup>-1</sup> with a SD of 1.69. The turfgrass site had an average value of 4.41 g kg<sup>-1</sup> with a SD of 1.30. In contrast to the SOC values, those of SIC were higher at 40 cm rather than lower (Table 2), which is typical of desert soils in the study region (Gile et al., 1981).

On the basis of the inference that the native grass site represents the amount of SIC that existed at the desert shrub site in the mid-1800s, an increase in SIC occurred when shrubs replaced the grassland (Fig. 4A,B). With regard to the conversion of desert shrubs to turfgrass, a notable increase in SIC at 10 cm accompanied this vegetative change in only 6 yr (Fig. 4A). This increase was greater at 10 cm than at 40 cm (Fig. 4B).

Isotopically, SIC  $\delta^{13}$ C values also reflect the change from  $C_3$  shrubs and  $C_4$  grass. The SIC content at the native grass site were too low to obtain accurate  $\delta^{13}$ C values, given the limitations of our continuous flow isotope ratio mass spectrometer. However,

at the desert shrub site, a value of -12.7 rose to -5.9% in the adjacent turfgrass site (Fig. 4C). On the basis of Eq. [3], this equates to an absence of  $C_4$  at the desert shrub site, followed by a rise to 43% in the turfgrass site in 6 yr (Fig. 4C). Little change in the isotope values occurred at the 40 cm depth (Fig. 4D). As for SOC, the  $\delta^{13}$ C values of SIC suggest a rapid change in isotopic signatures when  $C_4$  vegetation replaces  $C_3$  vegetation.

## **Treatment Effects Soil Organic C**

Did SOC change when the B4 and Ca(OH)<sub>2</sub> treatments were added to soil? Regardless of season or depth, the amount of SOC at the native grass, desert shrub, or turfgrass sites were little affected by either the B4 growth medium or Ca(OH)<sub>2</sub> (Fig. 5). This was expected, since SOC is primarily dependent on C in-

puts from plant growth and neither treatment was large enough or had enough time to affect plants.

Isotopically, SOC was also little affected by either the B4 or  $Ca(OH)_2$  treatment (Fig. 5). More variability occurred among sites than among treatments or seasons. For example, at 10 cm, the control samples at the desert shrub site averaged across February, May, and August were close to the pure  $C_3$  line of -25% (Fig. 5C). In contrast, the native grass and turfgrass sites at 10 cm depth were about midway between pure  $C_3$  and  $C_4$  (Fig. 5A,E). Similarly, at 40 cm depth, the desert shrub site had a  $\delta^{13}C$  value of -20.2%, which was closer to the pure  $C_3$  value (Fig. 5D) than the native grass or turfgrass sites, which were closer to pure  $C_4$  (Fig. 5B,F).

Table 2. Natural abundance of soil organic C (SOC) and soil inorganic C (SIC) in bulk samples and their isotopic compositions.

	SOC (g kg <sup>-1</sup> )		$\delta^{13}C_{SOC}$ (%)		SIC (g kg <sup>-1</sup> )		$\delta^{13}C_{SIC}$ (‰)		Soil temp.† (°C)		Soil CO <sub>2</sub> ‡ ( μL L <sup>-1</sup> )		
Month	10 cm	40 cm	10 cm	40 cm	10 cm	40 cm	10 cm	40 cm	10 cm	40 cm	10 cm	40 cm	Ppt.§ (mm)
Native gra	.SS												
Feb.	4.76	2.93	-17.5	-16.3	-	-	_	_	17.3	16.5	600	690	4.8
	4.00	3.80	-17.2	-17.6	_	_	_	_					
	4.52	2.69	-17.2	-16.6	-	-	_	_					
May	3.80	2.00	-15.9	-14.2	_	0.50	_	_	35.6	31.4	600	853	0.0
	2.97	2.16	-16.4	-16.0	-	0.98	_	_					
	$nm\P$	1.42	nm	-13.5	_	_	_	_					
Aug.	1.99	1.45	-15.8	-14.5	1.25	0.98	_	_	30.5	29.9	2,050	3,080	49.5
	2.38	nm	-16.8	nm	1.19	-	_	_					
	3.56	1.97	-15.5	-14.9	_	0.46	_	_					
Avg.	3.50	2.30	-16.54	-15.45	1.22	0.73	_	-					
SD	0.98	0.80	0.75	1.39	0.04	0.29	_	_					
Desert shr	ub												
Feb.	1.79	1.37	-23.4	-21.2	0.79	3.75	-12.7	-5.2	14.5	13.9	590	590	6.4
	1.71	1.77	-22.9	-21.8	1.17	4.52	-12.0	-5.1					
	1.54	1.45	-19.8	-21.1	5.24	9.04	-9.5	-3.9					
May	1.38	1.01	-23.1	-20.2	1.52	4.56	-16.7	-7.7	31.3	26.9	460	496	0.0
	1.43	2.14	-23.4	-20.1	0.91	3.69	-11.8	-2.3					
	0.98	1.21	-21.4	-19.5	4.47	12.10	-9.0	-6.7					
Aug.	1.93	1.54	-23.0	-19.0	1.40	8.07	-9.3	-6.5	31.0	29.7	1,670	1,945	50.5
	1.41	1.43	-23.3	-19.6	0.84	5.71	-11.7	-5.8					
	nm	1.25	nm	-19.1	0.92	4.46	-22.0	-7.4					
Avg.	1.52	1.46	-22.54	-20.18	1.92	6.21	-12.74	-5.62					
SD	0.29	0.33	1.28	0.99	1.69	2.90	4.19	1.73					
Turfgrass													
Feb.	2.23	1.68	-19.4	-18.6	3.24	5.54	-5.4	-7.9	11.4	11.2	2,075	2,900	86.4
	2.99	2.19	-16.5	-18.1	5.31	5.96	-6.5	-6.2					
	5.24	2.23	-16.1	-16.1	5.26	5.67	-5.5	-7.8					
May	1.37	1.84	-17.4	-17.7	7.13	6.20	-7.2	-6.3	20.1	19.5	7,533	10,486	245.6
	1.85	0.90	-15.9	-17.1	4.44	6.03	-8.8	-7.8					
	2.25	1.66	-18.0	-18.2	3.60	6.51	-5.8	-7.1					
Aug.	4.15	2.09	-17.4	-18.0	3.20	4.55	-2.6	-3.7	25.5	23.5	11,117	20,051	231.6
	1.93	1.77	-17.1	-17.3	3.36	4.40	-7.6	-4.9					
	3.36	1.65	-17.6	-17.0	4.11	6.22	-4.0	-6.0					
Avg.	2.82	1.78	-17.27	-17.57	4.41	5.68	-5.93	-6.41					
SD	1.25	0.40	1.07	0.77	1.30	0.74	1.88	1.43					

<sup>†</sup> Average of readings taken at 1-hr increments for the month-long duration of the experiment.

<sup>‡</sup> An average of three readings for desert shrub and turfgrass sites and two readings for native grass were taken on the day samples were harvested at the end of the month.

<sup>§</sup> Total precipitation for the duration of each monthly experiment.

<sup>¶</sup> nm, not measured, which indicates that this value was not measured because of instrument failure; –, not detected.

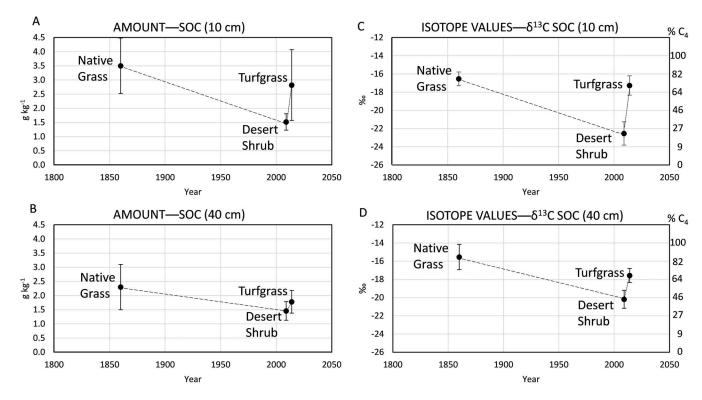


Fig. 3. Soil organic C (SOC) graphs plotting time versus SOC amounts at (A)10 and (B) 40 cm. The dashed line indicates the inference that the desert shrub site was formerly a native grassland in the mid-1800s (Buffington and Herbel, 1965; Gile, 1966). Soil organic C graphs plotting  $\delta^{13}$ C values at (C) 10 and (D) 40 cm and percentage of C from C<sub>4</sub> vegetation.

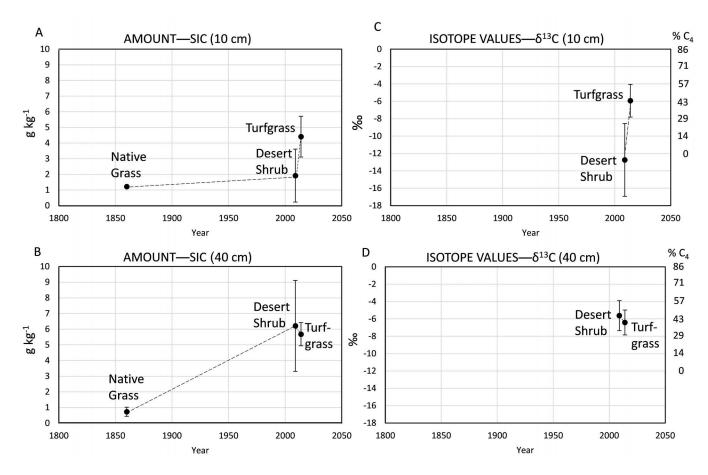


Fig. 4. Soil inorganic C (SIC) graphs plotting time versus SIC amounts at (A) 10 and (B) 40 cm. The dashed line indicates the inference that the desert shrub site was formerly a native grassland in the mid-1800s (Buffington and Herbel, 1965; Gile, 1966). Soil inorganic C graphs plotting  $\delta^{13}$ C values at (C) 10 and (D) 40 cm and percentage of C from C<sub>4</sub> vegetation

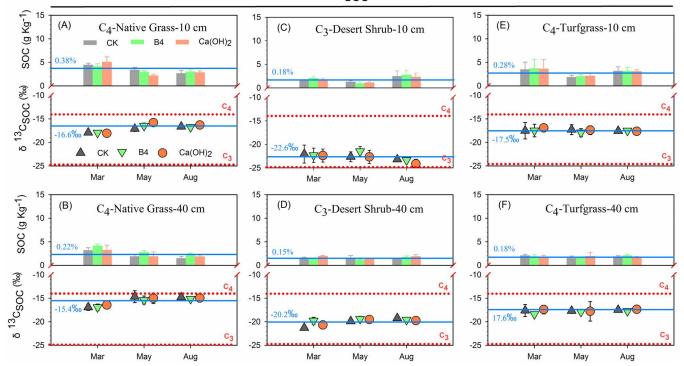


Fig. 5. Soil organic C (SOC) amounts and  $\delta^{13}$ C values in soil samples at (A,C,E) 10 and (B,D,F) 40 cm for the three study sites. (A,B) Native grass; (C,D) desert shrub; (E,F) turfgrass. The blue line shows the average C and  $\delta^{13}$ C values in control samples across February, May, and August. Red dashed lines locate the position of pure C<sub>3</sub> and C<sub>4</sub> vegetation.

#### Soil Inorganic C

Did SIC change when the B4 and  $Ca(OH)_2$  treatments were added to soil? The SIC response to the treatments was more variable than the SOC response but was still minor (Fig.

6). The treatment response was not apparent at either 10 or 40 cm (Fig. 6A,B). For the other two sites containing SIC, the largest response was at 10 cm depth in May at the desert shrub site (Fig. 6C). The Ca(OH)<sub>2</sub> treatment at 10 cm depth also had its

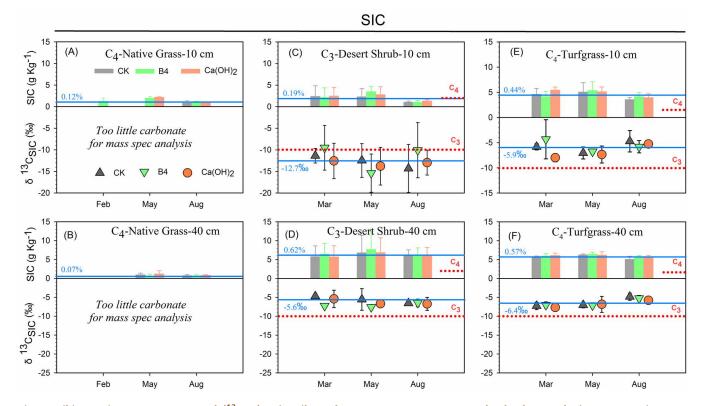


Fig. 6. Soil inorganic C (SIC) amounts and d<sup>13</sup>C values in soil samples at (A,C,E) 10 (B,D,F) 40 cm for the three study sites. (A,B) Native grass; (C,D) desert shrub; (E,F) turfgrass. The blue line shows the average C and  $\delta^{13}$ C values in control samples across February, May, and August. Red dashed lines locate the position of pure C<sub>3</sub> and C<sub>4</sub> vegetation. mass spec, mass spectrometry.

greatest response at the desert shrub site in May and at the turfgrass site in Feb (Fig. 6C,E). Still, there were no statistically significant differences at the p < 0.05 level among the control, B4, or Ca(OH)<sub>2</sub> treatments at either 10 or 40 cm.

Isotopically, SIC responses were more variable to the B4 and  $Ca(OH)_2$  treatments than their amount of SIC, especially at the desert shrub site at 10 cm (Fig. 6C). The B4 treatment at that site was higher than the control for February and August but lower for May (Fig. 6C). The  $Ca(OH)_2$  treatment, on the other hand, was lower for February and May but higher in August. Still, all sites lacked statistically significant differences at the p < 0.05 level.

#### **Microscopy**

Microscopy was used as additional evidence to determine whether SIC formed after the B4 medium was added to the soil. As shown in Fig. 2, one of the two B4 treatments at both 10 and 40 cm was examined with light and electron microscopy for calcified microorganisms in the form of calcified roots, fungal hyphae, or bacteria. Fungal biomineralization, for example, would be expected to appear like the image in Fig. 7A, which is common in the desert soils of the study area (Monger et al., 1991). The results, however, were negative. The closest example of biomineralization that could be found after examining all samples is shown in Fig. 7B. However, even this specimen displayed only weak, if any, evidence of calcification.

#### **Soil Inorganic C Accumulation Rates**

In addition to our examination of the total amounts of SIC produced by the addition of B4 and Ca(OH)<sub>2</sub>, we also re-examined the SIC data in the form of accumulation rates using Eq. [4], which magnifies the differences. In general, the accumulation under B4 was slightly greater than under Ca(OH)<sub>2</sub> but the variability was high and no statistically significant differences were found. Both B4 and Ca(OH)<sub>2</sub> typically had higher accumulation rates than the controls (Fig. 8A–F), except for a few cases when it was lower, which indicates dissolution in comparison to the control samples.

## **Correlations of SIC in Treated Samples to Physical Conditions**

Soil Inorganic C formation and dissolution is controlled by  ${\rm Ca^{2+}}$  and  ${\rm CO_3}^{2-}$  activity. These variables operate within the context of soil temperature, precipitation, and  ${\rm CO_2}$  concentrations (Breecker et al., 2009), which exert important controls over SIC formation in dryland soils in the study region. Soil inorganic C generated by B4 and  ${\rm Ca(OH)_2}$  treatments, therefore, was examined in light of these variables.

Soil temperatures at 10 cm during February, May, and August were similar between the native grass and desert shrub sites but, owing to the effect of irrigation, were 5 to 10°C cooler for the turfgrass site (Table 2; Fig. 8G–I). Temperatures at 40 cm followed a similar trend to those at 10 cm at all sites but were slightly cooler.

Precipitation, like temperature, was similar for the native grass and desert shrub sites but much greater for the turfgrass site because of the irrigation. The native grass site received 54 mm and the desert shrub site received 56 mm, in contrast to the turfgrass site, which received 1200 mm (Table 2; Fig. 8I, K, and G). In essence, irrigation changed the turfgrass site from a soil with an aridic moisture regime to a soil with an udic moisture regime.

Carbon dioxide concentrations closely followed soil moisture concentrations, as revealed by comparing Fig. 8G, 8H, and 8I with Fig. 8J, 8K, and 8L. At 10 cm depth, CO $_2$  ranged from 600 to 2050  $\mu L$  L $^{-1}$  at the native grass site, from 590 to 1670  $\mu L$  L $^{-1}$  at the desert shrub site, and from 2075 to 11,117  $\mu L$  L $^{-1}$  at the turfgrass site. At 40 cm depth, the increase was even more noticeable: CO $_2$  ranged from 690 to 3080  $\mu L$  L $^{-1}$  at the native grass site, from 590 to 1945  $\mu L$  L $^{-1}$  at the desert shrub site, and from 2900 to 20,051  $\mu L$  L $^{-1}$  at the turfgrass site.

To put seasonal CO2 variability in the context of diurnal variability, CO<sub>2</sub> was measured during a 24-hr period at the desert shrub and turfgrass sites on two dates: first in May (the hot, dry season) and again in August (the hot, moist monsoon season). These measurements revealed that diurnal CO2 variability was small compared to seasonal increases. At the desert shrub site in May, diurnal values ranged from approximately 350  $\mu L L^{-1}$ at 10 cm depth to 650  $\mu L$   $L^{-1}$  at 40 cm depth, in contrast to the August values, which ranged from  $1100 \,\mu L \, L^{-1}$  at  $10 \, cm$  to 1800  $\mu$ L L<sup>-1</sup> at 40 cm (Fig. 9A). At the turfgrass site, CO<sub>2</sub> was much higher and the concentration at 10 cm remained below the concentration at 40 cm, unlike the desert shrub site. In May, diurnal values at the turfgrass site ranged from 7500  $\mu L~L^{-1}$  at 10 cm depth to 17,000  $\mu$ L L<sup>-1</sup> at 40 cm, in contrast to the August values, which ranged from 11,000  $\mu$ L L<sup>-1</sup> at 10 cm and 21,000  $\mu L L^{-1}$  at 40 cm (Fig. 9B).

Although it is well known that temperature, rainfall, and soil  $CO_2$  are important factors for carbonate formation, an attempt to find statistically significant correlations among these variables individually was unsuccessful (p < 0.05). However, taken together, these factors were statistically significant (Table 3).

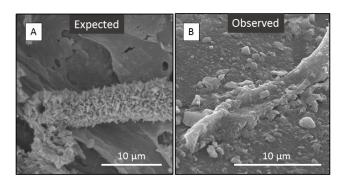


Fig. 7. Scanning electron micrographs. (A) Calcified fungal hyphae common in the native soils of the study region. If biomineralization in the soil treated with liquid growth medium had occurred, this is how we would have expected it to appear. (B) Fungal hyphae from soil treated with the liquid growth medium. Few, if any, calcite crystals are apparent on the hyphae, indicating that biomineralization did not occur as expected.

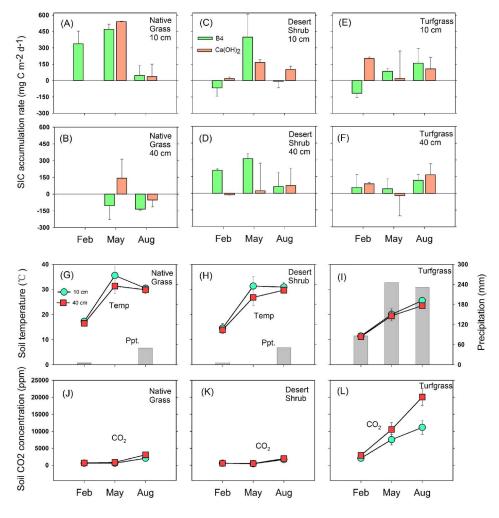


Fig. 8. Comparison of soil inorganic C (SIC) accumulation rates for B4 and  $Ca(OH)_2$  treatments at (A,C,E) 10 and (B,D,F) 40 cm for the 3 mo of the experiment. (G,H,I) Soil temperature, precipitation and (J,K,L) soil  $CO_2$  concentrations during the experiments. (G,J) Native turfgrass; (H,K) desert shrub; (I,L) turfgrass.

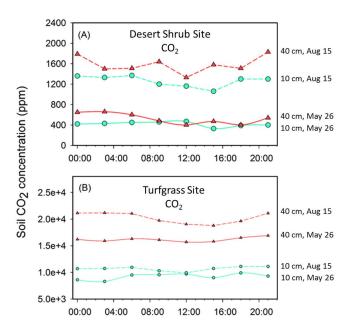


Fig. 9. Diurnal variations of soil  $\mathrm{CO}_2$  concentrations at the desert shrub (DS) and turfgrass (TG) sites before the monsoon season (measured on 26 May) and during the monsoon seasons (measured on 15 August).

The statistical analysis was repeated using a stepwise procedure for each factor. Still, there was no single factor effect. Thus, SIC in this study was regulated not by single parameters but by multiple factors.

#### **DISCUSSION**

How valid is the conclusion that a decline in SOC accompanied the conversion of native grasslands to desert shrub, as indicated by Fig. 3A? First, we must emphasize that we substituted space for time by making the inference that the vestigial grassland at Site 1 represents how the desert shrub soil at Site 2 would have appeared 150 yr ago. Still, such a loss of SOC would be expected based on the typical amount of C in a grassland soil versus the typical amount in a desert shrub soil. The SOC in the remaining grasslands of the study region is about 1% compared to less than 0.5% in local shrublands soils (Gile et al., 1981). However, it must be pointed out that SOC in a grassland is more homogeneously distributed at the landscape scale than SOC in a shrubland. Soil directly beneath some shrubs can actually have more SOC than grassland soils (Throop and Archer, 2008). It has also been pointed out that al-

though losses of SOC have occurred with the grass-shrub conversion, C storage of the ecosystem as a whole has changed little, especially in light of the extensive rooting system of the woody shrubs (Schlesinger and Pilmanis, 1998).

More striking was the increase in SOC resulting from the desert site being converted to turfgrass. In this case, we are making the inference that the turfgrass soil had the same C as the neighboring desert shrub soil before 2008. Given (i) the close proximity of the adjacent sites (100 m), (ii) the detailed soil mapping in the study area, and (iii) the uniform nature of the Torripsamment parent material, this seems to be a reasonable inference. In fact, such increases in SOC have been documented for almost 50 yr in the study area (Ruhe, 1967). At the time of Ruhe's study, some areas of New Mexico State University had been in turfgrass since the 1890s. Soil organic C in these irrigated lawns were found to be 3.7 to 12.5 times greater than in the neighboring desert shrub soils.

Carbon isotopes have been used to estimate turnover rates of organic matter for at least three decades (e.g., Balesdent et al., 1987; Boutton, 1991). In this study, the  $\delta^{13}$ C values of SOC at 10 cm depth indicate a turnover from 77% C<sub>4</sub> in the native

grass site's soil to 22%  $\rm C_4$  in the desert shrub site then to 70%  $\rm C_4$  in the turfgrass site. Just within a period of 6 yr since the desert shrub site was converted to turfgrass, the  $\rm \delta^{13}C$  values changed from -22.5 to -17.3‰, demonstrating how rapidly SOC can lose a  $\rm C_3$  signature and obtain a  $\rm C_4$  signature in this setting.

Soil inorganic C, in contrast to SOC, is typically viewed as a long-term soil component that forms according to the reaction  $Ca^{2+} + 2HCO_3^- \rightarrow CaCO_3 + CO_2 + H_2O$ .

In the study area, there is much soil-geomorphic evidence to support the view that SIC is a long-term mineral (Hawley et al., 1976). Radiocarbon dates that go back as far as 30,630 yr before the present also support this view (Monger et al., 1998). However, there also appears to be a superimposed dynamic carbonate component in native soils. In managed soils, like at the turfgrass site, this study suggests the possibility that SIC can be purposefully managed.

Soil inorganic C  $\delta^{13}$ C values are more commonly used to investigate paleoclimates than turnover rates (e.g., Cerling, 1984; Rabenhorst et al., 1984; Landi et al., 2003; Stevenson et al., 2005). Soil inorganic C values in this study, however, suggest that carbonate turnover rates might be surprisingly fast when C<sub>3</sub> shrubs replace C<sub>4</sub> grass or vice versa. Hence,  $\delta^{13}$ C values can be useful for investigating the neoformation of carbonate in managed systems (e.g., Li et al., 2007; Mi et al., 2008; Wang et al., 2014a).

The result that neither the B4 nor Ca(OH) $_2$  treatment had a significant impact on the amount or  $\delta^{13}$ C values of SOC was not surprising because neither treatment had enough time to affect plant growth or microbial decomposition of the existing organic matter. The treatments did, however, have a slightly greater impact on SIC, as shown by their greater variability. Both treatments caused slightly greater amounts of SIC to form than in the controls but the differences were not statistically significant.

Within the slight increase stimulated by the treatments, B4 produced more SIC than Ca(OH)<sub>2</sub>. The increased SIC resulting from the Ca(OH)<sub>2</sub> treatments is in agreement with many other researchers who have successfully modeled the precipitation and dissolution of Ca(OH)<sub>2</sub> by abiotic pathways (e.g., Hirmas et al., 2010). The increased SIC from the B4 treatments was expected to result from biomineralization via metabolic processes that mediate pH, CO<sub>2</sub>, and Ca (Phillips et al., 1987; Frankel and Bazylinski, 2003). However, no microbial calcification was found via microscopy.

Measurements of soil temperature, moisture, and  $\mathrm{CO}_2$  in similar soils in New Mexico by Breecker et al. (2009) led those authors to conclude that pedogenic carbonate is formed in the warm, dry conditions during May when  $\mathrm{CO}_2$  concentrations are low. Consequently, the  $\delta^{13}\mathrm{C}$  values of pedogenic carbonates would not record the mean growing season vegetation, as typically assumed, but would instead record disproportionately more  $\mathrm{C}_4$  vegetation. Other studies have also shown a seasonal effect on the timing of carbonate formation, especially as a function of

Table 3. ANOVA for soil inorganic C accumulation rate as a function of temperature (T), soil CO<sub>2</sub> concentration (SCO<sub>2</sub>), and precipitation (P).

Source of variation	df	Sum of squares	Mean of squares	F-value	<i>P</i> -value
T	1	65,896	65,896	3.78	0.063
SCO <sub>2</sub>	1	2,713	2,713	0.16	0.696
Р	1	23,151	23,151	1.33	0.260
$T \times SCO_2$	1	37,997	37,997	2.18	0.152
$T \times P$	1	6,326	6,326	0.36	0.552
$SCO_2 \times P$	1	150,803	150,803	8.66	0.007
$T \times SCO_2 \times P$	1	102,334	102,334	5.87	0.023

depth and when soil drying occurs (Ringham et al., 2016) and when respiration rates are higher (Oerter and Amundson, 2016). Our results do not support these conclusion, given the insignificant differences in SIC formation we observed among seasons. However, neither does our study dispute their conclusions because of the short duration of our experiment.

The observed average  $\delta^{13}$ C values of pedogenic carbonates at 10 cm at the desert shrub site was -12.7%, which is lower than the values below -11% commonly observed for carbonates in the study area (Deutz et al., 2002; Liu et al., 2007). Why are the values at this one location so low? Given a theoretical enrichment from diffusional and temperature-driven fractionation at 25°C (Cerling and Quade, 1993), we would expect values of around -10%. However, previous studies of carbonate formed on the roots of creosotebush have shown values as low as -18.5% (Monger et al., 2009). The samples at 10 cm at Site 2 (the desert shrub site) were collected in soil dominated by fine creosotebush roots, which might explain the low values, although further investigation is needed. We do not, however, attribute the low values to analytical error because (i) the other SIC  $\delta^{13}$ C values are reasonable and (ii) the analytic method used for obtaining these values was tested and verified with standards.

What role did Ca in irrigation water play in the observed increase in SIC at the turfgrass site? The concentration of Ca in the irrigation water was 2.6 mmol L $^{-1}$  and 1200 mm of irrigation was added in 2014. If we assume the same concentration and rate of irrigation for the previous 6 yr, then 749 g of Ca would have been added per m $^2$ . If all of this Ca reacted to form CaCO $_3$  in the upper 10 cm, then 1872 g of CaCO $_3$  would have been added to the top 10 cm of each m $^2$  of soil. Given a bulk density of 1.4 g cm $^{-3}$ , this amount could result in an increase of 1.3% CaCO $_3$ . Of course, some Ca would have leached and some would have been taken up by plants. Still, on a theoretical basis, Ca in irrigation water could account for the observed SIC.

Can irrigated agriculture sequester atmospheric CO<sub>2</sub> in the broad context of upstream CO<sub>2</sub> emissions? Any pumping of irrigation water that involves fossil fuel would need to be considered. Any processing and transportation of Ca fertilizer that involved fossil fuel would also need to be considered. Any Ca from preexisting CaCO<sub>3</sub> would need to be considered because it negates sequestration. Given these limitations, if irrigation is gravity-fed and if Ca in the irrigation water comes from the

chemical weathering of silicates, then some of the CO<sub>2</sub> from root and microbial respiration could be sequestered as CaCO<sub>3</sub>.

#### **CONCLUSIONS**

The main result of this study reveals that converting desert shrubs to turfgrass can increase SIC in less than 10 yr, which is unexpected, given the common view that SIC is a mineral that takes centuries to millennia to accumulate in soil. This experiment, therefore, supports other studies (e.g., Wang et al., 2014b), suggesting that SIC, like SOC, might be managed to sequester atmospheric CO<sub>2</sub> in desert environments. However, before these results can be generalized to other soils, it needs to be demonstrated that the source of Ca is derived directly from silicate minerals rather than preexisting carbonate, that the carbonate is durable and not subject to rapid dissolution, and that upstream CO<sub>2</sub> emissions are accounted for (Schlesinger, 1999).

The test for microbial biomineralization was negative. Although the liquid growth medium produced slightly more SIC than Ca(OH)<sub>2</sub>, differences were neither statistically different between treatments nor between treatments and controls. In addition, no microbial calcification was found by using electron microscopy. Further studies are needed that use more liquid growth medium and allow it to react for longer periods than 1 mo.

The C isotope results of this experimental pedology study have implications for paleoclimatic studies by increasing our understanding of "soil memory" (Targulian and Goryachkin, 2004; Monger and Rachal, 2013). First, this study shows that  $\delta^{13}$ C values can change in less than a decade for both SOC and SIC. Thus soils, like those in this study, can be overprinted and described as having "short memories." Second, comparing the relative changes in  $\delta^{13}$ C values between the 10-cm samples and the 40-cm samples supports the observation that subsoil B horizons have "longer memories" than surface A horizons (Gerasimova and Lebedeva, 2008). Thus an experimental pedology approach can supplement traditional paleopedology studies that must rely primarily on observation, deduction, and induction.

#### **ACKNOWLEDGMENTS**

The authors are grateful for the financial support from the National Natural Science Foundation of China (grant No. 41403072) and the Chinese Scholarship Council. Funding was also provided by the National Science Foundation to the Jornada Basin Long Term Ecological Research Program (DEB 125828) and the New Mexico State University Agricultural Experiment Station.

#### REFERENCES

- Balesdent, J., A. Mariotti, and B. Guillet. 1987. Natural <sup>13</sup>C abundance as a tracer for studies of soil organic matter dynamics. Soil Biol. Biochem. 19:25–30. doi:10.1016/0038-0717(87)90120-9
- Boquet, E., A. Boronat, and A. Ramos-Cormenzana. 1973. Production of calcite (calcium carbonate) crystals by soil bacteria is a general phenomenon. Nature 246:527–529. doi:10.1038/246527a0
- Boutton, T.W. 1991. Stable carbon isotope ratios of natural materials: II. Atmospheric, terrestrial, marine, and freshwater environments. In: C. Coleman and B. Fry, editors, Carbon isotope techniques. Academic Press, New York. p. 173–185. <a href="doi:10.1016/B978-0-12-179730-0.50016-3">doi:10.1016/B978-0-12-179730-0.50016-3</a>
- Boutton, T.W., S.R. Archer, and A.J. Midwood. 1999. Stable isotopes in ecosystem science: structure, function and dynamics of a subtropical

- savanna. Rapid Commun. Mass Spectrom. 13:1263-1277.
- Breecker, D.O., Z.D. Sharp, and L.D. McFadden. 2009. Seasonal bias in the formation and stable isotopic composition of pedogenic carbonate in modern soils from central New Mexico, USA. Geol. Soc. Am. Bull. 121:630–640. doi:10.1130/B26413.1
- Buffington, L.C., and C.H. Herbel. 1965. Vegetational changes on a semidesert grassland range from 1858 to 1963. Ecol. Monogr. 35:139–164. doi:10.2307/1948415
- Bughio, M.A., P. Wang, F. Meng, C. Qing, Y. Kuzyakov, X. Wang, et al. 2016. Neoformation of pedogenic carbonates by irrigation and fertilization and their contribution to carbon sequestration in soil. Geoderma 262:12–19. doi:10.1016/j.geoderma.2015.08.003
- Buyanovsky, G.A., and G.H. Wagner. 1983. Annual cycles of carbon dioxide level in soil air. Soil Sci. Soc. Am. J. 47:1139–1145. <a href="https://doi.org/10.2136/sssaj1983.03615995004700060016x">doi:10.2136/sssaj1983.03615995004700060016x</a>
- Cerling, T.E. 1984. The stable isotopic composition of modern soil carbonate and its relationship to climate. Earth Planet. Sci. Lett. 71:229–240. doi:10.1016/0012-821X(84)90089-X
- Cerling, T.E., and J. Quade. 1993. Stable carbon and oxygen isotopes in soil carbonates. In: P.K. Swart, K.C. Lohmann, J. McKenzie, and S. Savin, editors, Climate change in continental isotopic records. American Geophysical Union Monograph Ser. 78. American Geophysical Union, Washington, D.C. p. 217–231.
- DeJong, J.T., K. Soga, S.A. Banwart, W.R. Whalley, T.R. Ginn, D.C. Nelson, et al. 2010. Soil engineering in vivo: Harnessing natural biogeochemical systems for sustainable, multi-functional engineering solutions. J. R. Soc. Interface 8:1–15. doi:10.1098/rsif.2010.0270
- De Muynck, W., N. De Belie, and W. Verstraete. 2010. Microbial carbonate precipitation in construction materials: A review. Ecol. Eng. 36:118–136. doi:10.1016/j.ecoleng.2009.02.006
- Deutz, P., I.P. Montañez, and H.C. Monger. 2002. Morphology and stable and radiogenic isotope composition of pedogenic carbonate in late Quaternary relict soils, New Mexico: An integrated record of pedogenic overprinting. J. Sediment. Res. 72:809–822. doi:10.1306/040102720809
- Drees, L.R., and L.P. Wilding. 1987. Micromorphic record and interpretations of carbonate forms in the Rolling Plains of Texas. Geoderma 40:157–175. doi:10.1016/0016-7061(87)90020-6
- Eshel, G., P. Fine, and M.J. Singer. 2007. Total soil carbon and water quality: An implication for carbon sequestration. Soil Sci. Soc. Am. J. 71:397–405. doi:10.2136/sssaj2006.0061
- Eswaran, H., P.F. Reich, J.M. Kimble, F.H. Beinroth, E. Padmanabhan, and P. Moncharoen. 2000. Global carbon stocks. In: R. Lal, J.M. Kimble, H. Eswaran, B.A. Stewart, editors, Global climate change and pedogenic carbonates. Lewis Publishers, Boca Raton, FL. p. 15–27,
- Frankel, R.B., and D.A. Bazylinski. 2003. Biologically induced mineralization by bacteria. Rev. Mineral. Geochem. 54:95–114. doi:10.2113/0540095
- Gallegos, R.A. 1999. Biogenic carbonate, desert vegetation, and stable carbon isotopes. Masters thesis. New Mexico State Univ., Las Cruces, NM.
- Gerasimova, M., and M. Lebedeva. 2008. Contribution of micromorphology to classification of aridic soils. In: S. Kapur, et al., editors, New trends in soil micromorphology. Springer, Heidelberg. p. 151–162. doi:10.1007/978-3-540-79134-8
- Gibbens, R.P., R.P. McNeely, K.M. Havstad, R.F. Beck, and B. Nolen. 2005. Vegetation changes in the Jornada Basin from 1858 to 1998. J. Arid Environ. 61:651–668. doi:10.1016/j.jaridenv.2004.10.001
- Gile, L.H. 1966. Coppice dunes and the Rotura soil. Soil Sci. Soc. Am. J. 30:657–660.
  Gile, L.H., J.W. Hawley, and R.B. Grossman. 1981. Soils and geomorphology in the Basin and Range area of southern New Mexico: Guidebook to the Desert Project. New Mexico Bureau of Mines & Mineral Resources, Socorro, NM.
- Guo, Y., R. Amundson, P. Gong, and Q. Yu. 2006. Quantity and spatial variability of soil carbon in the conterminous United States. Soil Sci. Soc. Am. J. 70:590–600. doi:10.2136/sssaj2005.0162
- Hallsworth, E.G., and D.V. Crawford. 1965. Experimental pedology. Butterworths, London.
- Hawley, J.W., G.O. Bachman, and K. Manley. 1976. Quaternary stratigraphy in the Basin and Range and Great Plains provinces, New Mexico and western Texas. In: W.C. Mahaney, editor, Quaternary stratigraphy of North America. Dowden, Hutchinson & Ross, Inc., Stroudsburg, PA. p. 235–274.
- Hirmas, D.R., C. Amrhein, and R.C. Graham. 2010. Spatial and process-based

- modeling of soil inorganic carbon storage in an arid piedmont. Geoderma 154:486–494. doi:10.1016/j.geoderma.2009.05.005
- Khormali, F., H.C. Monger, and Y. Feng. 2014. Experimental micropedology: A technique for investigating soil carbonate biogenesis along a desert–grassland– forest transect, New Mexico, USA. Spanish J. Soil Sci. 4:1–18.
- Klappa, C.F. 1979. Calcified filaments in Quaternary calcretes: Organo-mineral interactions in the subaerial vadose environment. J. Sediment. Res. 49:955–968. doi:10.2110/jsr.49.955
- Koch, A., A. McBrantney, and B. Minasny. 2015. 4 per 1000 soil carbon to mitigate climate change. Global Policy. http://www.globalpolicyjournal. com/blog/24/11/2015/4-1000-soil-carbon-mitigate-climate-change (accessed 23 Sept. 2016)
- Lal, R. 2004. Soil carbon sequestration impacts on global climate change and food security. Science 304:1623–1627. doi:10.1126/science.1097396
- Lal, R. 2008. Carbon sequestration. Philos. Trans. R. Soc. Lond. B Biol. Sci. 363:815–830. doi:10.1098/rstb.2007.2185
- Landi, A., A.R. Mermut, and D.W. Anderson. 2003. Origin and rate of pedogenic carbonate accumulation in Saskatchewan soils, Canada. Geoderma 117:143–156. doi:10.1016/S0016-7061(03)00161-7
- Li, Z.P., F.X. Han, Y. Su, T.L. Zhang, B. Sun, D.L. Monts, et al. 2007. Assessment of soil organic and carbonate carbon storage in China. Geoderma 138:119–126. doi:10.1016/j.geoderma.2006.11.007
- Liu, X., H.C. Monger, and W.G. Whitford. 2007. Calcium carbonate in termite galleries—biomineralization or upward transport? Biogeochemistry 82:241–250. doi:10.1007/s10533-006-9067-x
- Mi, N.A., S. Wang, J. Liu, G. Yu, W. Zhang, and E. Jobbágy. 2008. Soil inorganic carbon storage pattern in China. Glob. Change Biol. 14:2380–2387. doi:10.1111/j.1365-2486.2008.01642.x
- Midwood, A.J., and T.W. Boutton. 1998. Soil carbonate decomposition by acid has little effect on  $\delta^{13}C$  of organic matter. Soil Biol. Biochem. 30:1301–1307. doi:10.1016/S0038-0717(98)00030-3
- Mikhailova, E.A., and C.J. Post. 2006. Effects of land use on soil inorganic carbon stocks in the Russian chernozem. J. Environ. Qual. 35:1384–1388. doi:10.2134/jeq2005.0151
- Monger, H.C. 2014. Soils as generators and sinks of inorganic carbon in geologic time. In: A.E. Hartemink and K. McSweeney, editors, Soil carbon. Progress in soil science. Springer International Publishing, New York. p. 27–36. doi:10.1007/978-3-319-04084-4\_3
- Monger, H.C., and D.M. Rachal. 2013. Soil and landscape memory of climate change: How sensitive, how connected? In: S.G. Driese and L.C. Nordt, editors, New frontiers in paleopedology and terrestrial paleoclimatology: Paleosols and soil surface analog systems. Society for Sedimentary Geology Special Publication 104. Society for Sedimentary Geology, Tulsa, OK. pp. 63–70.
- Monger, H.C., L.A. Daugherty, W.C. Lindemann, and C.M. Liddell. 1991. Microbial precipitation of pedogenic calcite. Geology 19:997–1000. doi:10.1130/0091-7613(1991)019<0997:MPOPC>2.3.CO;2
- Monger, H.C., D.R. Cole, J.W. Gish, and T.H. Giordano. 1998. Stable carbon and oxygen isotopes in Quaternary soil carbonates as indicators of ecogeomorphic changes in the northern Chihuahuan Desert, USA. Geoderma 82:137–172. doi:10.1016/S0016-7061(97)00100-6
- Monger, H.C., D.R. Cole, B.J. Buck, and R.A. Gallegos. 2009. Scale and the isotopic record of  $\mathrm{C}_4$  plants in pedogenic carbonate: From the biome to the rhizosphere. Ecology 90:1498–1511.  $\underline{\text{doi:}10.1890/08-0670.1}$
- Monger, H.C., R.A. Kraimer, S.A. Khresat, D.R. Cole, X. Wang, and J. Wang. 2015. Sequestration of inorganic carbon in soil and groundwater. Geology 43:375–378. doi:10.1130/G36449.1
- Oerter, E.J., and R. Amundson. 2016. Climate controls on spatial and temporal variations in the formation of pedogenic carbonate in the western Great Basin of North America. Geol. Soc. Am. Bull. 128(7–8):1095–1104 doi:10.1130/B31367.1
- Peters, D.P.C., J. Yao, O.E. Sala, and J.P. Anderson. 2012. Directional climate change and potential reversal of desertification in arid and semiarid ecosystems. Glob. Change Biol. 18:151–163. doi:10.1111/j.1365-2486.2011.02498.x
- Phillips, S., A. Milnes, and R. Foster. 1987. Calcified filaments- an example of biological influences in the formation of calcrete in South Australia. Aust.

- J. Soil Res. 25:405–428. doi:10.1071/SR9870405
- Post, W.M., W.R. Emanuel, P.J. Zinke, and A.G. Stangenberger. 1982. Soil carbon pools and world life zones. Nature 298:156–159. doi:10.1038/298156a0
- Rabenhorst, M.C., L.P. Wilding, and L.T. West. 1984. Identification of pedogenic carbonates using stable carbon isotope and microfabric analyses. Soil Sci. Soc. Am. J. 48:125–132. doi:10.2136/sssaj1984.03615995004800010023x
- Ringham, M.C., G.D. Hoke, K.W. Huntington, and J.N. Aranibar. 2016. Influence of vegetation type and site-to-site variability on soil carbonate clumped isotope records, Andean piedmont of Central Argentina (32– 34°S). Earth Planet. Sci. Lett. 440:1–11. doi:10.1016/j.epsl.2016.02.003
- Rodriguez-Navarro, C., M. Rodriguez-Gallego, K. Ben Chekroun, and M.T. Gonzalez-Muñoz. 2003. Conservation of ornamental stone by myxococcus xanthus-induced carbonate biomineralization. Appl. Environ. Microbiol. 69:2182–2193. doi:10.1128/AEM.69.4.2182-2193.2003
- Ruhe, R.V. 1967. Geomorphic surfaces and surficial deposits in southern New Mexico. Memoir 18. New Mexico Bureau of Mines and Mineral Resources, Socorro, NM.
- Schlesinger, W.H. 1982. Carbon storage in the caliche of arid soils: A case study from Arizona. Soil Sci. 133:247–255. doi:10.1097/00010694-198204000-00008
- Schlesinger, W.H. 1999. Carbon sequestration in soils. Science 284:2095. doi:10.1126/science.284.5423.2095
- Schlesinger, W.H., and E.S. Bernhardt. 2013. Biogeochemistry: An analysis of global change. Academic Press, Oxford, UK.
- Schlesinger, W.H., and A.M. Pilmanis. 1998. Plant-soil interactions in deserts. Biogeochemistry 42:169–187. doi:10.1023/A:1005939924434
- Schubert, B.A., and A.H. Jahren. 2012. The effect of atmospheric CO<sub>2</sub> concentration on carbon isotope fractionation in C<sub>3</sub> land plants. Geochim. Cosmochim. Acta 96:29–43. doi:10.1016/j.gca.2012.08.003
- Serena, M., B. Leinauer, M. Schiavon, B. Maier, and R. Sallenave. 2014. Establishment and rooting response of bermudagrass propagated with saline water and subsurface irrigation. Crop Sci. 54:827–836. doi:10.2135/ cropsci2013.07.0512
- Snyder, R.L., and S. Eching. 2007. PMDay.xls spreadsheet software for estimating daily or hourly reference evapotranspiration using the Penman-Monteith equation. University of California, Davis. http://biomet.ucdavis.edu/
- Stevenson, B.A., E.F. Kelly, E.V. McDonald, and A.J. Busacca. 2005. The stable carbon isotope composition of soil organic carbon and pedogenic carbonates along a bioclimatic gradient in the Palouse region, Washington State, USA. Geoderma 124:37–47. doi:10.1016/j.geoderma.2004.03.006
- Targulian, V.O., and S.V. Goryachkin. 2004. Soil memory: Types of record, carriers, hierarchy and diversity. Rev. Mex. Cienc. Geol. 21:1–8.
- Throop, H.L., and S.R. Archer. 2008. Shrub (*Prosopis velutina*) encroachment in a semidesert grassland: Spatial–temporal changes in soil organic carbon and nitrogen pools. Glob. Change Biol. 14:2420–2431. <a href="https://doi.org/10.1111/j.1365-2486.2008.01650.x">doi:10.1111/j.1365-2486.2008.01650.x</a>
- Verrecchia, E.P., and K.E. Verrecchia. 1994. Needle-fiber calcite: A critical review and a proposed classification. J. Sediment. Res. 64:650–664.
- Wang, J.P., X.J. Wang, J. Zhang, and C.Y. Zhao. 2014a. Soil organic and inorganic carbon and stable carbon isotopes in the Yanqi Basin of northwestern China. Eur. J. Soil Sci. 66:95–103. doi:10.1111/ejss.12188
- Wang, X.J., M.G. Xu, J.P. Wang, W.J. Zhang, X.Y. Yang, S.M. Huang, et al. 2014b. Fertilization enhancing carbon sequestration as carbonate in arid cropland: Assessments of long-term experiments in northern China. Plant Soil 380:89–100. doi:10.1007/s11104-014-2077-x
- Weems, S.L., and H.C. Monger. 2012. Banded vegetation-dune formation in the Medieval Warm Period and 20th century, Chihuahuan Desert, New Mexico, USA. Ecosphere 3(3):1–16. doi:10.1890/ES11-00194.1
- Wu, H., Z. Guo, Q. Gao, and C. Peng. 2009. Distribution of soil inorganic carbon storage and its changes due to agricultural land use activity in China. Agric. Ecosyst. Environ. 129:413–421. doi:10.1016/j.agee.2008.10.020
- Wu, L., Y. Wood, P. Jiang, L. Li, G. Pan, J. Lu, et al. 2008. Carbon sequestration and dynamics of two irrigated agricultural soils in California. Soil Sci. Soc. Am. J. 72:808–814. doi:10.2136/sssaj2007.0074