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Landscape patterns of vegetation change indicated by soil carbon isotope composition

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Abstract

Vegetation change, particularly from the grass to shrub life form, is a critical issue on the world's semiarid rangelands. Stable carbon isotope ($\delta^{13}C$) values and associated radiocarbon ages from soil organic matter (SOM) were used to evaluate vegetation change across five landscape positions at a small enclosed basin in southeastern Arizona. Light and dense SOM fractions were separated to distinguish recent vegetation changes. The direction and timing of vegetation change differed with landscape position along a gentle elevation gradient from the basin outlet to a nearby volcanic ridge top. C_4 perennial grasses have dominated the basin outlet, center, and toe slope landscape positions since at least 5000–6000 years BP, except for the dominance of C_3 plants at the bottom of the outlet excavation at 5000 years BP. This isotopic change is associated with rounded cobbles that may have been a stream channel, suggesting the presence of C_3 herbaceous or woody riparian vegetation. On mid-slope and ridge top landscape positions, where semidesert shrubs now dominate, the proportion of plants with C_4 metabolism calculated from mass balance mixing formulas decreased from approximately 60% as recently as 400 years BP to only 1.5% observed today. The light SOM fraction from mid-slope and ridge top surface soil horizons was approximately 30% C_4 and had a post-bomb date, suggesting that the conversion from grass to shrub occurred over the last several decades.
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1. Introduction

Vegetation change is a critical issue on rangelands throughout the world. The plant community present on a site determines its value for domestic livestock grazing, wildlife habitat, watershed protection, and recreation. One of the most serious problems on rangelands today is the increase in woody species in former grasslands. Since the turn of the century, many grasslands in the southwestern United States have been transformed into savannas or shrublands (Young et al., 1979; Cox et al., 1982; Bahre, 1985, 1991; Archer, 1994; Roundy and Biedenbender, 1995). In southeastern Arizona, written and photographic

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records indicate that since the late 1800s, grasses have declined in many areas, and mesquite (*Prosopis velutina* Woot.), and various shrubs and subshrubs have increased (Humphrey, 1987; Bahre, 1991; Hastings and Turner, 1965; Kidwell et al., 1998). This change in vegetation has often been accompanied by accelerated erosion and the loss of forage resources.

Although the increase in woody species on rangelands is often presented as a ubiquitous ecological phenomenon, neither natural nor anthropogenic vegetation change has been uniform across landforms (Darrow, 1944; Humphrey, 1958; Humphrey and Mehrhoff, 1958; Buffington and Herbel, 1965; McAuliffe, 1994, 1995; Boutton, 1996; Monger et al., 1998). Shrubs may have dominated certain ecological sites prior to European settlement due to soil or geomorphological constraints, creating mosaics of grasslands and shrublands that responded differently to disturbance. Geomorphic location affects soil variables, such as parent material, weathering, texture, depth, and the distribution and storage of soil water. These variables profoundly influence plants at the individual and community scales, particularly in regard to shrub and grass dominance. Different geomorphic surfaces in close proximity and exposed to essentially the same climatic conditions consequently may support very different vegetation. Therefore, it is reasonable to expect that plant communities developed on different geomorphic surfaces respond uniquely to natural and anthropomorphic forces driving vegetation change in any given area (Boutton, 1996; Monger et al., 1998). Understanding the dynamics of past vegetation and landscape-scale patterns of vegetation change allows resource managers to develop realistic goals for productivity, conservation, long-term site stability, and restoration.

The stable carbon (^{12}C and ^{13}C) isotopic composition of soil organic matter (SOM) can provide broad taxonomic resolution capable of distinguishing trees, shrubs, and forbs having the C_3 photosynthetic pathway from warm-season perennial grasses and other herbaceous and woody plants having the C_4 pathway. SOM reflects the carbon isotope composition of the plant material from which it originated, making it an appropriate indicator of the photosynthetic pathway of past vegetation (Deines, 1980; Tieszen and Archer, 1990; Cerling et al., 1993; Kelly et al., 1993; Boutton, 1996). Carbon isotopes in SOM can distinguish changes in vegetation patterns at a very site-specific scale, allowing the spatially distinct distributions of present and past communities to be traced across a landscape of heterogeneous geomorphic features (Boutton, 1996; Boutton et al., 1998).

Although carbon isotopes have been used extensively to investigate vegetation change, few studies have been conducted in southeastern Arizona. Cerling et al. (1998), using carbon isotopes in fossil equid tooth enamel, documented the expansion of C_4 ecosystems in the south central Great Plains and the Chihuahuan and Sonoran Deserts in Arizona and northern Mexico 4–7 million years ago. The encroachment of C_3 oak woodland (*Quercus emoryi* Torr.) and mesquite communities into C_4 grassland–savannas in southeastern Arizona has been demonstrated using stable carbon isotopes from SOM by several studies (McPherson et al., 1993; McClaran and McPherson, 1995; McClaran and Umlauf, 2000; Biggs, 1997). The results of these studies are consistent within the broad ranges of radiocarbon (^{14}C) dates reported, and all show increases in C_3 shrubs in the last 100–1700 years in areas formerly dominated by C_4 grasses. Several researchers in southeastern Arizona have discussed vegetation heterogeneity with respect to geomorphology and its effects on soil and microclimate (Darrow, 1944; Humphrey, 1958; McAuliffe, 1994). However, few studies have examined the relationship between vegetation change and landscape position. Monger et al. (1998) used carbon isotopes (in the form of soil carbonates) to assess change from grass to shrubs across landscape positions in New Mexico and Texas. They found that the conversion from warm-season grasses to desert shrubs accompanied Holocene erosion between 6400 and 2200 years BP, and that the change was more pronounced on middle piedmont slopes than on lower landforms.

A radiocarbon date for SOM represents an average age that includes the oldest, most decomposed material and the youngest, most recently deposited material. Separating SOM into lighter and denser fractions and dating the density separates can provide a means of identifying recent vegetation change (Boutton et al., 1998). The youngest SOM is expected to be associated with the less dense fraction and the older SOM with the denser and smaller sized particle fraction. A bimodal distribution of SOM into active and stable pools having mean residence times of less than a few decades and several hundred to thousands of years, respectively, is well supported (Jenkinson, 1981; Balesdent et al., 1988; Hsieh, 1992, 1993). The light fraction is generally considered to be a transitory pool dominated by readily decomposable plant and animal residues and microbial biomass that decompose in a few years (less than a decade in warm climates) (Trumbore et al., 1995). The light SOM fraction is expected to include bomb ^{14}C from atmospheric weapons testing and to yield a modern or postmodern ^{14}C age. The high-density fraction is composed of physically protected or chemically resistant pools representing SOM integrated over the past several decades to several hundred years (Balesdent et al., 1988; Trumbore et al., 1996).

Light fraction SOM is also expected to be more similar to current vegetation in stable carbon isotope composition than the dense fraction, which has been influenced by decomposition and changes in atmospheric carbon isotope composition due to the burning of fossil fuels since the industrial revolution. Because plant carbon isotope values closely track atmospheric values, this change is reflected in young SOM, with the most recent SOM in the upper part of the soil profile more closely matching current atmospheric carbon isotope composition than that deposited over 200 years ago and now located deeper in the soil.

The purpose of this research was to explore vegetation change across landscape positions on the Walnut Gulch Experimental Watershed (WGEW) in southeastern Arizona using carbon isotopes. Stable carbon and radiocarbon isotopes from SOM were used to construct a long-term vegetation chronology and to evaluate the relationships over time between vegetation and geomorphic location. The relative ages for light and dense SOM were compared to reveal the direction and timing of recent versus older vegetation change. The study area is a small basin surrounded by low volcanic hills, providing an ideal opportunity to study adjacent grasslands and shrublands and the geomorphic surfaces that support each. The ridges with semidesert shrubs are immediately adjacent to the basin with C_4 perennial grasses. SOM from basin outlet, basin center, toe slope, mid-slope, and ridge top landscape positions was evaluated. The modern vegetation composition changes dramatically across these five geomorphic locations, but it is not known if or when the plant communities diverged in the past.

2. Site description

The 150-km^2 WGEW is located in the San Pedro River Valley in southeastern Arizona's Cochise County (Renard et al., 1993). The watershed receives approximately 300 mm of bimodally distributed annual precipitation, with about 70% falling from July through mid-September. Both spatial and temporal precipitation variability are high (Osborn, 1983). Walnut Gulch lies in a transition zone between the Sonoran and Chihuahuan Deserts (Hastings and Turner, 1965; Brown, 1994). The research site is a small sediment-filled basin that is enclosed by ridges and drains only during infrequent, very high intensity precipitation events into the main channel of a larger subwatershed. The site is located in T. 20 S, R. 23 E, NW 1/4 S. 21; UTM coordinates, Zone 12, are 594750 m E, 3505500 m N. The basin is situated on Pleistocene age alluvial fans overlain by Holocene deposits (Angeles Alonso, 1997). There is only a 25-m elevation gradient between the ridges ($\sim$ 1430 m) and the basin ($\sim$ 1405 m), so there are insignificant temperature and precipitation differences that could affect vegetation $\delta^{13}C$ values. The major differences between the geomorphic locations are soils and soil moisture regimes.

A soil pit was excavated on each of the five geomorphic locations (Fig. 1). The outlet excavation was located at the lowest elevation where the basin narrows as it nears a large channel. Although the depth of the pit was greater than 2 m, bedrock was not encountered. The soil is classified as a fine, montmorillonitic, thermic Typic Haplotorrert (Breckenfeld, 1993). It is very deep, well drained, well mixed, clay enriched, and has calcium carbonate accumulation throughout the profile. Stage I carbonate began to develop at 71–81 cm and continued to 228 cm, consistent with a Holocene age for almost the entire profile (Birkeland et al., 1991). The outlet profile exposed well-rounded cobbles near the bottom of the excavation, possibly representing an old stream bed. The beginning of clay skins just above the cobbles suggested an older age at that depth, but

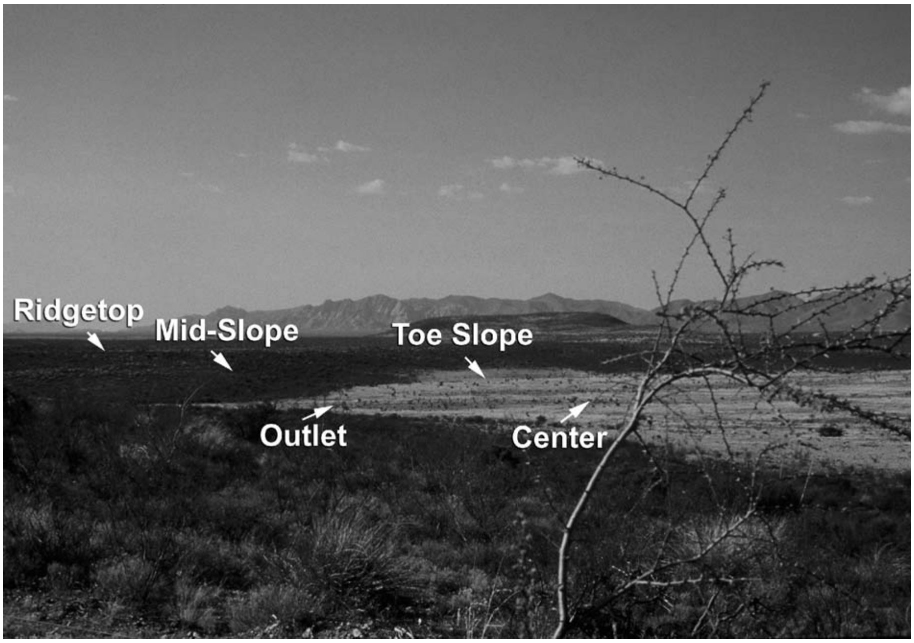

Fig. 1. Overview photograph showing the locations of the five landscape positions where soil was excavated for carbon isotope analysis at the enclosed basin study site on the Walnut Gulch Experimental Watershed.

the clay in the upper profile was probably deposited from eroding surfaces in the upper basin and surrounding hills. There was a buried B horizon under the cobbley alluvium that could be the previous basin surface. The carbonate matrix at the buried B horizon showed Stage III or IV development, bracketing the age between 100 and 500 ka (Birkeland et al., 1991).

The aspect at the basin outlet is that of a swale, with tobosa grass (*Hilaria mutica* (Buckl.) Benth.) sharing dominance with several other C_4 perennial grasses, including sideoats grama (*Bouteloua curtipendula* (Michx.) Torr.), blue grama (*Bouteloua gracilis* (H.B.K.) Lag.), vine mesquite (*Panicum obtusum* H.B.K.), and creeping muhly (*Muhlenbergia repens* (Presl.) Hitchc.). Line intercept transects measured in the fall of 1996 showed 85% of absolute canopy cover and 81% of species relative cover were accounted for by C_4 perennial grasses (Biedenbender, 2000). Annual and perennial forbs comprised 25% of absolute canopy cover and 19% of relative cover.

The basin center landscape position is located at approximately 1–3% slope in the middle of the basin. The basin center soil is a fine, montmorillonitic, thermic Typic Calciargid (Breckenfeld, 1993), also very deep, well drained, and derived from alluvial parent material. Red siltstone was encountered in this excavation at 186 cm. Like the basin outlet profile, clay decreased and quartz increased with depth, implying deposition of clay from surrounding areas. However, clay skins began at 13–26 cm and continued with depth, suggesting either clay development in place or profile stability for a long enough period of time for argillans to form. Soil carbonate was mixed with clay in almost equal proportions in the soil matrix, a pattern not found in the outlet profile, also indicating greater profile stability and/or age. The red siltstone underlying the profile could reflect a depositional environment such as a lake or lagoon at some time in the past (Pleistocene/Pliocene), where low turbulence and weak currents permitted the settling of fine silt-sized particles (Monroe and Wicander, 1995). The siltstone also included large areas of reduction (Shiers, 1997), indicative of an aquatic environment. Basin center vegetation is similar to the basin outlet and is dominated by C_4 perennial grasses, including tobosa grass, blue grama, and sideoats grama, that made up 76% of absolute canopy cover and 88% of relative cover in the 1996 census (Biedenbender, 2000). The forb component was 10% of absolute canopy cover and 12% of relative cover.

The toe slope profile is located on a gently sloping transition (3–5% slope) between the basin and a proximal volcanic ridge. Its classification is a fine loamy, montmorillonitic, thermic Typic Calciargid (Breckenfeld, 1993) developed on mixed alluvium over residuum. The soil is moderately deep and well drained. Red siltstone began at around 91 cm. Clay increased with depth, from about 40% near the surface (5–16 cm) to 70% at 49–68 cm, accompanied by a decrease in quartz with depth. Clay skins were observed throughout the profile, but were most evident in the 32–49-cm sample. The increase in clay with depth and the argillans suggested that the clay developed in place on residual alluvium. Calcite appeared at 32–49 cm as coatings on quartz and occurred as coatings and carbonate grains in the 49–68-cm sample. Coatings on grains signified Stage II carbonate and probably bracket the age of the profile to 10–50 ka (Birkeland et al., 1991).

The toe slope landscape position is a transitional zone between the grasslands of the basin center and outlet and the shrub-dominated uplands. Blue grama was the only C_4 perennial grass present on the toe slope site in the 1996 census, with 30% of absolute canopy cover and 67% of relative cover (Biedenbender, 2000). Perennial forbs comprised 14% of absolute canopy cover and 30% of relative cover. Burroweed (*Haplo-opappus tenuisectus* (Greene) Blake) made up a small component, 1% of absolute canopy cover and 3% of relative cover.

The mid-slope landscape position is approximately equidistant from the toe slope and ridge top. The slope is 10–15%. There are strong similarities between the mid-slope and ridge top soils. Rock fragment ground cover on the slope is almost 100% and is composed of limestone clasts up to 20–30 cm in diameter as well as sandstone and igneous lithics (Osterkamp, pers. Comm., 1997). The profile to a depth of approximately 180 cm contained 75–80% rock fragments by volume. Most of the rocks were less than 5 cm. The profile showed moderate calcium carbonate accumulation throughout, with coatings on all pebbles, hardening into massive calcrete below 180 cm.

Mid-slope vegetation is dominated by large semi-desert shrubs with an understory of subshrubs. Large shrubs are white thorn acacia (*Acacia constricta* Benth.), tarbush (*Flourensia cernua* DC.), creosotebush (*Larrea tridentata* (DC.) Coville), desert sumac (*Rhus microphylla* Engelm.), and bear grass (*Nolina microcarpa* Wats.), a semiherbaceous member of the Nolinaceae family. The most common subshrubs are desert zinnia (*Zinnia pumila* Gray) and dog weed (*Dyssodia acerosa* DC.), with a few mariola (*Parthe-enium incanum* H.B.K.), range ratany *(Krameria parvifolia* Benth.), and burroweed. C_4 grass cover is negligible, comparable to the 1.5% absolute canopy cover measured at the ridge top position (Biedenbender, 2000).

The ridge top soil is a coarse loamy, mixed, thermic Typic Petrocalcid (Breckenfeld, 1993). Ridge top soils are heavily enriched with calcium carbonate, and cemented carbonate begins at about 61 cm, indicating an age of 25,000 to over 400,000 years (Birkeland et al., 1991). Although clay comprised only a small part of the matrix, clay films appeared on the quartz grains, indicating that the soil has been developing in place since before the Holocene. Like the mid-slope landscape position, the ridge top is dominated by large semidesert shrubs (creosotebush, bear grass, white thorn acacia, tar bush, and desert sumac) with subshrubs in the understory (desert zinnia, dog weed, range ratany, mariola, burroweed). C_4 grasses are rare and confined to the protection of shrub canopies. At the excavation site, white thorn accounted for 23% of the absolute canopy cover and subshrubs for 30% in the 1996 census (Biedenbender, 2000). Relative woody vegetation cover was 98%, with only 1.5% C_4 grasses and 0.7% forbs. The C_4 grasses were three-awns (*Aristida* spp.) and black grama (*Bouteloua eriopoda* (Torr.) Torr.).

This study assumes that C_4 grasses were the dominant source of the SOM with stable carbon values characteristic of C_4 biomass because there are no C_4 shrubs and only an occasional succulent having crassulacean acid metabolism (CAM) in the enclosed basin plant communities. There are some C_4 forbs, particularly members of the Euphorbiaceae family. The line intercept transects documented 8% Euphorbiaceae canopy cover at the basin outlet, 4% at the basin center, 11% at the toe slope, and none on the ridge top (Biedenbender, 2000). For these forb species, particularly the annuals, biomass production is expected to be less per canopy cover length than grass production.

3. Methods

3.1. SOM density fractions

Researchers have used various means to separate SOM into chemical and physical components in attempts to isolate the oldest fraction for dating, or the youngest fraction for investigating relationships between soil carbon sequestration and atmospheric CO_2, and to calculate turnover times for various SOM pools (Gilet-Blein et al., 1980; Parton et al., 1987; Balesdent et al., 1988; Becker-Heidmann and Scharpenseel, 1986, 1989, 1992; Martin et al., 1990; Martin and Johnson, 1995). In this study, the $\delta^{13}C$ values of the low-density and high-density SOM fractions were compared in order to separate recent vegetation change from that occurring on longer time scales. The low-density fraction (<2 g/cm^3) was separated from the high-density fraction (>2 g/cm^3) with lithium metatungstate (LMT). This protocol was modeled after that used by Trumbore (1993), Trumbore and Zheng (1996), and Trumbore et al. (1996), who used sodium polytungstate (SPT) for SOM density separation.

3.2. Stable carbon isotopes in SOM

The carbon isotope ratio is expressed relative to the Pee Dee Belemnite (PDB) standard, a limestone from South Carolina that contains 1.123% ^{13}C. Results are calculated as delta carbon-13 values ($\delta^{13}C$) on a per mil basis (parts per thousand, ‰) according to the following formula:

$$\delta^{13}C = [(^{13}C/^{12}C \text{ sample}) - (^{13}C/^{12}C \text{ standard})] / (^{13}C/^{12}C \text{ standard}) \times 1000$$

$\delta^{13}C$ values for C_3 plants range from -22‰ to -38‰ with an average of -27‰, whereas values for C_4 plants range from -9‰ to -21‰ with an average of -13‰, resulting in distinct values between C_3 and C_4 plant tissues (Raven et al., 1981; Salisbury and Ross, 1985; Tieszen and Archer, 1990; O'Leary, 1993).

Soil was collected from excavations in approximately 10-cm increments, depending on the thickness of the horizon, and samples were always taken from immediately above and below horizon boundaries. Soil samples were oven-dried at 100–105 °C before the <2-mm fraction was separated by grinding and sieving. All visible roots, which were not numerous, were removed both prior to and following grinding and sieving. Calcium carbonate was eliminated by leaching with 1N HCl in an ultrasonic bath until effervescence ceased and a low pH was obtained. The length of leaching time varied with the calcium carbonate content of the sample; acid was changed as necessary.

To obtain CO_2 for mass spectrometry, samples were combusted at 900 °C with cupric oxide and silver foil. The evolved CO_2 was cryogenically purified under vacuum using liquid nitrogen to remove moisture and trace gases. The $\delta^{13}C$ values for the CO_2 were determined by analysis using a Finnigan MAT-Delta-S mass spectrometer at the University of Arizona. The precision of the mass spectrometric measurements was ± 0.1‰.

3.3. Mixing models

The proportion of past C_4 versus C_3 vegetation is often calculated using SOM $\delta^{13}C$ values in a simple mass balance mixing formula (Natelhoffer and Fry, 1988; Becker-Heidmann and Scharpenseel, 1992; Cole and Monger, 1994; Nordt et al., 1994; Wedin et al., 1995; Boutton, 1996; Boutton et al., 1998; Bernoux et al., 1998):

$$\delta^{13}C_{meas} = x(\delta^{13}C_4) + (1-x)(\delta^{13}C_3)$$

where $\delta^{13}C_{meas}$ = measured $\delta^{13}C$ values, $\delta^{13}C_4 = \delta^{13}C$ value for C_4 vegetation, $\delta^{13}C_3 = \delta^{13}C$ value for C_3 vegetation, and x = percent C_4 vegetation.

This two-compartment model can be used where the plant community has undergone a change in the photosynthetic pathway of the dominant vegetation. The $\delta^{13}C$ values for past C_3 and C_4 vegetation are based on the average values for contemporary C_3 and C_4 vegetation (Boutton et al., 1998; Nordt et al., 1994; Bernoux et al., 1998). Bernoux et al. (1998), working in central Texas, used estimated values of -13‰ for C_4 and -27‰ for C_3 vegetation. For this study, contemporary values for C_3 and C_4 vegetation (-13.5‰ and -26.4‰, respectively) were obtained by averaging above- and belowground $\delta^{13}C$ values for three individuals of four species of dominant (based on line intercept transects) C_4 grasses and four species of C_3 shrubs. Within-species standard deviations ranged from 0.03 ‰ to 0.6 ‰.

There is a well-documented in situ 1–6 ‰ increase with depth in the $\delta^{13}C$ values of bulk SOM developed from C_3 vegetation. There are several proposed reasons for this phenomenon, including illuviation of ^{13}C-enriched SOM fractions (Becker-Heidmann and Scharpenseel, 1986, 1989, 1992), microbial discrimination or mixing during decomposition (Natelhoffer and Fry, 1988; Wedin et al., 1995; Boutton, 1996; Agren et al., 1996), declining $\delta^{13}C$ values for atmospheric CO_2 since the beginning of large-scale burning of fossil fuel around 1800 AD (Tieszen and Archer, 1990; O'Leary, 1993; Toolin and Eastoe, 1993; Cerling et al., 1997), and the contribution of root biomass with greater $\delta^{13}C$ values than SOM (Biedenbender, 2000; Dzurec et al., 1985; Tieszen and Boutton, 1988; McPherson et al., 1993). It is difficult to separate the effects of diagenesis, post-industrial atmospheric CO_2 $\delta^{13}C$, and roots on the increase in $\delta^{13}C$ values with depth independent of vegetation change, since all three create an increase in $\delta^{13}C$ values of similar magnitude and direction. Nonetheless, the simple mixing formula unadjusted for these confounding variables can result in an overestimation of the proportion of past C_4 vegetation. In this study, results from the standard mixing formula were compared to a modified formula in which the C_3 end-member was increased 2 ‰ from the average value for contemporary C_3 vegetation (−26.4 ‰) to account for the expected increase in $\delta^{13}C$ values with depth.

3.4. Radiocarbon ages for soil organic matter

An exact, absolute numerical age for SOM cannot be determined by radiocarbon dating because of the continuous incorporation of soil carbon from freshly produced biomass. Radiocarbon dates for SOM represent the average age of all the carbon atoms in this open system, and SOM ages are always younger than the oldest organic matter and the age of initiation of soil formation (Gilet-Blein et al., 1980; Scharpenseel and Becker-Heidmann, 1992; Trumbore, 1996; Wang et al., 1996). Nevertheless, ^{14}C can be valuable in determining the relative ages of SOM at increasing soil depths. Solving the following equation for time (t) gives the number of years since the death of the plant or animal tissue being dated:

$$A = A_o e^{-\lambda t}$$

where: A = measured ^{14}C activity (dpm/g carbon); A_o = ^{14}C activity when sample tissue was alive; $\lambda = \ln 2/t_{1/2}$; $t_{1/2}$ = ^{14}C half-life = 5568 years.

Sample-specific ^{14}C activity is compared with that of the National Bureau of Standards oxalic acid (Faure, 1977). Like ^{12}C and ^{13}C, fractionation of ^{14}C occurs during biological and chemical processes, so sample activities are normalized to a $\delta^{13}C$ value of −25 ‰ (relative to PDB). The internationally accepted form for expressing radiocarbon ages is years BP, indicating that the age is calculated from 1950 AD using the original half-life of 5568 ± 30 years (Smart, 1991).

Radiocarbon dates were determined for light and dense fractions from the surface and mid-profile horizons of each landscape position, the deepest dense fraction sample from each position, and the horizon just above the cobbley alluvial horizon at the basin outlet position. Mid-profile samples were selected from depths at which transitions in $\delta^{13}C$ values and/or horizon boundaries were observed. Samples were submitted to the Accelerator Mass Spectrometry laboratory at the University of Arizona in the form of CO_2. Values greater than one yield post-bomb ages and result from ^{14}C enrichment by atomic weapons testing conducted after 1950 through the mid-1960s.

3.5. Statistical analyses

In this study, each soil pit constituted an unreplicated experimental unit remeasured over depth increments. The soil samples themselves were non-independent observations in which there was expected to be a positive correlation between depth and ^{14}C age, violating the analysis of variance assumption of independent observations (Kuehl, 1994). There was also a hypothesized correlation of unknown direction between depth and $\delta^{13}C$ value. Therefore, univariate descriptive statistics (means, ranges, standard deviations, absolute differences) were used to describe the data and address the research questions. The light and dense SOM fractions were statistically compared with paired *t*-tests across all soil depths.

4. Results

4.1. Stable carbon isotopes in SOM

At the basin outlet landscape position, $\delta^{13}C$ values for the present vegetation and the dense fraction throughout the profile reflected the dominance of C_4 plants (Table 1). The buried 2Bt horizon just below the cobbley alluvium at 228–243 cm deviated markedly from this consistently strong C_4 signal. The light SOM fraction was always more negative (depleted in ^{13}C) with respect to the dense fraction by 1.0–2.0‰. The $\delta^{13}C$ values for the light fraction were significantly more negative ($p<0.0001$) than the dense fraction when compared with a one-tail paired two-sample *t*-test. For all landscape positions, the amount of light SOM decreased with depth, resulting in inadequate material for analysis for nine samples.

At the basin center landscape position, the $\delta^{13}C$ value for the surface dense fraction SOM also showed a strong C_4 signal that remained consistently C_4 throughout the profile. The deepest horizon also had a more negative value, but not as extreme as the outlet position. The light fraction was always more negative (0.2–2.0‰) with respect to the dense fraction, except for the 85–102-cm depth in the Bt2 horizon, where the light fraction was slightly more negative. The absolute difference was approximately 1‰ in the A horizon, rising to almost 2‰ at the top of the Bt1 horizons, and falling thereafter. The dense and light fraction $\delta^{13}C$ values were significantly different ($p=0.006$) when compared with a one-tail paired two-sample *t*-test.

For the toe slope landscape position, the $\delta^{13}C$ value for the dense and light fraction SOM reflected a greater mix of C_3 and C_4 vegetation than the basin center and outlet positions. Absolute differences between the light and dense fractions ranged from 0.7‰ to 1.8‰. The dense and light fraction $\delta^{13}C$ values were significantly different ($p=0.0082$) when compared with a one-tail paired two-sample *t*-test.

Mid-slope landscape position $\delta^{13}C$ values for dense SOM also reflected a mix of C_3 and C_4 vegetation with a stronger C_3 trend. The light SOM followed the same pattern, but values were slightly more negative. The dense and light fraction $\delta^{13}C$ values were significantly different ($p=0.0004$) when compared with a one-tail paired two-sample *t*-test. Ridge top dense fraction SOM $\delta^{13}C$ values were similar to the mid-slope position. The absolute difference in $\delta^{13}C$ values between dense and light fractions was 2.7‰ and 3.6‰ for these two depths. The dense and light fraction $\delta^{13}C$ values were significantly different ($p=0.0455$) when compared with a one-tail paired two-sample *t*-test.

4.2. Proportion of C_4 vegetation from mixing models

Comparisons between the standard and modified mixing models (Table 2) revealed minor differences (1–6%) in calculated proportions of C_4 vegetation when $\delta^{13}C$ values were strongly C_4. As $\delta^{13}C$ values became increasingly negative, however, the model predictions diverged, with the standard model overestimating the proportion of C_4 vegetation by up to 15% (mid-slope landscape position, 106–122 cm depth). Applying the mixing formulas to SOM samples having δ^{13} values less than or equal to -13.5‰ (the average value for current C_4 vegetation at the study site) produced values greater than 100% for past C_4 vegetation, indicating that the values for past C_4 vegetation were less negative.

At the basin outlet, both mixing models suggested that the contribution of C_4 species to the dense fraction for the upper 10 cm of soil was over 90%. C_4 species represented 100% throughout the rest of the profile. Percent C_4 dropped sharply at the buried 2Bt horizon, corresponding to the pronounced C_3 signal at the bottom of the excavation just below the cobbley alluvium. The standard model yielded 65% and the modified model 55% C_4.

At the basin center, the percent C_4 vegetation calculated for the dense fraction using the standard model was 93% at the surface, rising to over 100% in the middle horizons and declining to 85% at 137–186 cm. Using the modified model, percent C_4 vegetation was 89% at the surface, also rising to 100% in the middle horizons, and declining to 79% at the lowest strata.

The toe slope percent C_4 vegetation calculated from the standard model for the dense fraction SOM

Table 1
$\delta^{13}C$ values (‰) with soil depth (cm) and radiocarbon ages (years BP) for surface, mid-profile, and basal excavations for dense (>2 g/cm^3) and light (<2 g/cm^3) SOM fractions from five landscape positions at the enclosed basin study site on the Walnut Gulch Experimental Watershed

Site and Horizon	Depth (cm)	Dense $\delta^{13}C$ (‰)	^{14}C Age (years BP)	Light $\delta^{13}C$ (‰)	^{14}C Age (years BP)
Outlet					
A	0–10	−14.1	post-bomb	−15.3	post-bomb
	10–20	−13.2		−14.6	
Bt1	20–31	−12.7		−14.4	
	31–42	−12.7		−14.1	
Bt2	42–52	−12.6		−14.4	
	57–67	−12.4		−14.4	
	71–81	−12.4		−14.2	
Bt3	81–92	−12.4		−13.6	
	92–103	−12.2		−13.9	
	103–114	−12.1		−14.1	
Btk1	114–129	−12.3	3780 ± 55	−13.8	3710 ± 80
	129–143	−13.0			
Btk2	143–163	−13.4		−14.4	
Btk3	163–180	−13.3			
Btk4	180–198	−13.6		−15.0	
Btk5	198–228	−13.5	5305 ± 60	−15.1	
2Bt	228–243	−18.4	5000 ± 70		
Center					
A	0–13	−14.7	post-bomb	−16.0	post-bomb
	13–26	−13.0		−14.1	
	26–40	−12.7		−13.7	
Bt1	40–55	−12.8		−14.6	
	55–70	−13.1		−14.1	
	70–85	−14.5	4910 ± 65	−14.7	
Bt2	85–102	−14.3		−14.1	
	102–119	−14.6			
	119–137	−14.7			
Btk	137–186	−15.8	6075 ± 65		
Cr	186 +				
Toe slope					
Ap	0–5	−15.9	post-bomb	−17.6	post-bomb
A	5–16	−14.0		−14.7	
Btk1	16–32	−13.4	1600 ± 40	−14.6	400 ± 40
	32–49	−13.8		−14.6	
Btk2	49–68	−13.8			
Btk3	68–91	−14.9	3965 ± 40		
Cr	91 +				
Mid-slope					
A	0–15	−18.2	465 ± 55	−21.3	post-bomb
	15–30	−16.3		−18.1	
Bk1	30–40	−16.4		−19.4	
	40–50	−16.7	2065 ± 40	−19.9	415 ± 45
	106–122	−19.5	1755 ± 45	−23.4	
Bk2	183 +				
Ridge top					
A	0–11	−18.5	425 ± 40	−21.2	post-bomb
	11–22	−17.5	975 ± 40	−21.1	post-bomb
Bk1	22–61	−17.7	2045 ± 40		
Bk2					

Table 2

SOM percent C_4 calculated with simple and modified mixing formulas using measured SOM dense fraction $\delta^{13}C$ values (‰) and values of −13.5‰ and −26.4‰ for C_4 and C_3 vegetation, respectively, based on vegetation averages at the study site; percent C_4 in present vegetation was determined from line intercept transect canopy cover

Site and depth (cm)	Dense, standard mixing formula	Dense, modified mixing formula	Light, standard mixing formula	Light, modified mixing formula
Outlet				
Present vegetation	81.1			
0–10	97.0	94.4	78.0	83.6
10–20	104.1	103.1	82.7	89.9
20–31	107.4	107.1	84.0	91.8
31–42	107.9	107.8	86.1	94.6
42–52	108.6	108.6	84.2	92.1
57–67	109.9	110.2	84.0	91.7
71–81	110.1	110.4	85.3	93.6
81–92	109.7	110.0	89.5	99.2
92–103	111.6	112.3	87.1	96.0
103–114	112.4	113.3	86.3	94.9
114–129	110.6	111.0	88.0	97.3
129–143	105.2	104.4		
143–163	102.3	100.9	83.9	91.6
163–180	102.9	101.6		
180–198	101.0	99.2	80.1	86.4
198–228	101.8	100.2	79.2	85.2
228–243	64.9	55.1		
Center				
Present vegetation	88.4			
0–13	92.7	89.1	73.2	76.9
13–26	105.3	104.6	86.2	94.8
26–40	107.9	107.7	88.6	98.0
40–55	106.8	106.4	82.9	90.2
55–70	104.3	103.3	86.0	94.6
70–85	94.1	90.8	81.8	88.7
85–102	95.6	92.7	86.0	94.5
102–119	93.2	89.8		
119–137	92.2	88.6		
137–186	84.5	79.0		
186+				
Toe slope				
Present vegetation	66.9			
0–5	83.9	78.3	62.5	62.2
5–16	97.6	95.2	81.8	88.8
16–32	102.3	100.8	82.9	90.2
32–49	99.7	97.7	82.4	89.5
49–68	99.1	97.0		
68–91	90.7	86.7		
91+				
Mid-slope				
Present vegetation	1.5			
0–15	66.1	56.6	38.0	28.5
15–30	80.6	74.3	59.1	57.5
30–40	80.1	73.6	50.6	45.8
40–50	77.8	70.9	47.0	40.9
106–122	56.4	44.7	24.1	9.3
183+				
Ridge top				
Present vegetation	1.5			
0–11	64.3	54.3	38.7	29.5
11–22	71.6	63.3	39.2	30.1
22–61	69.9	61.2		

was 84% in the 0–5-cm depth, increasing to 100% at 16–32 cm, and decreasing to 91% from 68 to 91 cm. Corresponding values from the adjusted model were 78%, 100%, and 87%.

The mid-slope and ridge top dense fractions were very similar, with standard formula rates of 66% and 64% C_4 at the surface, respectively, approximately 80% and 70% mid-profile, and 56% at 122 cm at the mid-slope position. The modified values were lower: 57% and 54% at the surface, respectively, 71–74% and 61–63% mid-profile, and only 45% for the mid-slope site at 106–122 cm.

For both models at all landscape positions, light fraction predictions followed the same pattern as the dense fraction, but the light fraction always produced lower estimates of C_4 vegetation than the dense fraction. The differences ranged from 2% to 35% less C_4 vegetation in the light fraction SOM.

4.3. Radiocarbon dates for soil organic matter

Radiocarbon dates (years BP) for the dense fraction (>2 g/cm^3) surface soil samples from all five landscape positions were post-bomb except for the ridge top geomorphic location (Table 1). At the basin outlet, the date for the dense SOM at 114–129 cm was 3780 ± 55 years BP, and at 198–228 cm, it was 5305 ± 60 years BP. A stratigraphic inconsistency in age occurred between the 198–228-cm depth just above the cobbley alluvial strata and the deepest layer just below it at 228–243 cm, which was dated at 5000 ± 70 years BP. This was one of two age inversions among the 25 samples dated.

The dense fraction SOM from the basin center 70–85 cm depth was dated at 4910 ± 65 years BP, and at 137–186 cm at 6075 ± 65 years BP. On the toe slope, the 16–32-cm depth had a date of 1600 ± 40 years BP, and the 68–91-cm depth had a date of 3965 ± 40 years BP. Mid-slope dense SOM at 0–15 cm had a radiocarbon age of 465 ± 55 years BP. At 40–50 cm, the age was 2065 ± 40 years BP, and at 106–122 cm, the age was 1755 ± 45 years BP, the second age inversion in this study. Ridge top dense SOM at 0–11 cm was dated at 425 ± 40 years BP, the 11–22-cm depth at 975 ± 40 years BP, and the 22–61-cm depth at 2045 ± 40 years BP.

Except for the surface samples, light fraction (< 2 g/cm^3) ages were always younger than dense fractions at the same soil depths. Light fraction surface samples were also post-bomb in age for all five landscape positions. For the basin outlet, the 114–129-cm depth had a light fraction radiocarbon date of 3710 ± 80 years BP. The toe slope light fraction at 16–32 cm had a date of 400 ± 40 years BP. For the light fraction SOM at 0–15 cm, the mid-slope age was post-bomb, and at 40–50 cm, the age was 415 ± 45 years BP. On the ridge top, both the surface 0–11 and the 11–22-cm depth light fractions were post-bomb in age.

5. Conclusions

5.1. Vegetation change and landscape position

For outlet, center, and toe slope geomorphic locations, there were absolute differences in $\delta^{13}C$ values of $< 1-4$ ‰ between present vegetation, light fraction SOM, and dense fraction SOM for the entire soil profile except the lowest strata. Although statistically significant, these differences were not large enough to warrant a conclusion regarding vegetation change, given that an enrichment in the dense fraction of several ‰ could be attributed to diagenesis, ^{13}C-depleted modern atmospheric CO_2, and ^{13}C-enriched roots. Calculating the proportion of C_4 versus C_3 vegetation from SOM $\delta^{13}C$ values using the mixing formulas supported the conclusion that the basin outlet, center, and toe slope locations remained strongly C_4 throughout almost the entire period of record.

By contrast, both SOM $\delta^{13}C$ values and mixing formula calculations showed dramatic vegetation changes on the upland landscape positions surrounding the enclosed basin. There were absolute differences in $\delta^{13}C$ values between present vegetation (-26.4 ‰) and dense fraction SOM of 9–11 ‰ and light fraction SOM of 4–9 ‰. The modified mixing formula calculations suggested a significant loss of the perennial C_4 grass component, indicating that C_4 plants comprised as much as 63% of ridge top and 74% of mid-slope vegetation in the past compared to only 1.5% now.

The $\delta^{13}C$ values and modified mixing formula calculations for the deepest strata indicated that at some point in the past, conditions at the lower elevations might have been swampy or riparian with a corresponding increase in C_3 herbaceous or woody vegetation. This outcome was not expected from the modern dominance of C_4 perennial grasses and must be interpreted with caution. $\delta^{13}C$ values throughout the basin outlet profile were strongly C_4 except for the intriguing, pronounced C_3 signal at the bottom of the excavation. Just below the cobbley alluvial layer at about 2.5 m, C_4 dropped from 100% to 55%. The center landscape position also exhibited a more negative signal at the deepest strata in the excavations than in the middle strata, dropping from 89% to 79% C_4. The toe slope decreased from 97% to 87% C_4, and the mid-slope decreased from 71% to only 45% C_4. If the outlet cobbley alluvium was an old streambed, this pattern could have originated from C_3 forbs, C_3 grasses or sedges, or C_3 mesic shrubs. Another possibility is that recent deep C_3 shrub roots have penetrated the lower strata and, because the amount of old SOM declines with depth, dominated the isotopic signal. This possibility is eliminated by the ^{14}C dates, which do not indicate recent carbon from deep tree roots.

$\delta^{13}C$ values for light SOM, mixing formula results, and present vegetation canopy cover data indicated a small increase in C_3 vegetation in the present plant communities at the outlet landscape position. The surface horizon (0–10 cm) light SOM $\delta^{13}C$ value (-15.3 ‰) was more negative than the value for current perennial C_4 grasses (-13.5 ‰). The modified mixing formula prediction for percent C_4 vegetation from surface horizon light fraction SOM (84%)

were very close to field measurements of C_4 canopy cover (81%), compared to estimates of 90–99% C_4 at 10–20 cm and deeper.

5.2. Timing of vegetation change

Radiocarbon dates provided a chronology for the vegetation changes suggested by the $\delta^{13}C$ SOM values. The SOM samples for all five landscape positions were mid- to late Holocene in age. Since SOM is an open system continually replenished by the addition of new material, the dates represent minimum ages. Keeping this caveat in mind, perennial C_4 grasses have dominated the lower elevations in the basin, as they do today, since at least 5000–6000 years BP. From at least 2000 to 400 years BP, C_4 grasses also made up a significant proportion of the vegetation on the slopes and ridges, where today C_4 grasses are practically nonexistent. The surface horizon dense fraction SOM radiocarbon date at the ridge top (425 ± 40 years BP) and the mid-slope (465 ± 55 years BP) suggested the possibility of a truncated soil from which the youngest dense SOM has been lost by erosion. The post-bomb dates for surface horizon light fraction SOM at the ridge top and mid-slope, combined with the modified mixing formula estimates of 30% and 29% C_4 vegetation, respectively, at those depths, suggests recent timing for grass declines on the slopes and ridge top.

One of the most intriguing questions posed by the $\delta^{13}C$ results concerned the time frame for the existence of the C_3 vegetation at the bottom of the outlet profile. The trend towards an increase in C_3 vegetation found at the bottoms of the basin outlet, basin center, toe slope, and mid-slope excavations in this study area suggested the possibility of a more mesic environment prior to at least 5000–6000 years BP, followed by an increase in grasses accompanied by mid-Holocene warming and drying after that time. The enclosed basin carbon isotope results agreed with those from packrat middens (Van Devender, 1990) and the Murray Springs pollen record (Mehringer et al., 1967) for a more mesic Altithermal in southeastern Arizona. The presence of a substantial proportion of C_3 vegetation on the mid-slope and ridge top landscape positions by at least 2000 years BP was also consistent with packrat midden analyses by Van Devender (1990, 1995) showing that desert shrubs, particularly creosotebush, appeared in the Chihuahuan Desert during the late Holocene around 4000 years BP. Studies by Monger et al. (1998) and McClaran and McPherson (1995) also suggested that woody increases were under way prior to anthropogenic influences such as livestock grazing and increasing atmospheric CO_2.

The results of this study supported evidence from the historical record (Humphrey, 1953, 1958; Hastings and Turner, 1965; Bahre, 1985, 1991; Kidwell et al., 1998) and other isotope studies (McPherson et al., 1993; McClaran and McPherson, 1995; Biggs, 1997; McClaran and Umlauf, 2000) for a decline in C_4 perennial grasses on southeastern Arizona rangelands, with an important qualification. The decline in grasses did not occur consistently across heterogeneous landscape positions despite exposure to similar land uses and climatic cycles. In addition, the separation and dating of dense and light fraction SOM in this study indicated that the decline in mid-slope and ridge top grasses occurred within the last several decades. These results contradicted Darrow's (1944) assertion that in Cochise County, where the WGEW is located, the grassland–desert shrub vegetation type associated with rocky slopes had undergone little change in species composition since European settlement.

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