Carbon and nitrogen stable isotope analysis of hunter–gatherers from the Coleman site, a Late Prehistoric cemetery in Central Texas

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Abstract

We report on a stable isotopic analysis of 17 hunter–gatherer burials from the Coleman site (41BX568), a Late Prehistoric Toyah Interval (700–350 years BP) occupation in Texas. Prior to our analysis, isotopic research on Toyah populations in Central Texas was represented by a single burial at site 41BX677. That burial showed an isotopic pattern suggestive of a diet heavily focused on CAM/C4 plants and C4 fauna. Coleman burials show a different pattern. While interpretations are complicated by high variability in the isotopic signatures of children and by differences in male and female diets possibly related to mate exchange, the 11 adult and adolescent burials at Coleman show a diet focused on C3 fauna and the use of both C3 and CAM/C4 plants. The moderate CAM/C4 plant use is a radical departure from a trend of increasing C3 plant use that characterized hunter–gatherers in this region for at least 6200 years prior to the start of the Toyah Interval. Protein sources among Coleman adults probably centered on deer, but also included high nitrogen (15N) animals, such as fish. Males seem to have differential access to these high nitrogen sources. Two different isotopic patterns, one reflecting a focus on C3 fauna and moderate use of CAM/C4 plants, and a second reflecting C4 fauna and extensive use of CAM/C4 plants, are represented during Toyah. While interpretations are complicated by small sample sizes, these two patterns could simply reflect temporal differences, different acquisition strategies based on availability, or hint at different subsistence strategies. It may also be the case that the 41BX677 individual represents an immigrant into the Central Texas region, one with a different isotopic history.

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1. Introduction

Much of prehistoric Texas maintained hunter–gatherer adaptations until historic contact (see Johnson and Hard, 2008). While we have adequate data, the ability to sustain a hunter–gatherer subsistence base appears to be related to humans intensifying on locally available plants and aquatic resources. For example, coprolites (e.g., Riley, 2012; Sobolik, 1988), floral and faunal remains (e.g., Dering, 1979; Lord, 1984), and bone isotope studies (e.g., Huebner, 1991) from dry shelters in the Lower Pecos region of Texas (Fig. 1) suggest the exploitation of seasonally available resources and provide evidence of the intensification on aquatic resources (e.g., Jurgens, 2008) and succulents (e.g., Dering, 1999). Recent isotopic work on human burials by Hard and Katzenberg (2011) suggests that intensive use of marine and freshwater resources supported large populations (e.g., Ricklis, 2004) in the Texas Coastal and Riverine Zones (Fig. 1). Detailed patterns of hunter–gatherer subsistence are not well documented in South Texas (Fig. 1) because of a dearth of investigation (Hester, 2004; Hester et al., 1989). More work has been done in Central Texas (Fig. 1), but we often lack high quality data sets (see Collins, 2004) that can be tied directly to diet (e.g., coprolites). Well-preserved floral and faunal assemblages are rare, and subsistence details are often inferred by indirect methods such as changes in feature frequency and type (e.g., Black and Creel, 1997; Thoms, 2009), and technological shifts (e.g., Tomka, 2001).

Researchers in Central Texas have not vigorously pursued the isotopic study of human remains, a source of direct subsistence data. Here we review extant isotopic data on populations in the Central Texas region. Burials dating to the prehistoric period with collagen carbon and nitrogen data, as well as carbon data isolated in carbonate from bone apatite, are limited. There are
reports on five burials with isotopic data for the Early Archaic (8900–6000 years BP), four from the Middle Archaic (6000–4000 years BP), and 17 interments for the Late Archaic (4000–1200 years BP) period. The Late Prehistoric period, divided into the Austin (1200–700 years BP) and Toyah (700–350 years BP) Intervals (Collins, 2004; Turner et al., 2011), consists of 15 burials with isotopic data, but 13 of these are from the earlier Austin Interval.

Following our review, we present an isotopic analysis of 17 interments using carbon and nitrogen from bone collagen and carbon from bone apatite. The samples were recovered at the Coleman site (41BX568; Potter et al., 2005) in Central Texas (Fig. 1). Radiocarbon dates on collagen from seven of these burials show a restricted time range (656–506 years BP) in the Toyah Interval. Excluding six pre-adolescent interments that had different diets, the 11 adult and adolescent Coleman individuals (6 males, 4 females, 1 undetermined) suggest an increasing dependence on C_4 and/or CAM plant resources, which is consistent with substantial use of plants such as succulents. These results reflect a radical divergence from a trend of increasing C_3 dependence that characterized the Central Texas region for at least 6200 years (ca. 6900–700 years BP). Collagen carbon and nitrogen values suggest the possibility that whitetailed deer may have been a significant protein source for the Coleman population, though there are hints of the consumption of other animals with high δ^{15}N values, such as fish, present in selected burials. While sample sizes are small, males dominate these cases of elevated δ^{15}N. There is no evidence in the Coleman isotopic patterns for a dependence on bison. Most researchers suggest these animals returned to the region in large numbers around 700 years BP after an absence of several centuries (Collins, 2004; Dillehay, 1974) and that these animals were an important food (e.g., Johnson, 1994; Shafer, 1977). The Coleman samples also diverge from the pattern shown by the previously reported Central Texas Toyah Interval isotopic sample that hints at a focus on bison (Cargill, 1996). The isotopic data suggest that intensification in this case is complex, with the possibility that at least two different subsistence systems, focused on different resources, may have been in place during the Toyah Interval. Alternatively, this may reflect previously unrecognized temporal patterns within the Toyah Interval, or may reflect isotopic signatures from outside the Central Texas region, with individuals migrating into the area and retaining some component of their isotopic signature of origin.

1.1. Stable carbon and nitrogen isotope analysis and paleodiet

Stable isotope research relies on established relationships in chemistry and biology (see Ehleringer, 1991; Sharp, 1997). Applications to prehistoric human populations rely on the observations that while bone turnover rate varies with age, sex, and type of bone (see Hedges et al., 2007; Parfitt, 2002), isotopic ratios of carbon and nitrogen in human bone reflect the average isotopic values of these elements in an individual’s diet over the last decades of life (Mays, 1998; Tykot, 2004). Stable isotopic ratios have been widely used for paleodietary reconstructions, and overviews of the methods, applications, and potential pitfalls can be found in a variety of sources (e.g., Ambrose, 1993; Katzenberg, 2008; Kellner and Schoeninger, 2007; Krueger and Sullivan, 1984; Lee-Thorp, 2008; Schwarzc, 2000; van der Merwe, 1992).
To understand the process by which stable carbon and nitrogen isotopes in bone reflect diet, we begin with considering how these isotopes are incorporated in plants. During photosynthesis, terrestrial plants incorporate atmospheric carbon into their tissue using the C3, C4, or CAM pathway (see Farquhar et al., 1989; O’Leary, 1988). These pathways have evolved in response to different ecological conditions (see Long, 1999). The C3 pathway is the most common, and plants that use this pathway, which discriminates against the heaviest isotope of carbon, include all trees, most bushes and shrubs, and cool season grasses (see Ehleringer et al., 1997; Ehleringer and Cerling, 2001). C3 plants have δ13C values that range from −37‰ to −20‰ (Kohn, 2010). Plants that use the C4 pathway, a pathway dominated by warm season grasses, have a more positive δ13C range, from ca. −16‰ to −9‰ (Deines, 1980). Cacti and other succulents dominate the third photosynthetic pathway, CAM. CAM plant δ13C ranges are variable (−10 to −20‰) and primarily overlap with the C3 group (Boutton et al., 1998; Cockburn, 1985; Griffiths, 1992).

The movement of carbon in terrestrial plants is relatively straightforward. However, marine and freshwater systems are more complex due in part to a greater diversity of available carbon sources. In addition to atmospheric CO2, aquatic carbon sources include dissolved inorganic carbonate and organic carbon incorporated into rivers and oceans. These sources are often enriched relative to atmospheric carbon, especially in ocean settings (Boutton, 1991). Plants and aquatic organisms in such settings tend to have more positive δ13C values (Boutton, 1991; Chisholm et al., 1982).

Our understanding of variation in nitrogen stable isotope values (δ15N) is underdeveloped. Enrichment is primarily tied to trophic levels in a given food web (Ambrose, 1986; Bocherens and Drucker, 2003; DeNiro and Epstein, 1981). Atmospheric nitrogen (N2) is the ultimate source of nitrogen in an ecosystem and has a δ15N of 0‰ (Mariotti, 1983), with soils commonly having slightly more positive δ15N values. Legumes absorb atmospheric nitrogen released through bacterial action, and these plants tend to have δ15N values that are between 1‰ and 3‰. Non-leguminous plants obtain nitrogen from the decomposition of soil organic matter by bacteria. These plants have higher δ15N values, with most falling between 2‰ and 6‰ (Ambrose, 1991; Shearer and Kohl, 1986).

Several factors can influence nitrogen ranges in soils and plants, as well as the δ15N in bone collagen of animals that feed on those plants. Nitrogen values in bone can increase, for example, under conditions of high evaporation and aridity (Ambrose, 1986; Cormie and Schwartz, 1996; Heaton, 1987; Heaton et al., 1986; Pate and Anson, 2008; Ugan and Coltman, 2011). There is also a stepwise enrichment in nitrogen values in bone that is tied to trophic levels (Ambrose and DeNiro, 1986; Bocherens and Drucker, 2003; DeNiro and Epstein, 1981). The δ15N found in bone collagen of herbivores feeding on plants is enriched 3‰–4‰ above the value of the plants consumed. A similar increase is present in omnivores and carnivores. Because there tends to be more steps in the food chain, this trophic increase in δ15N is especially apparent in aquatic food webs (see Minagawa and Wade, 1984).

When humans consume plants and animals the δ13C and δ15N isotopic signatures present in these resources are incorporated into the body with an additional fractionation or enrichment (Ambrose, 1993; Ambrose and Norr, 1993; Katzenberg, 2008). Different bone components have different patterns of incorporation. Bone collagen, the organic component of bone, contains both carbon (13C) and nitrogen (15N) isotopic signatures. Nitrogen isotopic values reflect protein intake, with animals being the primary nitrogen source in most human diets. Although the relationship is complex (Froehle et al., 2010), several researchers have shown that carbon in bone collagen also reflects intake related to protein (e.g., Ambrose et al., 1997; Kellner and Schoeninger, 2007; Krueger and Sullivan, 1984). The principal complication in linking collagen to diet is related to whether the dietary protein is from a C3 or a C4/ marine protein source. Controlled dietary studies (e.g., Ambrose and Norr, 1993; Jim et al., 2004, 2006; Tieszen and Fagre, 1993) have shown that there is a strong, linear relationship between δ13C values in collagen and diet but only within a C3 or a C4/marine protein group (Kellner and Schoeninger, 2007). In contrast, these same controlled studies demonstrate that δ15N in bone apatite is strongly correlated with the isotopic signature of the whole diet (Froehle et al., 2010; Kellner and Schoeninger, 2007). Apatite δ15N values alone provide no information about the protein source. Collagen and apatite are complementary in that the use of stable isotope data from both tissue types provides a more detailed reconstruction of past diets.

1.2. Previous isotopic research into prehistoric human diets in Central Texas

Stable carbon isotopic research on prehistoric human remains in Texas dates back at least to the early 1980s (e.g., Bement, 1994; Bousman, 1990; Huebner, 1991, 1994; Huebner and Comuzie, 1995). Work has been sporadic over the decades. In some studies, only collagen carbon, usually in association with radiocarbon dates, is included in the analysis (e.g., Rice, 2006) or the temporal placement of samples is unclear (e.g., Alvarez, 2005). Prior to the work reported here, only 41 samples of human bone with good temporal assignments, nitrogen isotopic data, and carbon isotopic information from collagen and apatite could be located for Central Texas. These samples come from seven sites that span roughly 6500 years in time. Table 1 summarizes these Central Texas isotopic data.

Many of the Archaic period Central Texas isotopic data are from 41KR241 (Fig. 1; Bement, 1994). Sixteen burials analyzed from this site span roughly 4900 years (ca. 6900–2000 years BP) and account for all Early (n = 5) and Middle (n = 4) Archaic samples, as well as seven Late Archaic samples (Table 1). No information on sample pretreatment, analytical conditions, or sample quality measures (e.g., C:N ratios) is available for the 41KR241 samples. Additional Late Archaic samples come from 41BX1 (Fig. 1; Table 1) and are reported by Hard and Katzenberg (2011; see Lukowski, 1988). Six interments from this site represent a restricted time range (2350–1690 years BP). Four burials from 41HY161 and 41HY163 (Fig. 1), which date between 3510 and 1215 years BP (Munoz et al., 2011), complete the Late Archaic samples (Table 1).

There are 15 Late Prehistoric burials with isotopic data reported for Central Texas. Based on the recovery of an Edwards style projectile point (see Turner et al., 2011), Cargill (1996) reports an Austin Interval burial at 41BX952 (Fig. 1; Table 1). Data on file at the Texas Archeological Research Laboratory list isotopic results from 12 Austin Interval individuals at 41WM230, the Loeve-Fox site (Huebner, 1995; Previtt, 1974, Table 1 and Fig. 1). Cargill (1996) also reports isotopic data for an individual from 41BX677 (Fig. 1; Table 1) directly dated to 530–300 years BP (Tennis, 1994). The date encompasses the end of the Toyah Interval (700–350 years BP) and some of the Protohistoric (350–250 years BP). Finally, Munoz et al. (2011; see also Lohse, 2011) report data from an individual dating to 515 ± 20 years BP from 41HY161 (Fig. 1; Table 1). With the exception of the recent work on 41HY161, no information on sample pretreatment, analytical conditions, or sample quality is available for these Late Prehistoric samples.

When we consider the stable carbon δ13C values for collagen and carbonate (apatite) for the 41 previously analyzed Central Texas individuals (Fig. 2; Table 1), 39 cluster near the C3 protein line developed by Kellner and Schoeninger (2007; Froehle et al., 2010). This suggests a dependence on C3 feeding animals (e.g., deer). There
is a strong temporal pattern within this C₃ protein group with an increase in the use of C₂ resources through time in the overall diet as shown in the δ¹³C_Carbonate values (see Bement, 1994:104), which should correlate with overall diet. The Early Archaic samples cluster to the right along the δ¹³C_Carbonate scale and have a δ¹³C_Carbonate mean of −7.2‰. This value decreased to a mean of −8.8‰ in the Middle Archaic and fell to −9.7‰ by the Late Archaic. This trend of increasing C₂ resources continues into the subsequent Austin Interval. The δ¹³C_Carbonate average of −13.1‰ represents a 3.4‰ decrease from the Late Archaic mean. There is a 5.9‰ shift from the Early Archaic mean to the Late Prehistoric Austin Interval δ¹³C_Carbonate mean. This trend of greater dependence on C₂ resources, and thus C₂ plants, is consistent with increased use of geophytes such as camas (Camassia scilloides) and, to a lesser extent, C₂ sotol (Dasylirion sp.) over this period (see Black and Creel, 1997; Dering, 2003; Mauldin et al., 2003; Thoms, 2008, 2009).

Two burials in Fig. 2 do not fit this C₃ pattern. Both date to the Toyah period, the same period as the Coleman material. Carbon values derived from collagen (−10.0‰; −10.35‰) and apatite (−5.3‰; −7.4‰) from these burials, one from 41BX77 (Cargill, 1996) and a second from 41HY161 (Munoz et al., 2011), cluster near the C₄ protein line. A dependence on C₄ protein and C₄/CAM plants is indicated (see Table 1). C₄ grazing bison are a primary candidate for that protein source given the post-700 year BP age (see Collins, 2004; Dillehay, 1974; Shafer, 1977). However, both samples have higher nitrogen values than would be expected with a dependence on bison. Seventeen Central Texas bison dating to this same time have an average δ¹⁵N value of 6.2‰ ± 0.9 (Lohse et al., 2012). A dependence on bison would produce nitrogen values in humans of about 9.7‰, assuming a trophic enrichment of 3.5‰. The δ¹⁵N value for the 41HY161 sample is 13.3‰, well outside the range produced by bison consumption. Munoz et al. (2011:348–349) conclude that a diet dependent on marine resources is the most likely candidate for this carbon and nitrogen signature and that this burial probably reflects the inland migration of a coastal resident. The individual's isotopic patterns are consistent with coastal burials (see Hard and Katzenberg, 2011). We do not consider this sample further. The 41BX677 individual's nitrogen value is 10.7‰ (Cargill, 1996:120–122). This δ¹⁵N value may reflect a dependence on bison, especially if other high nitrogen sources (e.g., fish, soft-shell turtle) were included in the diet. However, many of these other high nitrogen sources have a C₄ carbon signature in inland settings (see Hard and Katzenberg, 2011), and such a carbon signature is not reflected in the location of this case on the C₃/marine protein line (Fig. 2). While substantial bison use seems to be a possible scenario for the 41BX677 burial, it is...
conceivable given both the high $^{15}$N value and the strong C4 diet signature, that the 41BX677 individual may also reflect an individual from outside the region.

2. Materials and methods

2.1. The Coleman site (41BX568)

Regardless of what accounts for the differences in the 41BX677 burial, this Central Texas Toyah age individual consumed a radically different diet when compared to earlier samples in Table 1. The increasing dependence on C3 plants and animals characteristic of the previous 6200 years (6900–700 years BP) is suddenly replaced by C4 animals and C4/CAM plants. The isotopic analysis of the Coleman material (41BX568; Fig. 1) provides an opportunity to confirm this pattern and refine our understanding of this drastic shift in resource structure.

Excavated in late 1995 and early 1996 under salvage conditions by volunteers associated with the Southern Texas Archaeological Association and the Texas Historical Commission, the Coleman site minimally contained partial remains from 20 individuals buried in two distinct groups within a sand and gravel quarry (Fig. 3). Seven burials were present in the first group with at least 13 individuals present in Group 2 (Potter, 2005a; Pickering and Potter, 2005a,b). Portions of the second burial group had been extensively disturbed. Both groups were at roughly the same elevation, and the initial cluster was discovered 50–90 cm below a buried surface from which excavators suggested that the burial pits originated (Potter, 2005b). Two small hearths (Features 5 and 7 in Fig. 3) were present on that buried surface. Charcoal from those features produced corrected, calibrated dates of 705–621 years BP and 725–542 years BP at a two-sigma range (Potter, 2005b:25). Researchers (Pickering and Potter, 2005a,b) suggested that the remains dated to the end of the Austin Interval (1200–700 years BP), although the ranges primarily overlap with the Toyah interval. The suggested temporal placement in the earlier period was influenced by the recovery of a broken Austin Interval arrow point near one of the disturbed burials (Pickering and Potter, 2005b:55) and the previous absence of Toyah Interval cemeteries.

The remains recovered included one neonate and six children, which we combine into a group labeled “immature,” and three adolescents (burials 12, 13b, 16b) and ten adults that we designate as “mature.” Six burials are male, four are female, and the remaining ten were fragmentary and sex could not be determined (Pickering and Potter, 2005a). The remains are housed at Our Lady of the Lake University. In 2012, we began an analysis of these remains in order to clarify their temporal placement through direct bone collagen radiocarbon dating of selected samples and to develop dietary information through stable isotopic analysis of nitrogen and carbon in bone collagen and carbon in carbonate isolated from bone apatite.

2.2. Bone preparation methods

We selected bone fragments, primarily consisting of rib segments, from 17 different burials, from the Coleman site for stable isotope analysis and selective radiocarbon dating. No samples were available for burials 10, 11, and 14a, all in the more heavily disturbed second group (Fig. 3). All sample preparation was conducted at the Center for Archaeological Research at The University of Texas at San Antonio (CAR-UTSA). For all analyses, initial steps focused on cleaning bone samples. A rotary tool with a sanding attachment was initially used to lightly clean any foreign material observed on the bone surface. This was followed by
multiple cleanings in ultra-pure water in an ultrasonic bath. When the rinse water was clear, we removed and dried the samples under low heat.

For collagen samples, we crushed dried bone into small fragments (0.5–2 mm size) with a ceramic mortar and pestle and sonicated them in ultra-pure water. We changed water after each run, and the process continued until the rinse water was clear. About 100 mg of dried bone was then weighed into glass test tubes. Samples were decalcified by reacting with 0.5 N HCl at 4°C for 30 h (Bocherens et al., 1991; DeNiro and Epstein, 1981; Longin, 1971). We rinsed samples to neutral and subsequently treated them with 0.1 N NaOH for up to 45 min. The samples were again rinsed to neutral. They were then solubilized in 0.01 N HCl at 70°C for 11 h. The supernatant was filtered into glass vials, frozen, and freeze-dried under vacuum. Once dried, 600 µg of collagen sample was placed into tin capsules for bulk stable carbon and nitrogen isotope analysis. For the Coleman collagen samples, we processed sample sets that include 12 individual samples at a time, including two samples of modern deer with known isotopic ranges used as internal standards.

For dating, we followed an acid–base–acid procedure for collagen preparation (see Brock et al., 2010; Minami et al., 2004). We initially crushed samples with a ceramic mortar and pestle. These were then sonicated in ultra-pure water, with the water changed after each run until the rinse water was clear. We then dried samples at low heat. For a given sample, we weighed out two changed after each run until the rinse water was clear. We then weighed out two changed after each run until the rinse water was clear. We then weighed out two.

Samples were decalci
dried samples at low heat. For a given sample, we weighed out two.

We initially crushed samples with a ceramic mortar and pestle.

They were covered with 0.5 N HCl and refrigerated for 18 h. The samples were again rinsed to neutral. They were filtered into glass vials, frozen, and freeze-dried. The samples were then solubilized in 0.01 N HCl at 4°C for 30 h. After washing to neutral, the sub-samples were treated with 0.1 N NaOH for up to 45 min at room temperature and again washed to neutral. They were covered with 0.5 N HCl and refrigerated for 18 h. The 0.5 N HCl was replaced with 0.01 N HCl without exposing the decalcified bone to air. Samples were then solubilized in a dry bath at 70°C for 20 h. The liquid was then filtered into glass vials, frozen, and freeze-dried. Sample vials were sealed and shipped to DirectAMS for analysis. Ultrafiltration methods (see Potter and Reuther, 2012) were not used on these samples as we anticipated the bone to be no older than 1000 years (see Potter, 2005b:25).

Following the initial rotary cleaning and ultrasonic wash, bone fragments designated for carbon isotope analysis from apatite were sampled with a rotary drill at low speed. About 150 mg of ground bone was weighed into glass test tubes, to which we added a 5% phosphoric acid and incubated for at least 1 h at 70°C to produce CO2 for analysis. Isotopic standards used for carbonates at the CPSIL include NIST standards (NBS 18, NBS 19) and lithium carbonate (LSVEC). Collagen and carbonate δ13C values are reported in per mil relative to the Vienna Pee Dee belemnite standard and δ15N values are reported relative to AIR. Based on replicative analysis, the CPSIL has an uncertainty of ±0.10‰ for δ13Ccollagen, ±0.20‰ for δ15N, and ±0.10‰ for δ13Ccarbonate.

3. Results and discussion

The results of the stable isotopic analysis of the 17 burials are shown in Table 2. Data are reported for each burial with data on multiple runs from a given burial averaged. The collagen samples all had atomic C:N ratios between 3.2 and 3.4, within the commonly accepted range of 2.9–3.6 (Ambrose and Norr, 1992; DeNiro, 1985; van Klinken, 1999). All samples also had good δ13C and δ15N returns (Table 2) further suggesting that the collagen was of good quality (Ambrose and Norr, 1992). The analysis of samples from three modern deer (Deer1, Deer2, and Deer4) used as internal standards at CAR-UTSA produced expected results for carbon and nitrogen in collagen. For Deer1, a single collagen analysis prepared at the same time as the Coleman samples produced a δ13C value of −20.2‰, and a δ15N of 6.2‰ (Deer1 mean of 52 runs δ13C = −20.1±0.1‰, δ15N = 6.1±0.1‰). Two samples from Deer2 produced an average δ13C of −20.8‰ and a δ15N of 6.3‰ (Deer2 mean of 33 runs δ13C = −20.8±0.3‰, δ15N = 5.9±0.3‰), and a collagen sample from Deer4 yielded a δ13C of −22.7‰ and a δ15N of 4.3‰ (mean of 27 runs δ13C = −22.8±0.2‰, δ15N = 4.0±0.2‰).

The values of the CAR-UTSA internal standards processed and run with the Coleman carbonate samples suggest that the procedures used yielded consistent results. When run with the Coleman samples, our Deer2 carbonate produced a carbon value of −14.6‰ identical to that of the mean δ13Ccarbonate based on three previous runs, while the Deer4 carbonate yielded a δ13Ccarbonate value of −15.3‰ slightly higher than the Deer4 average of −15.0‰ (n = 3). We did not conduct an independent assessment of the quality of the carbon recovered from Coleman bone carbonate samples.

We processed collagen for radiocarbon dating from seven samples (Table 2). The dates fall within a small range (Table 3, Fig. 4a, b). Considering the minima and maxima of the calibrated two-sigma ranges, the seven calibrated dates cluster between 656 and 506 years BP (Table 3) with OxCal (Bronk Ramsey, 2009); a time range that is in the middle of the Toyah Interval (700–350 years BP).
The values suggest that diets associated with the immature group enriched in $^{15}$N relative to the mature samples. There is also collagen sample information and radiocarbon results for Coleman (41BX 568). Table 3 presents box plots for (Fig. 4b). The OxCal Span function (not shown; see Bronk Ramsey, 550 cal years BP, with an end use between 630 and 470 cal years BP beginning use of the Coleman cemetery at between 700 and (Turner et al., 2011:51). The OxCal Boundary function places the Collagen sample information and radiocarbon results for the Coleman site.

### Table 2

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<td>12.95</td>
<td>3.35</td>
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<td>9.5</td>
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<td>3.23</td>
<td>-8.8</td>
<td>2</td>
<td>Male</td>
<td>Yes</td>
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<sup>a</sup> 1 = immature (0–10 years of age); 2 = mature (+10 years of age).

3.1. Isotopic patterns reflected in the Coleman burials

There was considerable variability in the stable carbon and nitrogen data from the 17 Coleman burials analyzed (see Table 2). The major source of this variation can be seen in Fig. 5, which presents box plots for $\delta^{13}$Ccollagen, $\delta^{13}$Carbonate, and $\delta^{15}$N contrasting immature (0–10 years of age) burial data (n = 6) with those values generated from mature (adult and adolescent) skeletons (n = 11). The values suggest that diets associated with the immature group probably contained more C3 resources and were also slightly enriched in $^{15}$N relative to the mature samples. There is also significantly greater variability in the diets of the immature group relative to the mature group, despite the smaller sample size of the immature group. Some of these differences may be related to patterns of breastfeeding and weaning behavior (see Clayton et al., 2006; Katzenberg and Pfeiffer, 1995). While we lack sufficient sample size and age data to explore these differences, diets associated with the single neonate and five children were isotopically distinct from those of older individuals in the Coleman sample (Fig. 5). Because most comparative isotopic data sets in the region lack information on children, we focus on the 11 mature individuals in subsequent discussions.

The $\delta^{13}$Ccollagen and $\delta^{13}$Carbonate values for the 11 mature Coleman burials are shown in Fig. 6. The Coleman burials tend to cluster near the C3 protein line, suggesting a reliance on C3 animals. The $\delta^{13}$Carbonate values, however, reflect moderate use of C4/CAM resources in the overall diet. Likely candidates from this area are CAM succulents, such as prickly pear, which have low protein content (Hard and Katzenberg, 2011). The consumption of this or similar CAM resources would result in an increased $\delta^{13}$Carbonate signature without significantly increasing the $\delta^{13}$Ccollagen Values. The $\delta^{13}$Carbonate values for the Coleman burials, which are influenced primarily by protein intake, are consistent with the consumption of several different animals that feed on C3 plants (e.g., deer).

Also identified in Fig. 6 is the sex of the Coleman individuals for the 10 cases where the sex of the individual could be determined (Table 2; Pickering and Potter, 2005a). While the mean $\delta^{13}$Carbonate and $\delta^{13}$Ccollagen Values for males ($-8.6^{\text{male}}, -16.4^{\text{male}}$) in our sample are essentially identical to those of females ($-8.8^{\text{female}}, -16.4^{\text{female}}$), greater isotopic variability is reflected in the four female isotopic results relative to the six males (Fig. 6), especially in $\delta^{13}$Ccollagen.

The standard deviations on $\delta^{13}$Ccollagen for males and females are $0.64^{\text{male}}$ and $0.90^{\text{female}}$, respectively. While the standard deviation on the female $\delta^{13}$Carbonate data ($1.49^{\text{female}}$) is over three times as great as that for males ($0.41^{\text{male}}$). The greater variability in female isotopes and the clustering of male isotopic values is intriguing, especially given recent arguments by Bouman and Quigg (2006). Working with isotopic data from the Chihuahuan Desert, the Central Texas region, and the Lower Pecos area, and assuming that the isotopic distribution of a given population of hunter–gatherers would form a normal distribution, they

### Table 3

<table>
<thead>
<tr>
<th>Collagen samples</th>
<th>NAU</th>
<th>NAU</th>
<th>DirectAMS</th>
<th>Radiocarbon age</th>
<th>Calibrated years BP</th>
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<td>3.22</td>
<td>-15.58</td>
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<td>611</td>
<td>655–547 (95.4%)</td>
</tr>
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<td>D-AMS 1206-62</td>
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</tr>
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<td>656–551 (95.4%)</td>
</tr>
<tr>
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<td>3.26</td>
<td>-18.18</td>
<td>D-AMS 1206-63</td>
<td>598</td>
<td>651–542 (95.4%)</td>
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</table>
investigate the distribution of δ¹³C collagen values using normal probability plots. They suggest that isotopic patterns in their plots, revealing multiple populations, are related to mate exchanges between regions during the Archaic Period (Bousman and Quigg, 2006:133–136). While additional investigations are needed and while our sample size is small, the greater variability in female carbon isotopic values at the Late Prehistoric Coleman site is consistent with a pattern of mate exchange in which females migrated into the area from several different regions and retained some component of their isotopic signature of origin (e.g., Schulting and Richards, 2001). This pattern implies a patrilocal residence system, the most common residence pattern in ethnographic samples of hunter-gatherers (Ember, 1975, 1978; Kelly, 1995:271) and one that has recently been shown to have significant time depth (e.g., Lalueza-Fox et al., 2011). While female carbon isotopic values at Coleman are more variable, the mean values of males and females for carbon are virtually identical for both collagen and carbonate.

Fig. 5. Box plots of δ¹³C collagen (top), δ¹³C carbonate (middle), and δ¹⁵N values from burials 0–10 years of age (Immature) and greater than 10 years of age (Mature) recovered at the Toyah-aged Coleman site. In spite of the smaller samples size (n = 6), the range of values in the immature group of samples are greater than that shown by the 11 adolescent and adult, designated mature, interments. The locations of the medians and the 1st and 3rd quartiles are also different. These different patterns suggest that the diet of individuals in the immature group was substantially different from that of those individuals grouped as mature from the Coleman site.

Fig. 6. δ¹³C collagen and δ¹³C carbonate (‰) for 11 mature (adolescent and adult) individuals from the Coleman site. Also identified are male (n = 6), female (n = 4), and undetermined (n = 1) individuals. Data are listed in Table 2. As in Fig. 2, the C₃ and C₄ marine protein regression lines follow Kellner and Schoeninger (2007) and Froehle et al. (2010).
carbonate derived components. As shown in Fig. 7, a bivariate plot of $\delta^{15}$N values against $\delta^{13}$C values for Coleman individuals by sex, this is not the case with nitrogen. While the clustering within the male values and the dispersion among females discussed previously is clearly visible in this plot, note that the top five $\delta^{15}$N values are all male (Fig. 7). The average $\delta^{15}$N value for Coleman males is $9.4 \pm 0.26^{\%}_{\text{se}}$, more than $0.7^{\%}_{\text{se}}$ higher than the female average of $8.7 \pm 0.56^{\%}_{\text{se}}$. In this limited sample, males have higher $\delta^{15}$N values relative to females.

Higher $\delta^{15}$N values in males are common in the literature, with differences reported for a variety of locations and time periods (e.g., Ambrose et al., 2003; Craig et al., 2009; Richards et al., 2006; Schurr and Powell, 2005). Unlike the Coleman sample, most of these cases are agriculturally based. Possible explanations for these differences in these cases include differential access to foods as a function of status (e.g., Ambrose et al., 2003) as well as physiological differences related to the short-term impacts of pregnancy (e.g., Fuller et al., 2004, 2006; Schurr and Powell, 2005). Reported cases of nitrogen differences between males and females among hunter–gatherer groups, who are assumed to have widespread food sharing practices, are not common in the literature (but see Kusaka et al., 2010). We assume that differences in our sample most likely reflect differential food access rather than any “pregnancy effect,” the short-term impact of which might be difficult to document in collagen (see Nitsch et al., 2010). While it is possible that the nitrogen differences, like the carbon differences noted previously, are related to patterns of mate exchange, it is also possible that the differences reflect a sexual division of labor, with males having greater access to foods with higher $\delta^{15}$N values because of a more direct involvement in their acquisition.

### 3.2. Diachronic patterns

The isotopic patterns reflected in the Coleman burials are a radical departure from previous regional patterns in both nitrogen and carbon. For example, Fig. 8 shows the overall Coleman $\delta^{15}$N values for adults and adolescents relative to earlier periods (see Table 1). The Coleman samples are higher overall than all earlier periods and represent a substantial increase in $\delta^{15}$N relative to the preceding Austin Interval pattern (Fig. 8). Samples below the median Coleman value of $9.2^{\%}_{\text{se}}$ are within the range that would be produced by human consumption of several C3 fauna, including white-tailed deer ($\text{mean } \delta^{15}N = 5.7 \pm 1.2^{\%}_{\text{se}}$, Hard and Katzenberg, 2011) if we assume a $3.5^{\%}_{\text{se}}$ trophic level enrichment. However, human $\delta^{15}$N values above $9.2^{\%}_{\text{se}}$ suggest the use of some high nitrogen dietary options. Given the inland setting and a dependence on C3 protein sources, the Coleman $\delta^{15}$N values in these higher ranges (Fig. 8) would likely require input from fish, reptiles (e.g., soft-shell turtle), or birds that have elevated $\delta^{15}$N levels (see Hard and Katzenberg, 2011: Table 4a, b).

Fig. 9 illustrates the means and standard deviations for carbon isotopes during the Early, Middle, and Late Archaic periods, the Austin Interval, and the Late Prehistoric Toyah Interval mature (adult and adolescent) samples from the Coleman site. Table 2 lists the Coleman data.

#### 3.3. Late Prehistoric (Toyah) adaptation

The Coleman data are also distinct from the previous Toyah sample in that there is no indication among these 11 individuals of a dependence on a C4/CAM protein source such as bison, a pattern possibly reflected in the 41BX677 burial. If the 41BX677 burial represents a distinct isotopic pattern for the region rather than an isolated case, these data suggest the possibility of two different subsistence patterns present in the region during the Late Prehistoric Toyah Interval. One pattern focused on C4/CAM resources, including what is probably C4 feeding bison. A second pattern, represented by the Coleman samples, relied on C3 protein, and included some higher nitrogen faunal resources, as well as moderate use of CAM/C4 plants.
How, or if, these two patterns interacted is not clear. Consistent with the extant direct radiocarbon dates, it is possible that the two different patterns reflect a diachronic shift within the Toyah Interval, with a dependence on C3 protein early and a late shift to C4 protein sources. A shift to predominantly C4 feeding bias has been suggested to occur several hundred years before the Coleman dates (see Dillehay, 1974; Johnson, 1994). This shift, and the associated changes in lithic tool assemblages, defines the Toyah Interval (Kemnotts and Boyd, 2012). Refining our understanding of the timing of this shift can most effectively occur with additional direct dates and isotopic analysis of Toyah Interval bone samples. If additional dates confirm that several different adaptive strategies co-existed during Toyah, interactions between them may have been mediated though various social mechanisms (see Arnn, 2012).

A second possibility is that what we have characterized as two distinct strategies simply reflect different short-term temporal responses to a highly variable Late Prehistoric environment and resource structure (see Dering, 2008; Mauldin et al., 2012), with populations emphasizing C3 or C4 protein resources based on availability. The stable isotopic record from burials may not be the best data set to pursue the possibility of short-term dietary fluctuations, as low bone turnover rates may obscure evidence of oscillations. Precise turnover rates for human rib collagen are not available. However, the turnover rates are much higher (ca. 10−30%) in adolescents (see Hedges et al., 2007), and at least three of the 11 mature Coleman samples are between 10 and 20 years of age. These three samples (burial 12, 13b, 16b) as a group do have a more positive δ13Ccollagen average of −15.7‰ (range −15.1‰ to −16.1‰) when contrasted with the δ13Ccollagen mean of −16.6‰ (range −15.6‰ to 17.5‰) for the remaining mature samples (see Table 2). It may be the case that these three early samples show some evidence of dietary variability within the Coleman mature group. However, contrasting the Coleman patterns in Fig. 6 with the location of the previous Toyah case (41BX677) in Fig. 9 suggests two distinct patterns, with no intermediate cases present. While we cannot eliminate the possibility that these patterns reflect short-term responses to C3/C4 protein availability without additional samples, the lack of intermediate cases is not consistent with that scenario.

Finally, it may be the case that the 41BX677 sample, like the eliminated Toyah sample from 41HY161 (Munoz et al., 2011), reflects an individual from outside the region. As we noted previously, the nitrogen value for the 41BX677 sample (10.7‰) is at the upper end of the expected range if there was a significant dependence on C4 bison, which has an average δ15N of 6.2‰ during this period. In addition, both the δ13Ccollagen (−10.9‰) and δ13Ccarbonate (−5.3‰) values for the 41BX677 sample are the highest of any samples listed in Table 1, suggesting a diet dominated by CAM/C4 plants and C4 animals, with little inputs from C3 resources. As shown by Hard and Katzenberg (2011: Tables 4a and 6) C3 resources dominate the available plant and animal resources in inland settings. For plants, C3 resources include all mast resources (e.g., oak, pecan, hickory, and walnut), all geophytes (e.g., camas, onion, garlic), and most others seed/pod producing plants, including mesquite (Hard and Katzenberg, 2011: Table 6). C4 animal resources include deer, cottontail rabbits, large birds (turkey, geese/swan), most reptiles and inland fish (Hard and Katzenberg, 2011: Table 4a). Low dependence on these resources seems unlikely in inland settings. However, additional Toyah age isotopic samples, as well as consideration of trace elements in all samples (see Burton, 2008), might help to determine if there are non-local individuals present at this time.

4. Conclusions

The present study used carbon and nitrogen stable isotope values generated from bone collagen, and carbon values from carbonate found in apatite, coupled with direct radiocarbon dates on bone, to characterize dietary aspects of burials at the Coleman site, a Late Prehistoric hunter—gatherer cemetery in Central Texas. The patterns investigated here highlight the complexities present in hunter—gatherer adaptations, including potential subsistence diversity, differential resource access based on sex, and patterns of interaction. Isotopic signatures demonstrate that both age and sex affected isotopic variability. Those burials from young individuals showed evidence for both a more variable diet, and higher nitrogen intake when contrasted with results from mature (+10 years of age) interments. These differences are probably related to patterns of breastfeeding and weaning. Focusing on the mature samples, isotopic data also indicate differences between the sexes, through samples sizes are small (n = 10). Coleman females (n = 4) have more varied carbon isotopic signatures, consistent with a pattern of mate exchange in which females migrate into the region. Coleman males have higher average δ15N values when compared to females. This may be related to greater access to higher nitrogen resources, such as fish, at the time of resource acquisition. Overall, adults and adolescents at Coleman have carbonate and collagen carbon values that show a dependence on both C3 and C4/CAM plant resources. Collagen carbon and nitrogen values are consistent with a dependence on C3 animals, such as white-tailed deer, supplemented by fish or other inland resources with high δ15N values. There is no evidence in the Coleman isotopic data for a dependence on bison. Our review of regional isotopic data shows that the Coleman samples reflect a radical divergence in subsistence from patterns shown in earlier isotopic samples as well as from the single, previously analyzed Central Texas Late Prehistoric Toyah sample
from 41BX677. While future research should focus on a re-analysis of the pre-1997 cases listed in Table 1, all of which lack both C:N ratios (see Ambrose and Norr, 1992) and analytical details (see Jardine and Cunjak, 2005), those regional data show a trend of increasing C₃ plant dependence dating back to at least 6900 years BP. The burials from the Coleman site clearly demonstrate a break from that long-term trend with a shift toward a moderate use of CAM/C₄ plants. While this increased use of CAM/C₄ plants is reflected in both the 41BX677 sample and in the Coleman adult and adolescent burials, the 41BX677 sample stands out in two ways. First, the carbonate value suggests a substantial CAM/C₄ plant dependence with little C₃ inputs and second, the collagen carbon value suggests the extensive use of C₄ fauna. The focus on C₄ fauna is in sharp contrast to the Coleman pattern, which has a more C₃ protein signature. These two patterns may reflect temporal differences, different acquisition strategies based on availability, or hint at different subsistence strategies. The 41BX677 individual may also represent an immigrant into the region, one that retains a significant component of their isotopic history of origin. The sex of this individual is not known, but patterns in the Coleman burials hint at movement of females into the region, possibly as a function of mate exchange.

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Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.jas.2012.09.032.

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