***Supplementary Text* for Uno et al., Large mammal diets and paleoecology across the Oldowan-Acheulean transition at Olduvai Gorge, Tanzania from stable isotope and tooth wear analyses**

Tooth enamel stable isotopes and vegetation

Today in eastern Africa nearly all woody vegetation uses the C_3 photosynthetic pathway, or Calvin cycle, and nearly all low elevation (<1500 m) grasses, some sedges (e.g., *Cyperus papyrus*), and select shrubs (e.g., Amaranthaceae) use the C_4 pathway, or Hatch-Slack cycle (Cerling and Harris, 1999; Jones and Milburn, 1978; Livingstone and Clayton, 1980; Sage et al., 2007; Tieszen et al., 1979; Young and Young, 1983). The carbon isotope ratio of C_3 plants is on average around −28 ‰ with an observed range of about −36 to −23 ‰. The wide range in $\delta^{13}C$ values in C_3 plants is controlled by environmental conditions: more negative values are found in closed canopy forests whereas xeric conditions—common in lowland eastern Africa—lead to more positive values (e.g., −26 to −23 ‰). C_4 plants exhibit a narrower range of values, from about −14 to −10 ‰ in eastern Africa (Cerling et al., 2003).

To establish the end-member $\delta^{13}C$ values in tooth enamel ($\delta^{13}C_{enamel}$) for C_3 and C_4 diets, the atmospheric $\delta^{13}C$ value ($\delta^{13}C_{atm}$) and the enrichment in ^{13}C between diet and tooth enamel must be determined. We use the mean value of −6.7 ‰ for the $\delta^{13}C_{atm}$ based on the high resolution planktonic foraminifera record from 1.8 to 1.6 Ma from Tipple et al. (2010). We chose the 1.8 to 1.6 Ma interval because it best matches the age range for Olduvai Bed II ages.

We use the $\delta^{13}C_{atm}$ value of −6.7 ‰ along with biosynthetic fractionation factors for C_3 and C_4 plants to calculate the $\delta^{13}C$ values for C_3 and C_4 plants from 1.8 to 1.6 Ma (**Table S1**). Next, we apply the enrichment factor ($\varepsilon^*_{enamel\text{-}diet}$) of 14.1 ± 0.5 ‰ as determined by Cerling and Harris (1999) for large ungulates to calculate $\delta^{13}C_{enamel}$ endmember values of −11.9 ‰ and + 2.8 ‰ for pure C_3 and C_4 diets, respectively (**Table S1**). The 14.1 ‰ enrichment factor probably doesn't apply to all large ungulates. It likely best describes the enrichment in foregut fermenting bovids. Large mammals with other digestive physiologies that do not match the foregut fermenters in terms of methane production, such as equids, rhinos, proboscideans, suids, and hippos likely have slightly lower $\varepsilon^*_{enamel\text{-}diet}$ values (Passey et al., 2005a). Note that the application of the enrichment factor, $\varepsilon_{enamel\text{-}diet}$ *, is from rearrangement of the following equation,

$$\varepsilon^* = \alpha^* - 1 \quad (1)$$

from Craig (1954), where alpha (α^*) is the apparent fractionation factor between diet and enamel, defined as

$$\alpha^* = (1000+\delta^{13}C_{enamel})/(1000+\delta^{13}C_{diet}) \quad (2)$$

We include these equations here as a review of the mathematically correct way to calculate endmember $\delta^{13}C_{enamel}$ values from $\delta^{13}C_{diet}$ values, rather than simply adding $\varepsilon^*_{enamel\text{-}diet}$ (14.1 ‰) to the $\delta^{13}C_{diet}$ values to determine endmember $\delta^{13}C_{enamel}$ values.

Sampling and Methods

Enamel sampling, pretreatment and isotopic analysis

The protocol for bulk sampling involved drilling along broken enamel surfaces whenever possible, or in some cases a lateral tooth surface, using a Dremel handheld drill with carbide (Brasseler) or diamond grit impregnated (Lasco) bits at low speed (~1000 RPM). No occlusal surfaces were drilled so as to preserve these surfaces for morphological or microwear studies. The sample surface was prepared by abrading the enamel surface with the drill bit. On lateral surfaces, this removed surface adherents and the outermost enamel (*ca.* 100 μm). If present, cementum was drilled away to expose enamel. After visual inspection of the prepared surface with a hand lens, a narrow groove about 1mm deep and 1 to 2 mm wide was drilled parallel to the growth axis of the tooth. Sample groove lengths varied based on tooth geometry (enamel thickness and crown length), but ideally were 10 to 15 mm long to average out potential seasonal variability in diet and water. Sample masses ranged from about 3 to 15 mg.

Three equid molars were serially sampled along the growth axis of the tooth to generate intratooth isotope profiles. Cementum was cleared away using the Dremel to expose a window of the fossil enamel. The exposed area was ~5mm wide and ran the length of the tooth crown (50 to 60 mm). Samples were drilled every 3 mm along the growth axis of the tooth, with sample grooves oriented normal to the growth axis. Sample grooves were $\leq$ 1 mm deep, ~1 mm wide, and 3 to 6 mm long. Sample masses were about 3-5 mg. Enamel powders from bulk and intratooth profile samples were pretreated at LDEO prior to stable isotope analysis with a weak oxidizer followed by buffered acid. Samples were treated with 3% NaOCl (bleach) for 30 minutes in 1.7 ml centrifuge tubes that were stirred every 10 minutes on a vortex mixer. After the reaction period, samples were centrifuged and the supernatant was removed. Each sample was then rinsed three times with distilled water. The rinse procedure involved adding de-ionized (DI) water, stirring on the vortex mixer, then centrifuging the sample and removing the supernatant. Next, samples were treated with 0.1 M Na-acetate buffered acetic acid for 30 minutes, as above. Following three distilled water rinses, they were loosely covered and dried overnight in a fume hood.

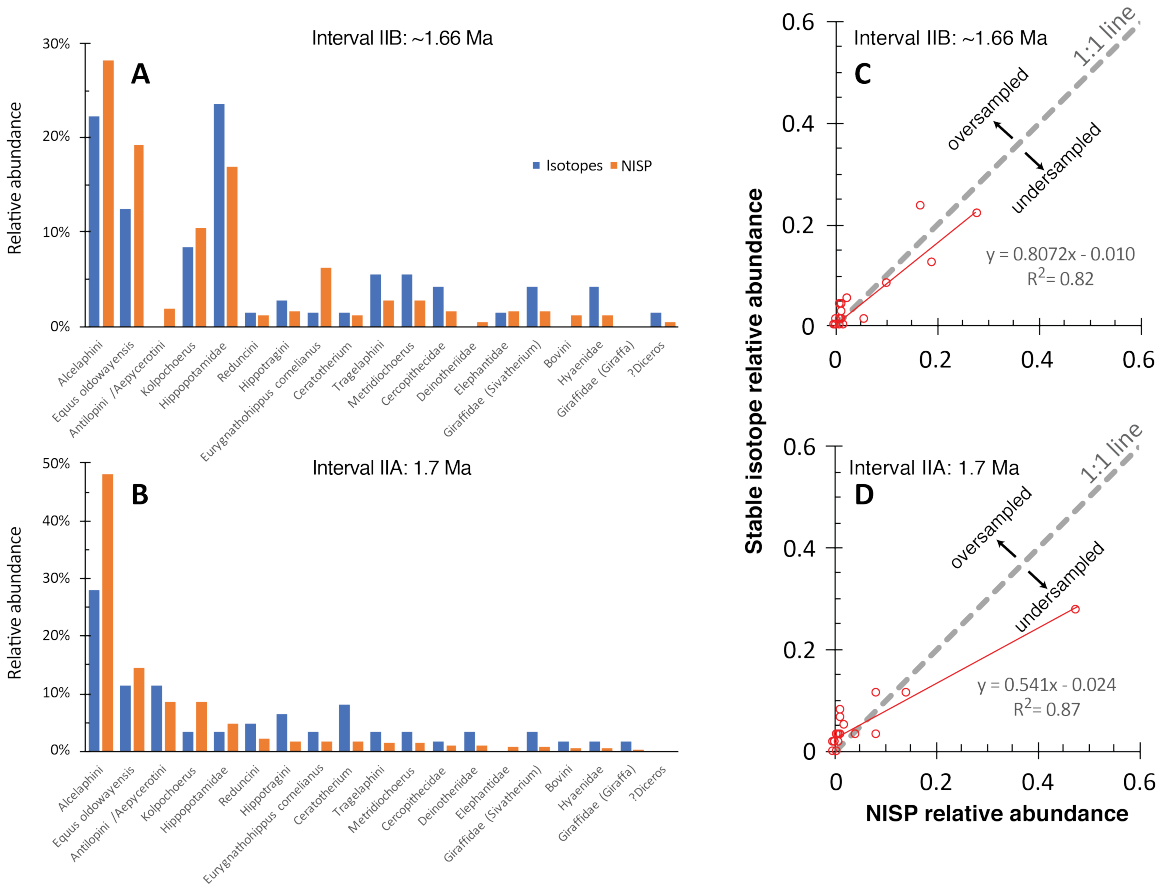
Approximately 300 to 600 μg of powdered enamel sample were weighed out into a silver capsules, along with NBS-19 standard (20-50 μg). Samples and standards were roasted in vacuo for 2 hours at 60° C to remove adsorbed water, then transferred to round bottom,

glass reaction vials. A strand of silver wool was added to each vial to oxidize any SO_2 produced in the reaction. Sample vials were loaded immediately into a (Thermo) Kiel IV device or transferred into an oven held at 60° C until analyzed.
Fossil enamel samples were reacted with phosphoric acid at 70° C for 10 minutes in vacuo on a Kiel IV device. Exsolved gases were cryogenically transferred to a microvolume held at −170° C, and then transferred to second microvolume with the first held at −70 °C to remove water. The purified sample CO_2 was inlet into a dual-inlet Delta V Plus isotope ratio mass spectrometer. Carbon and oxygen isotope ratios were corrected using international and internal standards. The standard deviation of NBS-19 throughout sample runs was ~0.05 ‰ for $\delta^{13}C$ and <0.10 ‰ for $\delta^{18}O$. Oxygen isotope values were converted from CO_2 (gas) to fossil enamel (mineral) values using a fractionation factor of 1.00799, which was calculated using the temperature dependent equation for fossil enamel (Eq. 4) in Passey et al. (2007).

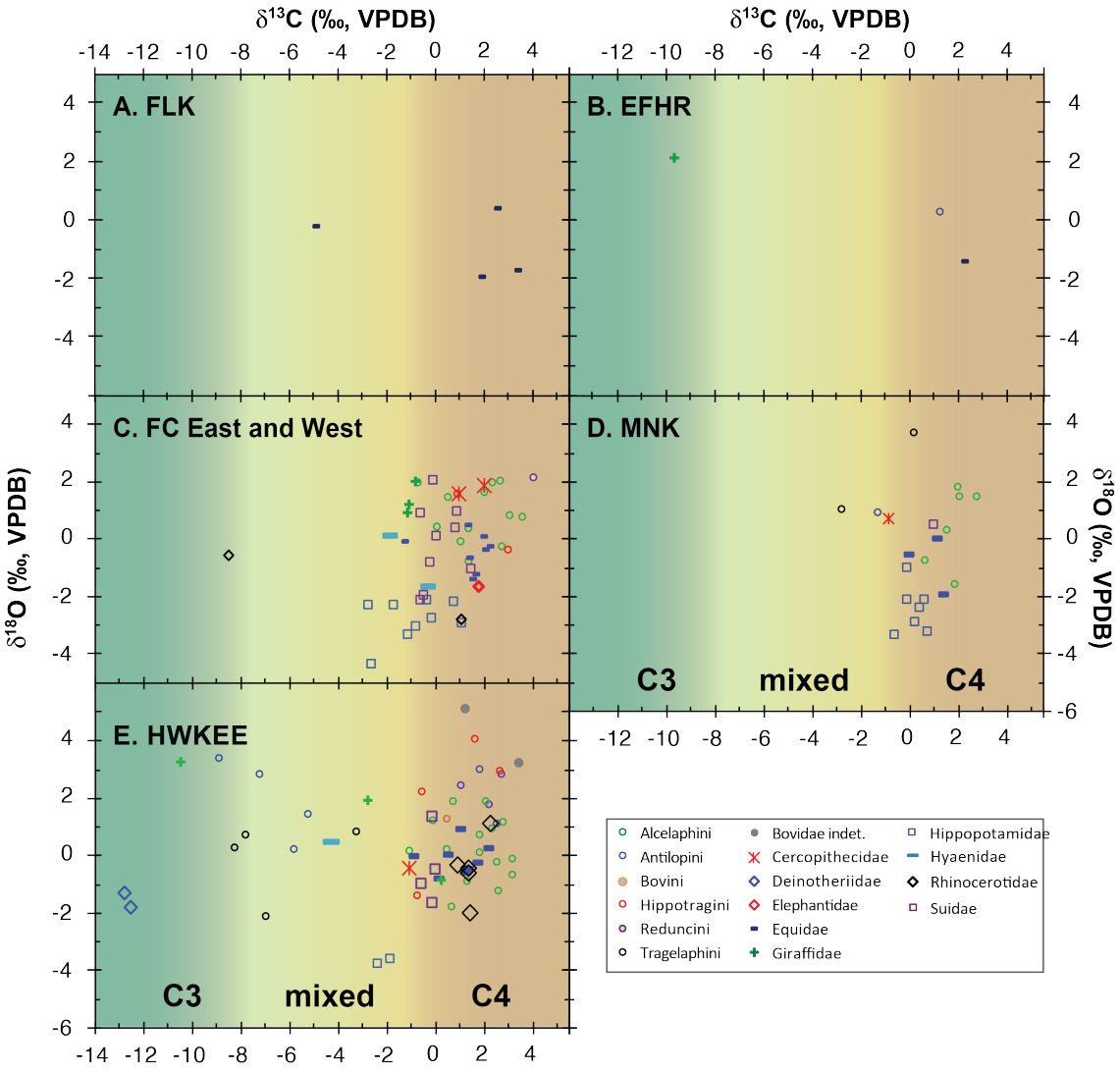
Intratooth stable isotope profiles and inverse modeling procedure

The inverse model requires input parameters related to tooth formation, isotope sampling geometry, and isotope analysis. Input parameters relevant to tooth enamel formation, or amelogenesis, include initial enamel density (f_i), enamel appositional length (l_a) and maturation length (l_m) (Passey and Cerling, 2002). We use parameters measured on *Equus ferus przewalskii* by (Blumenthal et al., 2014) as estimates for *Equus oldowayensis* molars used here, where f_i = 22%, l_a = 6 mm, l_m = 28 mm. Sample input variables include distance between samples (3 ± 0.5 mm) and depth (0.7 mm ± 0.2 mm). The final input variables are the measured carbon and oxygen isotope data and their associated uncertainties, which were smoothed with a 3-point weighted mean (0.25:0.5:0.25). A measured error term, E_{meas}, is computed from measurement uncertainties in isotope values and sample measurements. This term ultimately governs model sensitivity it is used to determine an appropriate damping factor (ε^2). The model requires selection of a damping factor that minimizes the difference between E_{meas} and the prediction error (E_{pred}). A detailed description of additional model parameters and the regularization method is given in Passey et al. (2005b). Model code was downloaded and adapted from Passey et al. (2005). The inverse model assumes constant growth rate. However, it is widely accepted that ungulate cheek teeth have non-linear growth rates, particularly towards the end of crown extension (Bendrey et al., 2015; Zazzo et al., 2012).

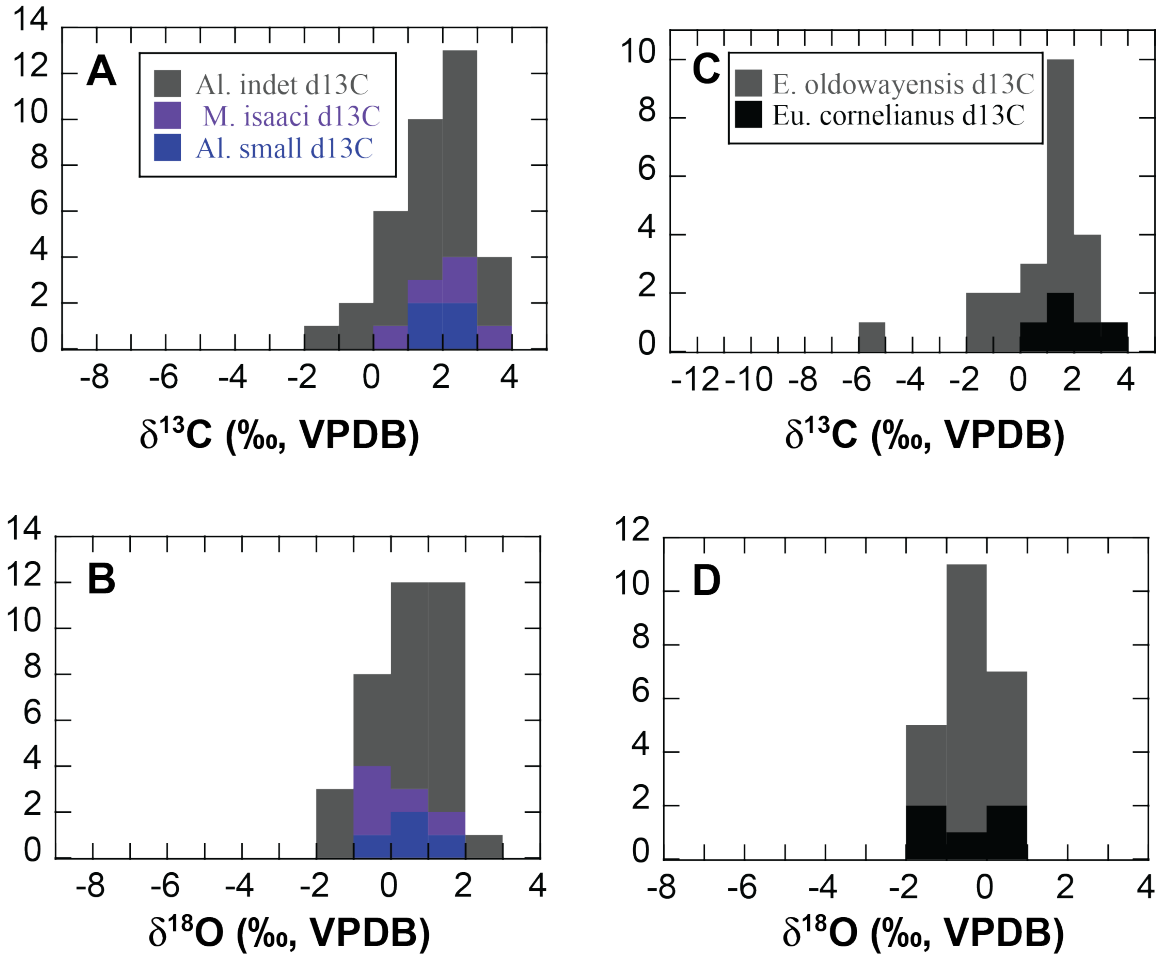
Supplementary Figures



SOM Figure S1. Bar graphs (A, B) and cross plots (C, D) of relative taxonomic abundances of isotope vs. NISP data sets for interval IIA and IIB.



SOM Figure S2. Cross plots of tooth enamel carbon and oxygen isotopes from all sites investigated in this study: A) FLK West, B) EF- HR, C) FC East and West, D) MNK, and E) HWK EE. Shaded regions indicate C_3-dominated (green), mixed C_3-C_4 (yellow), and C_4-dominated (orange) diets based on $\delta^{13}C$ values. Data are plotted at the family level except for bovids, which are plotted by tribe.



SOM Figure S3. Histograms of A) $\delta^{13}C$ and B) $\delta^{18}O$ for three forms of Alcelaphini (*Megalotragus issaci*, Alcelaphini small sp., and Alcelaphini sp. indet.) from Bed II. Neither isotope shows niche partitioning among the Alcelaphini. Panels C) and D) show $\delta^{13}C$ and $\delta^{18}O$ for the two Equidae species in Bed II, *Equus oldowayensis* and *Eurygnathohippus cornelianus*. The latter tend to have more positive $\delta^{13}C$ values that indicate a greater proportion of C_4 vegetation in their diet although sample size is small (n =5). There is no difference in $\delta^{18}O$ values.

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