1. Introduction

Under present atmospheric CO₂ concentrations, C₄ plants are minor or absent in cool, high-elevation environments (Ehleringer et al., 1997; Collatz et al., 1998; Sage et al., 1999; Edwards and Smith, 2010). The modern distribution of C₄ plants is likely a result of preferential adoption of C₄ photosynthesis by clades adapted to warm, mesophytic conditions (>22 °C mean growing season temperature, ca 1200 mm yr⁻¹ rainfall), where the C₄ photosynthetic pathway imparts a competitive advantage (Collatz et al., 1998; Edwards and Smith, 2010). Ecosystem models incorporate a 22 °C minimum mean monthly temperature requirement for C₄ plants in addition to a 25 mm month⁻¹ minimum precipitation threshold for all plants (Collatz et al., 1998; Still et al., 2003). Therefore, these models reconstruct few C₄ plants at latitudes >50° or elevations >2500 m asl (e.g., Fig. 1), which may result in underestimates of global primary production (Still et al., 2003).

The existence of >50 C₄ plant species on the Tibetan Plateau and other high-elevation regions challenges this 22 °C threshold criterion (Fig. 1; Tieszen et al., 1979; Cavagnaro, 1988; Wang et al.,...
and heterogenous, ranging from 0 to 80% in Southeast Asia, the Philippines, and Indonesia (Z. Liu et al., 2009), showing no C4 grasses on the Tibetan Plateau, but consistent minor C4 grass abundance in Southeast Asia, the Philippines, and Indonesia (white dots; H, Huanxian; W, Weinan (Liu et al., 2005); X, Xunyi (Zhang et al., 2003); M, Mangshan (Peterse et al., 2011)). C4 plants abundance is variable and heterogeneous, ranging from 0 to 80% in Southeast Asia, the Philippines, and Indonesia (Stil1 et al., 2003). Inset: C4 grass abundance at 9 ka simulated with the CCSM3 transient simulation (Z. Liu et al., 2009), showing no C4 grasses on the Tibetan Plateau, but consistent minor C4 grass abundance in Southeast Asia, the Philippines, and Indonesia (white; see Fig. 35 for more time slices).

The Tibetan Plateau and other high-elevation regions experience amplified responses to global climatic change due to cloud-and-snow-albedo feedbacks (X. Liu et al., 2009; Diffenbaugh et al., 2012). High-elevation plant ecosystems must adapt to amplified climatic change under already harsh (cold, arid) conditions, and are therefore bellwethers of climatic and environmental change. Re-constructions of plant ecosystem variability during climate regimes different than today elucidate mechanisms that cause plant ecosystem change, and provide information to improve ecosystem model accuracy in sensitive high-elevation regions.

We present a sub-millennial-resolution 32-ka-long reconstruction of C4 plant abundance on the northeastern Tibetan Plateau based on δ13C of leaf wax C28 n-acids (δ13CC28) from Lake Qinghai (37°N, 100°E; 3194 m above sea level; Fig. 1). We augment our findings with a 16-ka-long pollen record from Lake Qinghai (Ji et al., 2005), a detailed interpretation of the 32-ka-long pollen record is in preparation (S. Xue, personal communication). Our work builds on research by An et al. (2012), who developed an age model and initial climate reconstructions for these drill cores are described in previous work (An et al., 2012). Free lipids were extracted from freeze-dried samples with an Accelerated Solvent Extractor 200 (Dionex) using dichloromethane (DCM):methanol 9:1 (v:v). N-alkanoic acids were prepared by first separating the TLEs on solid phase Aminopropyl silica gel flash columns using three column volumes of DCM:Isopropanol 2:1 (v:v) and 4% acetic acid in diethyl ether. The acid fraction was methylated in anhydrous 5% HCl in methanol at 60 °C overnight. Hydroxyl-carboxylic acids were removed from the methylated acid fractions using silica gel flash columns with DCM as the eluant, after eluting apolar compounds with hexane.

1. Determining past C4 plant abundance

Different enzyme-mediated reactions in the C3 and C4 photosynthetic pathways impart distinct carbon isotope compositions on plant biomass: C4 plants are ca 15‰ enriched in 13C relative to C3 plants (O’Leary, 1981). These distinct δ13C compositions are also reflected in leaf waxes (Chikaraishi and Naraoka, 2007; Rao et al., 2008). Leaf waxes are n-alkyl lipids that form a protective layer on leaves; plants in arid regions produce particularly high abundances of leaf waxes (Shepherd and Griffiths, 2006). Leaf waxes are abraded from the leaf surface and carried to a lake basin by wind and/or water and are preserved well in Quaternary lake sediments (Castañeda and Schouten, 2011). We use leaf wax isotopic end-member values for C3 and C4 plants, discussed below, to calculate C4 plant abundance.

2. Methods

2.1. Lipid biomarker extraction and purification

131 samples were obtained from ICPD drill cores LQDP05 1A and LQDP05 1F for organic analysis. The stratigraphy, age model, and initial climate reconstructions for these drill cores are described in previous work (An et al., 2012). Free lipids were extracted from freeze-dried samples with an Accelerated Solvent Extractor 200 (Dionex) using dichloromethane (DCM):methanol 9:1 (v:v). N-alkanoic acids were prepared by first separating the TLEs on solid phase Aminopropyl silica gel flash columns using three column volumes of DCM:Isopropanol 2:1 (v:v) and 4% acetic acid in diethyl ether. The acid fraction was methylated in anhydrous 5% HCl in methanol at 60 °C overnight. Hydroxyl-carboxylic acids were removed from the methylated acid fractions using silica gel flash columns with DCM as the eluant, after eluting apolar compounds with hexane.

2.2. Carbon isotope analysis

The carbon isotope ratio of fatty acid methyl esters (FAMES) was determined on an HP 6890 gas chromatograph interfaced to a Thermo Finnigan Delta V plus isotope ratio mass spectrometer through a combustion reactor at Brown University. Isotope values...
are expressed in per mille (‰) relative to Vienna Pee Dee Belemnite (VPDB). The FAME δ13C values were corrected for the isotopic contribution of the carbon in the methyl group added during methylation. The analytical error, calculated using the results of duplicate analyses of each extract, is 0.4‰ (1σ) and an external FAME standard, injected once after every sixth sample injection, had an analytical error of 0.7‰ (1σ).

### 2.3. Transient simulation

The transient simulation of the past 21,000 years was carried out using the low-resolution version (T31×3) of the Community Climate System Model, version 3 (CCSM3; Z. Liu et al., 2009; He, 2011; Liu et al., 2012). CCSM3 is a global, ocean-atmosphere-sea ice-land surface climate model coupled without flux corrections. The atmospheric component is the Community Atmosphere Model version 3 at a horizontal resolution of T31 (approximately 3.75° in latitude and longitude) and 26 hybrid vertical levels. The land model, which predicts plant functional types using a dynamic vegetation scheme, uses the same grid as the atmospheric component. The ocean model is an implementation of the Parallel Ocean Program model with longitudinal resolution of 3.6° and variable latitudinal resolution, with finer resolution in the North Atlantic and tropics (~0.9°). The vertical resolution is 25 levels extending to a depth of 5.5 km. The dynamic-thermodynamic sea ice model uses the same horizontal grid and land mask as the ocean component. The T31×3 present-day control simulation reproduces the major features of global climate, with improved ocean circulation compared to earlier versions of this model (Yeager et al., 2006). Results from the higher-resolution (T42×1) version of CCSM3 for Last Glacial Maximum and middle Holocene conditions capture important paleoclimate patterns documented by proxy records (Otto-Blesner et al., 2006).

Detailed information about the forcings used in the transient experiment can be found in He (2011), Liu et al. (2012), and Z. Liu et al. (2009). Briefly, orbital forcing varied transiently (Berger, 1978), and greenhouse gas (CO2, CH4, and N2O) concentrations were based on ice core measurements (Joos and Spahni, 2008). Continental ice sheets were based on the ICE-5G reconstruction (Peltier, 2004) and altered approximately once per thousand years. Meltwater forcing (MWF) prior to 14.5 ka was the DGL-A scheme (Z. Liu et al., 2009), which includes an abrupt termination at the Bolling-Allerod. Following this, major periods of MWF were prescribed from 14.4 to 13.9 ka to the North Atlantic and Southern Ocean and from 12.9 to 11.7 ka and from 9.0 to 8.2 ka to the North Atlantic (He, 2011; Liu et al., 2012).

To generate time series of climate variables from the transient simulation for the Lake Qinghai area, we averaged the four closest grid cells to the lake (Fig. 2). This choice was made to average out random fluctuations from individual grid cells while accounting for the relatively low resolution of the model and the large topographic gradients in this region. The transient evolution of temperature and precipitation for these four grid cells is representative of monsoonal Asia as a whole (Figs. 2, S2 and S3).

### 2.4. CMIP5 model results

To provide an estimate of conditions that the Tibetan Plateau plant ecosystem will be experiencing in ca 2050 AD, we extracted temperature and pCO2 data for the Tibetan Plateau from Coupled Model Intercomparison Project Phase 5 (CMIP5) model projections. Temperature and precipitation anomalies for the Lake Qinghai area at ca 2050 AD were calculated from CMIP5 model projections, using the Representative Concentration Pathways (RCP) 2.6 and 8.5 (years 2040–2059 AD) relative to historical climate simulations (years 1986–2005 AD). We used all models in the CMIP5 archive as of May 2013 that provided the information necessary for our analysis (Table S1). For models providing ensembles for the RCPs, we analyzed just one ensemble member given the expectation that differences among the models are typically larger than differences between ensemble members. The grid resolutions vary among the models and to ensure that we consistently averaged the same spatial area we first bilinearly interpolated results from all models to the CCSM4 grid. Then, we calculated the area average for a 3° × 3° box centered on the location of Lake Qinghai (37° N, 100° E).

### 3. C28 n-acid carbon isotopes as a proxy for C4 plant abundance

#### 3.1. Leaf wax sources

The most abundant n-acid chain lengths in Lake Qinghai sediments are C26 and C28. Terrestrial plants in the modern Lake Qinghai catchment produce mainly mid- to long-chain n-acids, with a peak at C24. C26 and C28 n-acids compose ca 10% each of total terrestrial plant leaf wax production (Wang and Liu, 2012). In contrast, aquatic plants produce mainly C16 n-acids. C26 is <10%, and C28 is <5% of total aquatic plant leaf wax production (Wang and
Aquatic pollen is a minor component (<1% average) of the total pollen flux throughout the entire 32-ka record, except from 20.5 to 15.5 ka, when aquatic pollen averages 5%, and briefly peaks at 65% at ca 18 ka (Fig. 3; Ji et al., 2005; S. Xue has generated aquatic pollen data for the past 32 ka in Lake Qinghai and reconstructs pollen abundance up to 65% at 20.5, 18, and 15.5 ka, personal communication). Terrestrial plants are therefore the dominant source of long-chain n-acids, with the possible exception of brief intervals between 20.5 and 15.5 ka. We report δ13C of the C28 n-acid because aquatic plants produce fewer C28 n-acids, and are less likely to contaminate the terrestrial signal.

3.2. Carbon isotope end members

In order to convert δ13C of wax to C4 plant abundance, we must determine reasonable C3 and C4 plant C28 n-acid δ13C values for the
The δ13C of C3 fatty acid in C3 trees and herbs from Thailand and Japan, where mean annual precipitation is ca 1500 mm, is −36.9 ± 1.7‰ (Chikaraishi and Naraoka, 2007). When C3 plants are drought-stressed, as is the case for plants on the northeastern Tibetan Plateau, their biomass becomes enriched in 13C by up to 3.0‰ relative to non-drought-stressed C3 plants (Passey et al., 2002). A compilation of carbon isotope fractionation during CO2 uptake and fixation (Δleaf) by trees throughout the world shows that trees in dry regions have lower Δleaf, and therefore higher δ13C, than trees in wet regions, even within a single genus (Diefendorf et al., 2010). C3 plants at higher elevations also have lower Δleaf, perhaps due to decreased atmospheric pressure (Li et al., 2007; Diefendorf et al., 2010; Zhou et al., 2011). Assuming that this relationship applies to grasses and herbs as well as trees, we apply an empirically derived equation for leaf δ13C enrichment: Δleaf = 4.20(±0.26) log10(MAP) − 0.06(±0.01)*Δlatitude + 9.31(±0.30). MAP = mean annual precipitation in mm. (Diefendorf et al., 2010) using an average catchment MAP of 370 mm and an elevation of 3500 m. The combination of aridity and high-elevation at Lake Qinghai would cause up to 5.3 ± 2.2‰ enrichment of C3 plant δ13Cwax compared to plants in Japan and Thailand (Chikaraishi and Naraoka, 2007; Diefendorf et al., 2010). The modern landscape around Lake Qinghai is dominated by C3 plants, and the minimum late Holocene δ13Cwax, in Lake Qinghai sediments is −32.9‰. This is 4.2‰ enriched in 13C relative to δ13Cwax of C3 trees and herbs in Japan and Thailand, close to the predicted 5.3‰ enrichment. We therefore use −33 ± 2‰ for the C3 plant δ13Cwax end member at Lake Qinghai.

The glacial period was drier than the Holocene at Lake Qinghai (Colman et al., 2007; An et al., 2012), likely causing C3 plant δ13C to be more enriched. Although we do not have quantitative estimates of precipitation amount, a site with 100 mm growing season precipitation would contain C3 plants 2‰ enriched relative to modern C3 plants at Lake Qinghai using the equation above (Diefendorf et al., 2010). Thus, the glacial C3 plant δ13Cwax end member value may have been closer to −31‰, which would decrease our calculated C4 plant abundance during the glacial period by 5–10% (Fig. 3B). Note that the −31‰ C3 plant δ13Cwax end member results in negative C4 plant abundance during the late Holocene, and is therefore likely only applicable to the glacial period.

3.2.2. C4 plant δ13Cwax end member

The δ13C of C28 fatty acid in C4 plants from Thailand and Japan, where mean annual precipitation is ca 1500 mm, is −21.4 ± 2.1‰ (Chikaraishi and Naraoka, 2007). Unlike C3 plants, drought-stressed C4 plants do not exhibit changes in biomass δ13C (Passey et al., 2002). Drier conditions on the Tibetan Plateau, or changes in precipitation amount between the glacial and Holocene periods likely do not affect the C4 plant δ13Cwax end member. There is evidence that C4 Chenopodiaceae s.s., a dominant form of C4 plants in central Asian deserts, have lower Δleaf at high-elevations (0.1‰ per 100 m; Pyankov et al., 2000; Van de Water et al., 2002). At 3500 m on the northeastern Tibetan Plateau, the C4 plant δ13Cwax end member value would be −18‰, ca 3‰ enriched in 13C relative to C4 plant δ13Cwax at low elevations (Chikaraishi and Naraoka, 2007).

3.2.3. Aquatic plant δ13Cwax end member at Lake Qinghai

Aquatic plants are intermittently abundant in the Lake Qinghai record (Fig. 3; Ji et al., 2005). Aquatic pollen in the Lake Qinghai record is mainly Myriophyllum sp. with minor amounts of Potamogeton sp. and Sparganium sp. Carbon isotopes of bulk Potamogeton sp. material from Lake Qinghai are −17.8 to −15.9‰ (W. Liu et al., 2013). Cladophora sp., aquatic photosynthetic plant that do not produce pollen and produce only a small relative proportion of long chain n-acids, have bulk carbon isotope values of −33.6 to −28.6‰ (Wang and Liu, 2012; W. Liu et al., 2013). C28 n-acid carbon isotopes tend to be depleted relative to bulk values, and therefore the bulk values cited here are maxima for aquatic δ13Cwax end members. The studies of plants at Lake Qinghai do not provide information regarding the concentration of n-acids produced by aquatic and terrestrial plants, although other research demonstrates that terrestrial plants produce more n-acids than aquatic plants (Gao et al., 2011). Aquatic plants are therefore unlikely to have contributed significant amounts of n-acids to the Lake Qinghai sediments throughout most of the record. One exception to this is the interval at 18 ka, when aquatic pollen is −65‰ (S. Xue, personal communication). Depleted δ13Cwax values, ca −34‰ also occur in this interval, and may be a result of increased aquatic plant productivity, particularly Cladophora sp., which has depleted bulk δ13C values.

3.3. C4 plant abundance, error estimates, and alternate interpretations

Lake Qinghai δ13Cwax ranges from −32.9 to −21.9‰ (Fig. 3). δ13Cwax was most 13C-depleted from 32 to 21 ka and 6 to 0 ka and most 13C-enriched during the Lateglacial and early Holocene (13.7–8.3 ka; Fig. 3). This 10‰ range is most likely a result of changing C3 and C4 plant biomass in the Lake Qinghai catchment. We therefore use the isotopic end members, −33 ± 2‰ for C3 plants and a range from −21 to −18‰ for C4 plants, and the Lake Qinghai δ13Cwax record to calculate C4 plant abundance from 32 ka to the Late Holocene (Fig. 3B; black line end members: −33‰ for C3 and −18‰ for C4 plants). C3 plants were dominant (>80%) during the glacial period, with two intervals of abrupt C4 plant expansion. During the Lateglacial and early Holocene, C4 plants were abundant (ca 50%) in the Lake Qinghai catchment. The modern C3-dominated ecosystem developed ca 6 ka. Due to uncertainty in our end member estimates, we present a range of possible C4 plant abundance (Fig. 3B).

We stress that aridity-driven changes in C1 plant 13CO2 discrimination would cause δ13Cwax variations no greater than ca 3‰. If aridity were the main cause of changes in Lake Qinghai δ13Cwax, the dry late Holocene would be more enriched in 13C than the wet early Holocene (Passsey et al., 2002; Wang et al., 2003; Diefendorf et al., 2010; An et al., 2012; Dietze et al., 2013; Hudson and Quade, 2013). This is opposite the trend that we observe (Fig. 3). Furthermore, CAM plants, such as Crassulaceae and Isoetes, are rare in this part of the Tibetan Plateau, in both modern surveys and in the pollen record, and so would likely not influence δ13Cwax (Chen and Peng, 1993; Ji et al., 2005; Herzschuh et al., 2010). Thus, we interpret the Lake Qinghai δ13Cwax record as indicating changes in terrestrial plant ecology, specifically changes in C3 and C4 plant biomass.

4. Discussion

We propose that two distinct sets of environmental conditions caused C4 plants to flourish in the Lake Qinghai catchment: 1. From 21 to 15 ka: C3 plants dominated, but arid conditions caused Lake Qinghai level to drop, exposing the lake bed, which was colonized by early-succession, cold- and salt-tolerant C4 Chenopodiaceae s.s. and, 2. During the Lateglacial and early Holocene: moderate pCO2 and warm, wet summers caused C4 Chenopodiaceae s.s. and Poaceae to flourish throughout northeastern Tibet (Fig. 4). We use these boundary conditions to incorporate precipitation into the
model by Ehleringer et al. (1997) that predicts the CO2 and temperature conditions that favor C3 or C4 plants (Fig. 5, curves representative for the Tibetan Plateau). The boundary conditions between C3 and C4 plants calculated by Ehleringer et al. (1997) cannot explain the changes in C4 plant abundance that we reconstruct at Lake Qinghai. We therefore constructed a schematic representation of the effects of precipitation (Fig. 5). Therein, isohyets represent dry (ca 100 mm/growing season; Last Glacial), wet (ca 330 mm/growing season; early Holocene), and moderate precipitation conditions (ca 220 mm/growing season; modern) and serve as boundaries between conditions that favor C3 and C4 plants. The isohyets were drawn to have slopes similar to those calculated by Ehleringer et al. (1997), and were positioned to represent a best fit to our %C4 plant results. Our results support recent findings that the conditions that favor C4 plant productivity shift not only in response to temperature and pCO2, but also in response to growing season precipitation (Edwards and Still, 2008; Edwards and Smith, 2010).

4.1. Plant ecosystems in the Lake Qinghai catchment during the glacial period

During the glacial period (32–14 ka), Lake Qinghai δ13Cwax ranged from −34.0 to −22.5‰, indicating C3 plants were dominant (Fig. 3). Overall primary productivity was low from 32 to 15 ka, as indicated by low pollen counts (Fig. 3) and multiple bulk organic geochemical proxies (X. Liu et al., 2013). Mean annual temperature reconstructed on the Chinese Loess Plateau (CLP; Peterse et al., 2011) and summer temperature at Lake Qinghai in the transient simulation of the CCSM3 (Z. Liu et al., 2009) were 3–6°C lower than present (Figs. 2–4, and S2; see SI). Precipitation, reconstructed using carbonate in Lake Qinghai sediments and simulated by
layers that date to ca 19 ka indicate low levels at Lake Qinghai (Colman et al., 2007). Aquatic plant pollen is not readily transported over long distances (Cook, 1988), so the presence of abundant aquatic plant pollen at the drill site from 21 to 15 ka suggests that aquatic plants were growing near the drill site (Fig. 3; Ji et al., 2005; S. Xue, personal communication). These plants generally root in water that is <7 m deep (Sheldon and Boyle, 1977), so this provides further evidence for lower Lake Qinghai levels. Other records at Lake Qinghai and elsewhere in Tibet indicate dry conditions and lower lake levels during the glacial period (Herschuh et al., 2010; Mügler et al., 2010; An et al., 2012; X. Liu et al., 2013; Y. Wang et al., 2013). The Qinghai lake bed did not desiccate completely, however, as sedimentation was continuous with no evidence for subaerial exposure at the drill site, located at the depocenter of the lake (An et al., 2012).

Glacier maxima in the Dalijia Mountains, 220 km southeast of Lake Qinghai, at 21.8 ± 1.5 ka and 17.3 ± 1.5 ka, are evidence of maximum cold conditions, corresponding to the lowest temperatures reconstructed on the CLP (Fig. 4; Pétursson et al., 2011; J. Wang et al., 2013). Despite cold and arid conditions, low pCO2 would have favored C4 plants (Figs. 4 and 5; Jouzel et al., 2007). Chenopodiaceae s.s. are early succession plants, and C4 photosynthesis is more abundant in Chenopodiaceae s.s. lineages that exhibit salt-tolerance and succulence, both of which would have been critical adaptations to surviving the arid glacial period on the Tibetan Plateau (Pyankov et al., 2000; Wang, 2007; Kadereit et al., 2012). Chenopodiaceae s.s. pollen was abundant (20–40%) during the glacial δ13Cwax peaks (Fig. 3; Ji et al., 2005; S. Xue, personal communication). We hypothesize that C4 Chenopodiaceae s.s. colonized the exposed, salty lake bed when Lake Qinghai levels dropped during the glacial period. This increase in C4 Chenopodiaceae s.s. would have been a local phenomenon, constrained to exposed lake beds (Fig. S1). This hypothesis can be tested in future studies: other desiccated lake beds may also show intermittent increases in δ13Cwax, whereas terrestrial sediment deposits (e.g., loess) on the Tibetan Plateau would not contain evidence for these δ13Cwax increases.

4.2. Transition to the Lateglacial period

At ca 16 ka, the westerly winds strengthened, likely in response to Heinrich Event 1 (H1; Sun et al., 2011; An et al., 2012). From 16 to 14.5 ka, tree pollen increased in abundance and δ13Cwax decreased, indicating that C3 plants dominated during H1 (Fig. 3; Ji et al., 2005). Immediately following H1, δ13Cwax increased, but returned to consistently low values (<32‰) from 14.5 to 13.7 ka. Artemisia represented 80% of pollen from 14.5 to 13.7 ka, which would explain the strong C3 plant signal in δ13Cwax. Other proxies indicate increasing temperature, increasing summer precipitation, and increasing seasonal insolation contrast during this time period (Figs. 3 and 4), so it is unclear why the δ13Cwax and pollen suggest stable ecological conditions from 14.5 to 13.7 ka.

4.3. C4 plants were abundant on the Tibetan Plateau during the Lateglacial and early Holocene

During the Lateglacial and early Holocene (13.7–8.3 ka), Lake Qinghai δ13Cwax ranged from −29.5 to −21.8‰, corresponding to an average of 50% C4 plants (Fig. 3). We hypothesize that this increase in C4 plant abundance was caused by a combination of environmental changes: high seasonal insolation contrast, moderate pCO2 and warm conditions, and greater summer precipitation.

Winter insolation was low and summer insolation was high at Lake Qinghai during the early Holocene, resulting in a seasonal insolation contrast of ca 350 W m−2 (Fig. S4; Laskar et al., 2004).
Cold winters and springs likely inhibited C3 plant growth, whereas short, hot, wet summers favored C4 plant productivity (Ehleringer et al., 1997; Pyankov et al., 2000). Global pCO$_2$ was 240–260 ppmv (Jouzel et al., 2007), values that favor C4 photosynthesis only at higher temperatures (Fig. 5; Ehleringer et al., 1997). Late-glacial and early Holocene temperature was 1–3 °C higher than present, according to a pollen record from Lake Luanhaizi in the nearby Qilian mountains, the CCSM3 transient simulation, and the temperature reconstruction on the CLP (Figs. 2 and 4; Z. Liu et al., 2009; Herzschuh et al., 2010; Peterse et al., 2011). Precipitation increased from 13.7 to 12 ka and peaked at 12 to 8 ka, according to δ$^{18}$Ostracode from Lake Qinghai, the pollen record from Lake Luanhaizi, and the CCSM3 transient simulation (Figs. 2–4; Liu et al., 2007; Z. Liu et al., 2009; Herzschuh et al., 2010; An et al., 2012). Higher lake levels, suggesting wetter conditions, existed throughout Tibet during the early Holocene, including at Lake Donggi Cona, 200 km southwest of Lake Qinghai (Dietze et al., 2013; Hudson and Quade, 2013).

Warmer conditions were favorable for primary production, as indicated by increased pollen flux at ca. 14.8 ka and multiple bulk organic geochemical proxies from Lake Qinghai (Fig. 3; An et al., 2012; X. Liu et al., 2013). Poaceae and Chenopodiaceae s.s. each contributed to 20% of total pollen by 12–8 ka, with Poaceae remaining constant during the late Holocene, and Chenopodiaceae s.s. pollen increased slightly during the Younger Dryas (Fig. 3; Ji et al., 2005). Tree pollen decreased to 10% during the Younger Dryas, and increased abruptly to 20% at the end of the Younger Dryas, when total pollen counts also increased. Although the existing data do not distinguish C3 from C4 plant pollen, the 10$^{13}$C$_{wax}$ increase in δ$^{13}$C$_{wax}$ cannot be explained by changes in C3 plant water stress, and so C4 plants must have flourished during the Lateglacial and early Holocene. Relatively warm, wet conditions, occurring at a time of maximum seasonal contrast during the Lateglacial and early Holocene, would be favorable to both C3 Poaceae and C4 Chenopodiaceae s.s. This increase in C4 plant abundance may have resulted from a few extant taxa that proliferated on the Qinghai–Tibetan Plateau during this period, or may have resulted from C4 plant species shifting to higher elevations as climate became more conducive to their growth (Fig. S1). In either case, the Lateglacial and early Holocene increase in C4 plant abundance was driven by regional and global environmental conditions, and therefore C4 plants were likely dominant throughout the region around Lake Qinghai.

4.4. Mid-Holocene development of a C$_3$-plant-dominated ecosystem

The mid-Holocene δ$^{13}$C$_{wax}$ decrease is mirrored by decreasing bulk organic matter δ$^{13}$C (X. Liu et al., 2013). Aquatic plants exert only a minimal influence on δ$^{13}$C$_{wax}$, so we can more confidently conclude that this decrease in carbon isotope values is due to a decrease in C4 plant biomass. The initial decline of C4 plants during the mid-Holocene (8–6 ka) coincided with high counts of tree pollen, as well as decreasing precipitation and insolation, as well as decreasing seasonal contrast (Figs. 2–4, and 5; Liškar et al., 2004; Ji et al., 2005; Peterse et al., 2011; An et al., 2012). C$_3$ plants dominated the Lake Qinghai catchment after ca. 6 ka, coincident with high pCO$_2$ and modeled increasing May temperatures and winter precipitation, which favored C$_3$ plant productivity (Figs. 2–4; Still et al., 2003). Whereas arboreal pollen declined during the late Holocene, likely due to decreased precipitation (Herzschuh et al., 2009), Chenopodiaceae s.s. and Poaceae remained constant, suggesting that C3 plants within these clades became dominant, replacing C$_4$ Chenopodiaceae s.s. and Poaceae that had dominated during the early Holocene (Fig. 3). Today, high pCO$_2$ means that most C$_4$ plant communities are near the edge of their ecological niche, especially those in cold regions (Fig. 5). The predicted 2–3 °C temperature increase on the Tibetan Plateau by 2050 AD will not be sufficient to offset the deleterious effects of 450–500 ppmv CO$_2$, likely resulting in the demise of C$_4$ plants in cold, dry, high-elevation regions (Fig. 5; Taylor et al., 2011).

4.5. C$_4$ plants in high-elevation ecosystems

In contrast to the northeastern Tibetan Plateau, high-elevation sites in equatorial Africa and South America hosted flourishing C$_4$ plant communities during the glacial period. The different timing of maximum C$_4$ plant abundance is likely due to different temperatures and seasonal contrasts. The equatorial sites had higher mean annual temperatures than the northeastern Tibetan Plateau during the glacial period (Figs. 4 and S4). Seasonal insolation contrast is minimal at the equator, ranging from 40 to 60 W m$^{-2}$, with minimum insolation contrast occurring during the glacial period when there were maximum C$_4$ plants (Fig. 54; Berger et al., 2006). Thus, seasonal insolation contrast may not play a dominant role in plant ecosystem variability at the equator. Instead C$_4$ plants flourished during the glacial period likely due to sufficiently warm rainy seasons and low pCO$_2$ (Figs. 4 and S4; Jouzel et al., 2007; Loomis et al., 2012).

At latitudes similar to Lake Qinghai but at lower elevation on the CLP, C$_4$ plants rarely exceeded 40% abundance. Despite their relatively low abundance, C$_4$ plants persisted on the CLP until the late Holocene, perhaps due to warmer, wetter summers (Fig. 4; Zhang et al., 2003; Liu et al., 2005).

5. Conclusions

Our paleoecological data demonstrate that C$_4$ plants can thrive on the Tibetan Plateau, and provide benchmarks for ecosystem models that infer <0.5% C$_4$ plants on the Tibetan Plateau today. The CCSM3 transient simulation infers 0% C$_4$ grasses even during time periods when Lake Qinghai δ$^{13}$C$_{wax}$ suggests up to 50% C$_4$ plants (Figs. 1 and 3, and 55; Still et al., 2003; Z. Liu et al., 2009). Although we have evidence for abundant C$_4$ plant biomass around Lake Qinghai during the glacial period, it will be most important to correctly model the increase in C$_4$ plant abundance during the Lateglacial and early Holocene, which was likely a more widespread phenomenon. Arid- and cold-adapted Chenopodiaceae s.s. likely evolved C$_4$ photosynthesis as a further adaptation to cold, dry, salty conditions on the Tibetan Plateau (Kadereit et al., 2012), and therefore do not fit with the warm, wet thresholds for C$_4$ plants that are currently incorporated into ecosystem models (Collatz et al., 1998; Still et al., 2003). As the spatial resolution of climate and ecosystem models increases, it will be necessary to account for unusual ecosystem dynamics, including cold-, arid-, salt-adapted C$_4$ Chenopodiaceae s.s., in spatially heterogeneous, high-elevation regions.

The combination of saline soils, high irradiance, and summer-focused precipitation on the Tibetan Plateau inherently favor C$_4$ plants, whereas cool conditions and high pCO$_2$ favor C$_3$ plants. A delicate ecological balance therefore exists on the Tibetan Plateau and in other high-elevation regions, where we document large paleoecological transitions between C$_3$ and C$_4$ plants. Lake Qinghai δ$^{13}$C$_{wax}$ provides a paleoecological perspective for studies of modern distributions of C$_4$ plants, which find that temperature, precipitation, and pCO$_2$ all play important roles (Ehleringer et al., 1997; Collatz et al., 1998; Sage et al., 1999; Edwards and Smith, 2010; Kadereit et al., 2012). Due to relatively high pCO$_2$ today, C$_4$ plant communities in cold, dry, high-elevation regions are at the edge of their ecological niche, and will likely decline further as pCO$_2$ increases (Taylor et al., 2011).
Author contributions

YH designed the study; EKT, JZ, PW, and LG performed sample preparation and analysis; EKT, YH, SMC, and SCC performed data analysis and interpretation; CM provided model output and analysis; EKT wrote the main paper and the Supplementary Information, with input from YH, CM, SMC, and SCC.

Acknowledgments

We thank R. Tarozo for lab assistance and E. J. Edwards and one anonymous reviewer for providing feedback on the manuscript. We thank the International Continental Scientific Drilling Program, the United States National Science Foundation, the National Natural Science Foundation of China and the Ministry of Science and Technology of China for their support for the Lake Qinghai Drilling Project, which was led in part by A. Zhisheng. We thank Z. Liu and B. Otto-Bliensner for sharing CCSM3 output. The CCSM3 transient simulation is supported by the P2C2 program at the US National Science Foundation, the A abrupt Change Program, EaSM program, and INCITE computing program at the US Department of Energy, and NCAR. We acknowledge the World Climate Research Programme's Working Group on Coupled Modelling, which is responsible for CMIP, and we thank the climate modeling groups (listed in Table S1) for producing and making available their model output. For CMIP the US Department of Energy's Program for Climate Model Diagnosis and Intercomparison provides coordinating support and led development of software infrastructure in partnership with the Global Organization for Earth System Science Portals. This research was funded by NSF grant #ATM-0902085 to Y. Huang and a NSF Graduate Research Fellowship to EKT.

Appendix A. Supplementary data

Supplementary data related to this article can be found at http://dx.doi.org/10.1016/j.quascirev.2013.12.014.

References
