

Carbon isotopic signatures of soil organic matter correlate with leaf area index across woody biomes

Brenton Ladd^{1,2*}, Pablo L. Peri³, David A. Pepper⁴, Lucas C. R. Silva⁵, Douglas Sheil^{6,7,8}, Stephen P. Bonser², Shawn W. Laffan⁴, Wulf Amelung⁹, Alf Ekblad¹⁰, Peter Eliasson^{10,11}, Hector Bahamonde³, Sandra Duarte-Guardia¹ and Michael Bird^{12,13}

¹Facultad de Ciencias Ambientales, Universidad Científica del Sur, Lima 33, Perú; ²Earth and Environmental Sciences, Evolution and Ecology Research Centre, School of Biological, UNSW Australia, Sydney NSW 2052, Australia; ³Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET)-cc332 (9400), INTA EEA, Santa Cruz, Rio Gallegos, Argentina; ⁴Earth and Environmental Sciences, School of Biological, UNSW Australia, Sydney NSW 2052, Australia; ⁵Department of Land, Air and Water Resources, University of California Davis, CA 95616, USA; ⁶School of Environmental Science and Management, Southern Cross University, PO Box 157, Lismore NSW 2480, Australia; ⁷Center for International Forestry Research (CIFOR), GPO Box 113 BOCBD, Bogor 1600, Indonesia; ⁸Department of Ecology and Natural Resource Management, Norwegian University of Life Sciences, PO Box 5003, NO-1432 Ås, Norway; ⁹Soil Science and Soil Ecology, Institute of Crop Science and Resource Conservation (INRES), University of Bonn, Nussallee 13, D-53115 Bonn, Germany; ¹⁰School of Science and Technology, Örebro University, SE-701 82 Örebro, Sweden; ¹¹Department of Physical Geography and Ecosystem Science, Lund University, Sölvegatan 12, SE-223 62 Lund, Sweden; ¹²Department of Ecology, Swedish University of Agricultural Sciences (SLU), Box 7044, SE-750 07 Uppsala, Sweden; and ¹³Centre for Tropical Environmental and Sustainability Science, School of Earth and Environmental Sciences, James Cook University, PO Box 6811, Cairns, Qld 4870, Australia

Summary

1. Leaf area index (LAI), a measure of canopy density, is a key variable for modelling and understanding primary productivity, and also water use and energy exchange in forest ecosystems. However, LAI varies considerably with phenology and disturbance patterns, so alternative approaches to quantifying stand-level processes should be considered. The carbon isotope composition of soil organic matter ($\delta^{13}\text{C}_{\text{SOM}}$) provides a time-integrated, productivity-weighted measure of physiological and stand-level processes, reflecting biomass deposition from seasonal to decadal time scales.
2. Our primary aim was to explore how well LAI correlates with $\delta^{13}\text{C}_{\text{SOM}}$ across biomes.
3. Using a global data set spanning large environmental gradients in tropical, temperate and boreal forest and woodland, we assess the strength of the correlation between LAI and $\delta^{13}\text{C}_{\text{SOM}}$; we also assess climatic variables derived from the WorldClim database.
4. We found that LAI was strongly correlated with $\delta^{13}\text{C}_{\text{SOM}}$, but was also correlated with Mean Temperature of the Wettest Quarter, Mean Precipitation of Warmest Quarter and Annual Solar Radiation across and within biomes.
5. *Synthesis.* Our results demonstrate that $\delta^{13}\text{C}_{\text{SOM}}$ values can provide spatially explicit estimates of leaf area index (LAI) and could therefore serve as a surrogate for productivity and water use. While $\delta^{13}\text{C}_{\text{SOM}}$ has traditionally been used to reconstruct the relative abundance of C_3 versus C_4 species, the results of this study demonstrate that within stable C_3 - or C_4 -dominated biomes, $\delta^{13}\text{C}_{\text{SOM}}$ can provide additional insights. The fact that LAI is strongly correlated to $\delta^{13}\text{C}_{\text{SOM}}$ may allow for a more nuanced interpretation of ecosystem properties of palaeoecosystems based on palaeosol ^{13}C values.

Key-words: climate, isoscapes, leaf area index, paleoecosystems, plant–soil (below-ground) interactions, productivity, stable isotopes, water stress, $\delta^{13}\text{C}$, $\delta^{13}\text{C}_{\text{SOM}}$

*Correspondence author. E-mail: brenton.ladd@gmail.com.

Introduction

The factors that control carbon isotope composition within a single plant are well understood at the leaf scale (Farquhar, Ehleringer & Hubick 1989). $\delta^{13}\text{C}$ values in plants in natural vegetation communities have also been studied extensively and shown to be affected by rainfall, temperature, soil water content, irradiance and soil nitrogen values (Song *et al.* 2008). Two recent meta-analyses of $\delta^{13}\text{C}$ values in plants that use the C_3 photosynthetic pathway demonstrate a strong correlation between rainfall and leaf $\delta^{13}\text{C}$ values and has led to the suggestion that $\delta^{13}\text{C}$ may be a useful proxy for quantifying aridity in palaeohabitats (Diefendorf *et al.* 2010; Kohn 2010). The $\delta^{13}\text{C}$ values of tree rings have proved useful in quantifying climatic and atmospheric influences and to reconstruct water use efficiency of individual trees and forest stands over time (Silva & Anand 2013), although restrictions may apply (Silva & Horwath 2013).

The situation with $\delta^{13}\text{C}$ values in soil is different. The carbon isotope composition of soil organic matter ($\delta^{13}\text{C}_{\text{SOM}}$) is significantly determined by the fraction of litter derived from C_3 versus C_4 plant species (Wynn & Bird 2007; Lloyd *et al.* 2008), by the contribution of litter from mycorrhizal fungi (Clemmensen *et al.* 2013) and by soil processes associated with the decomposition of organic matter (Baisden *et al.* 2002; Hobbie 2005) or a combination of factors (Silva *et al.* 2013). The C_4 photosynthetic pathway is predominant in tropical grasslands and open savannas. As a result, the $\delta^{13}\text{C}_{\text{SOM}}$ isotopic signature is similar to that found in the biomass of C_4 plants (-15 to -9‰), which is distinct from the isotopic signature of C_3 plants (-21 to -40‰) (Staddon 2004) that are dominant in forests. Values of $\delta^{13}\text{C}_{\text{SOM}}$ can thus be used to quantify the relative abundance of C_3 versus C_4 species in tropical ecosystems, and because these carbon ratios are sufficiently long lasting in the soil carbon pool, $\delta^{13}\text{C}$ can be used to quantify changes in tropical ecosystem structure and function over time (Wynn & Bird 2007). There are also promising first indications that $\delta^{13}\text{C}_{\text{SOM}}$ values might be useful for more than quantifying the relative abundance of C_3 and C_4 species. For example, Cerling *et al.* (2011) used a correlation between tree cover and $\delta^{13}\text{C}_{\text{SOM}}$ in tropical ecosystems to interpret the carbon isotope signatures of African palaeosols in terms of palaeoshade and human evolution. This analysis indicated that several early hominin fossil sites had been savannas rather than woodland or forest, as some had claimed previously (Cerling *et al.* 2011).

Despite the various advances, we remain uncertain as to whether $\delta^{13}\text{C}_{\text{SOM}}$ can provide insight into vegetation structure and function across the full range of ecosystems: from those dominated by C_3 through C_3/C_4 mixed to those dominated solely by C_4 plants. Early observations suggest that $\delta^{13}\text{C}_{\text{SOM}}$ could do so. Drucker *et al.* (2008) used $\delta^{13}\text{C}$ as a proxy for shade in temperate palaeoplant communities. Ladd *et al.* (2009) demonstrated that $\delta^{13}\text{C}_{\text{SOM}}$ is strongly correlated with LAI in extant temperate forest and woodland. Measurements of $\delta^{13}\text{C}_{\text{SOM}}$ have also been used successfully in tropical ecosystems to study decadal patterns of grass species invasion in sites undergoing restoration (Silva *et al.* 2013) and centennial

to millennial shifts from savannas to forests (Silva *et al.* 2008, 2010), which already indicates that $\delta^{13}\text{C}_{\text{SOM}}$ has broad application beyond the temperate zone. The aforementioned study by Cerling *et al.* (2011) also used $\delta^{13}\text{C}_{\text{SOM}}$ to reconstruct the fraction of woody cover in tropical ecosystems. It thus seems reasonable to assume that a relationship between LAI and $\delta^{13}\text{C}_{\text{SOM}}$ will be applicable across present-day tropical, temperate and boreal ecosystems. We assess this possibility by summarizing studies that included the joint assessment of these variables; specifically, we test the hypothesis that $\delta^{13}\text{C}_{\text{SOM}}$ is related to LAI across large environmental gradients and applicable at the continental to global scale.

Materials and methods

STUDY SITES AND LAI MEASUREMENT

We used data from 9 boreal, 45 temperate and 43 tropical old-growth forest and woodland ecosystems. The temperate and tropical data represent a synthesis of existing studies that reported measurements of $\delta^{13}\text{C}_{\text{SOM}}$ in top soil (to 10 cm depth), in which measurements of leaf area index (LAI) of the forest canopy were made, and for which we were able to obtain geographical coordinates precise enough to allow for GIS analyses (Williams *et al.* 2002; Silva *et al.* 2008, 2010; Ladd *et al.* 2009, 2013). The boreal forest samples and measurements were collected from unmanaged mature stands of *Picea abies* (L.) Karst., using the methods described in Ladd *et al.* (2009); it was assumed that any systematic variation in $\delta^{13}\text{C}$ of atmospheric CO_2 (source carbon) was incorporated into $\delta^{13}\text{C}_{\text{SOM}}$ at all sites, but the influence was less than the error estimate in $\delta^{13}\text{C}_{\text{SOM}}$ measurements (Tans, De Jong & Mook 1979). Likewise, the isotopic analyses for the sites described in Ladd *et al.* (2013) followed the methods described in Ladd *et al.* (2009). To standardize and control for the well-known effects of phenology on LAI values (Asner, Scurlock & Hicke 2003), we measured LAI at the peak of the austral summer for the Southern Hemisphere sites (January, February) and at the end of the summer season for the Northern Hemisphere sites (August, September). Our LAI measurements were made using the digital photography method of Macfarlane *et al.* (2007). At each sampling location, at least 20 digital images of the forest canopy were taken and used to calculate LAI using a modified version of the Beer–Lambert light extinction coefficient. At open woodland sites where canopy LAI is more variable, we took up to 40 digital images to compensate for increased variability; see Macfarlane *et al.* (2007) for further description of the digital photography method.

The sampled forest and woodland communities were selected from mature populations under near steady state conditions, spanning a wide range of environments and site productivities. Mean annual temperature (MAT) ranged between 3.3 and 26.4 °C, mean annual precipitation (MAP) between 304 and 2013 mm per annum and the average height of mature trees from 2 to 40 m across all sites.

SAMPLING AND ISOTOPIC ANALYSIS OF SOILS

Within each quadrat used for LAI measurement, we collected nine replicate soil samples at 0- to 10-cm depth. From the nine soil samples collected within each LAI quadrat, we created three composite samples, that is. each composite sample contained an equal amount of soil from three soil samples. The stable isotope composition of these soil samples (expressed as $\delta^{13}\text{C}$ in per mill units as calculated in relation to Vienna

Pee Dee Belemnite standard) was measured at the University of Bonn after dry combustion using an elemental analyzer (Flash EA, 1112 Series, Thermo Electron GmbH, Bremen, Germany) coupled with a Delta V Advantage isotope ratio mass spectrometer (Thermo Electron GmbH). Repeated measures of laboratory standards and selected $\delta^{13}\text{C}_{\text{SOM}}$ samples had a standard error equal to or $< 0.02\text{‰}$.

SPATIAL DATA – CLIMATE AND SATELLITE-DERIVED LAI MEASUREMENTS

The climate parameters for each site were derived from the WorldClim data set (Hijmans *et al.* 2005). WorldClim contains global geographical surfaces for 19 different climatic parameters that describe rainfall, temperature and variation in those parameters at a resolution of 0.008333° (approximately 1 km). Incoming solar radiation (Joules $\text{m}^{-2} \text{year}^{-1}$) was calculated from the Solar Radiation tool in ARCGIS version 9.3.1 (ESRI, California, USA), with topography data from the 3-arc-second resolution NASA Shuttle Radar Topography Mission Digital Elevation Model (SRTM DEM) of the globe (Jarvis *et al.* 2008). Satellite-derived LAI values were obtained from the MODIS 15A2 product (downloaded from: www.daac.ornl.gov, more information about this product can be found on the website), which is a 1-km resolution, 8-day composited data set. For each site, satellite LAI values were downloaded for each 8-day period from the first year in which the source study was published (see Appendix S1 in Supporting Information). The MODIS product includes quality control (QC) values. A QC value equal to 0 indicates that the value is reliable: not influenced by cloud or other atmospheric anomalies (Wang *et al.* 2005). The average of the usable data (where QC = 0) was thus used to calculate long-term average LAI for each site.

DATA ANALYSIS

We used a multivariate approach with a stepwise procedure to identify the most important variables driving changes in $\delta^{13}\text{C}_{\text{SOM}}$ and to identify and remove variables that were collinear with other explanatory variables in the data set, following the method described by Fox (2002, p 216). First, we calculated variance inflation factors (VIFs) for the complete set of explanatory variables. We then began an iterative process in which we deleted the variable with the highest VIF score, recalculated VIF scores for the remaining variables and then repeated this process until all remaining variables had VIF scores < 10 (after Quinn & Keough 2002). Through this iterative process, we identified nine non-collinear explanatory variables. The interactive effect of these nine variables on $\ln(\text{LAI} + 1)$ -transformed ground-based LAI values was then assessed using multiple linear regressions (see Appendix S2). The parsimony of nine different regression models that varied in complexity (i.e. the number of independent variables ranged from 1 to 9) was then assessed using Akaike's information criterion (AIC). The regression model with the best AIC ranking contained eight variables, and the relative importance of each of these eight environmental variables was then assessed by partitioning the sum of squares from the regression analysis (see the η^2 values of Table 1). All statistical analyses were performed using XLSTAT (Addin Soft, Paris, France).

Results

Drawing on our data set from 43 tropical, 45 temperate and nine boreal sites, we found a strong relationship between $\delta^{13}\text{C}_{\text{SOM}}$ and ground-based measurements of LAI (Fig. 1a).

Table 1. Summary statistics for the regression model for prediction of ground-based $\ln(n + 1)$ -transformed LAI with the best AIC ranking. SS = sum of squares, P = probability, d.f. = degrees of freedom and η^2 is percentage of variation of the r^2 of the regression model explained by each independent variable. Q = Radiation $\text{J m}^{-2} \text{year}^{-1}$, $\text{ISO}_{\text{THERM}}$ = Isothermality ((Mean Diurnal Range/Temperature Annual Range)*100), MT_{WQ} = Mean Temperature of Wettest Quarter, MT_{DQ} = Mean Temperature of Driest Quarter, $\text{PREC}_{\text{SEAS}}$ = Precipitation Seasonality (Coefficient of Variation), PREC_{WQ} = Precipitation of Warmest Quarter, PREC_{CQ} = Precipitation of Coldest Quarter. See www.worldclim.org for further detail on the climate parameters and Appendix S2 for a comparison of AIC values across all 9 regression models. $F = 65.66$, d.f. = 8,88; $P < 0.0001$, $R^2 = 0.85$. Model – $\ln(\text{LAI} + 1) = -1.89 - 2.03\text{E-}10*Q - 0.10*\delta^{13}\text{C}_{\text{SOM}} + 1.47*\text{ISO}_{\text{THERM}} + 2.43\text{E-}02*\text{MT}_{\text{WQ}} - 2.6\text{E-}02*\text{MT}_{\text{DQ}} + 4.67\text{E-}03*\text{PREC}_{\text{SEAS}} + 1.3\text{E-}03*\text{PREC}_{\text{WQ}} + 6.09\text{E-}04*\text{PREC}_{\text{CQ}}$

Source	d.f.	SS	F	$P > F$	η^2
Q	1	1.2	32.112	< 0.0001	5.23
$\delta^{13}\text{C}_{\text{SOM}}$	1	10.3	274.503	< 0.0001	44.7
$\text{ISO}_{\text{THERM}}$	1	0.16	4.439	0.038	0.72
MT_{WQ}	1	4.26	113.403	< 0.0001	18.4
MT_{DQ}	1	0.91	24.348	< 0.0001	3.9
$\text{PREC}_{\text{SEAS}}$	1	0.002	0.055	0.816	0.008
PREC_{WQ}	1	2.07	55.225	< 0.0001	9.0
PREC_{CQ}	1	0.79	21.215	< 0.0001	3.4
Error	88	3.30			

In contrast, there was no correlation between the MODIS-derived estimates of LAI and $\delta^{13}\text{C}_{\text{SOM}}$ (Fig. 1b). When we considered a broad range of forest and woodland ecosystems, simultaneously, we found $\delta^{13}\text{C}_{\text{SOM}}$ is the most important correlate of ground-based LAI, explaining 45% of the variance in LAI values (Table 1, Figs 1 and 2). This was a slightly weaker correlation than reported by Ladd *et al.* (2009), which only considered a single biome (temperate forest and woodland). The WorldClim climatic variables, Mean Temperature of the Wettest Quarter and Mean Precipitation of the Warmest Quarter, were also strong correlates of ground-based LAI, perhaps reflecting the synergistic impact of temperature and water on plant productivity (Table 1). Annual solar radiation (Q) was an additional correlate of ground-based LAI explaining 5.2% of the variance in the data. A multiple regression model that included $\delta^{13}\text{C}_{\text{SOM}}$, solar radiation and six climatic variables obtained from WorldClim finally explained 85% of the variance in ground-based LAI values in this global data set (Table 1). The small difference between the root mean squared error (RMSE) and the 'press RMSE' values (Appendix S2) indicates that the regression model has been robust and not sensitive to the presence or absence of a limited number of observations within the data set.

Discussion

Our results (see Figs 1 and 2, Table 1 and Appendix S1 and S2) confirm our hypothesis, showing that $\delta^{13}\text{C}_{\text{SOM}}$ values are well correlated with key ecosystem parameters across a wide range of forest and woodland ecosystems, explaining jointly

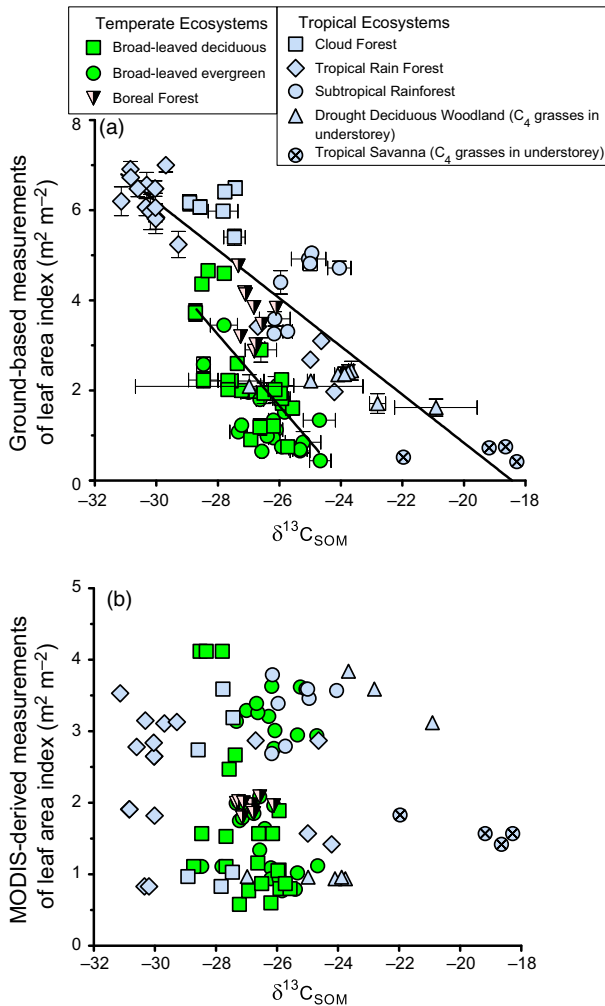


Fig. 1. (a) The relationship between ground-based measurements of leaf area index (m^2 of tree canopy per m^2 of ground) and the $\delta^{13}\text{C}$ of soil organic matter ($\delta^{13}\text{C}_{\text{SOM}}$) across a broad range of forest and woodland ecosystems. (b) The relationship between MODIS-derived leaf area index and $\delta^{13}\text{C}_{\text{SOM}}$ at the same geographical locations. The geographical coordinates of the field sites and the raw data are given in Appendix S1.

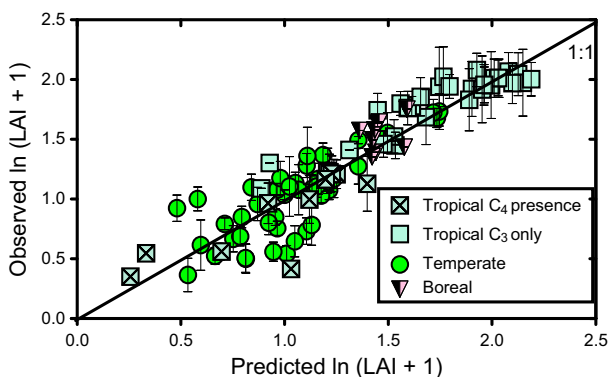


Fig. 2. Ground-based LAI (Mean \pm SE) as a function of $\delta^{13}\text{C}_{\text{SOM}}$, solar radiation and the climatic variables included in the statistical model described in Table 1. $y = x + 8E-15$. $R^2 = 0.85$.

with the climate indices over 85% of the variance in ground-based LAI. Although $\delta^{13}\text{C}_{\text{SOM}}$ was the single best predictor of ground-based LAI across both tropical and temperate ecosystems, the relationship was stronger in tropical ecosystems. This reflects the much greater relative contribution of C_4 plants to the productivity of tropical ecosystems compared to temperate and boreal ecosystems (Wynn & Bird 2007; Ladd *et al.* 2009).

THE UNRESOLVED MECHANISTIC BASIS OF THE CORRELATION

To understand the correlation between LAI and $\delta^{13}\text{C}_{\text{SOM}}$, we must consider the different factors affecting isotopic discrimination in plants and soil organic matter (SOM). The largest fractionation step to occur between the $\delta^{13}\text{C}$ of atmospheric CO_2 and the $\delta^{13}\text{C}$ of SOM happens physiologically at the leaf-atmosphere interface during CO_2 assimilation (Farquhar, Ehleringer & Hubick 1989; Ladd *et al.* 2009). Assimilated CO_2 is used to produce plant biomass, which eventually senesces and enters the soil where it determines $\delta^{13}\text{C}_{\text{SOM}}$ values (Peri *et al.* 2012). For the sites in southern Patagonia, we can state with confidence that $\delta^{13}\text{C}_{\text{SOM}}$ values are largely a reflection of the isotopic signal of the plants growing on site, which is modified slightly by soil processes – a modification that correlates positively with rainfall (see Fig. 3a in Peri *et al.* 2012).

Re-fixation of respired, isotopically depleted CO_2 can occur below dense forest canopies (high LAI) (Sternberg *et al.* 1997). Such re-fixation may strengthen the correlation between $\delta^{13}\text{C}_{\text{SOM}}$ and LAI as this process is likely to be less significant in more open, low LAI woodlands. The ratio of CO_2 assimilation to stomatal conductance (A/g) decreases for plant foliage deep within a forest canopy, which in turn causes $\delta^{13}\text{C}$ values to become more negative (Koch *et al.* 2004). Increased self-shading as canopy density increases may thus also be a causal factor in the negative correlation between $\delta^{13}\text{C}_{\text{SOM}}$ and LAI.

According to a recent application of the Baldocchi & Bowling (2003) CANISOTOPE model, which incorporates microclimatic and ecophysiological complexity over tree canopy depth, Voelker *et al.* (2014) attributed canopy variation in $\delta^{13}\text{C}$ and in tree-ring width across environmental gradients to variation in vapour pressure deficit (VPD) and light levels. The correlation between $\delta^{13}\text{C}_{\text{SOM}}$ and LAI across the environmental gradients we studied (Fig. 1a) is consistent with this mechanism. The possibility that a negative correlation between LAI and $\delta^{13}\text{C}_{\text{SOM}}$ is determined by ecophysiological processes represented in models like CANISOTOPE should be evaluated.

APPLICATIONS TO EXTANT ECOSYSTEMS

Ecosystem LAI determines the potential for a forest canopy to absorb solar radiation per unit area and is therefore a good indicator of potential productivity at the ecosystem scale. LAI is also a reliable proxy for the evaporative surface of a forest

canopy and thus provides a good indication of the potential for a forest to use water (Eamus *et al.* 2006), so it is useful to ask whether measurements of $\delta^{13}\text{C}_{\text{SOM}}$ values can enhance our ability to quantify ecosystem functioning at the stand scale beyond the high-frequency, high-resolution data available from remote sensing (i.e. MODIS). The answer we believe is yes, for three reasons. Firstly, $\delta^{13}\text{C}_{\text{SOM}}$ measurements are obtained from the slow-cycling soil carbon pool and therefore provide a temporally integrated measure of ecosystem functioning (Ometto *et al.* 2006; Ladd *et al.* 2009). This is because $\delta^{13}\text{C}_{\text{SOM}}$ values reflect the contribution to the soil on a carbon mass basis from the source vegetation (i.e. above- and below-ground litter deposition) over many years. LAI in contrast varies seasonally and annually, for example in response to natural year-to-year variations in mean annual rainfall (Eamus *et al.* 2006). With $\delta^{13}\text{C}_{\text{SOM}}$, we can therefore avoid the need for repeated (and time consuming) measurements of LAI across different seasons and in different years to account for phenological variations that occur with LAI (Asner, Scurlock & Hicke 2003).

Secondly, although repeated measurements of LAI are more viable with satellite-based sensors, satellite-derived estimates of LAI are known to saturate at high levels of LAI, which limits their utility in global analyses (Song 2013). Comparison of the ground-based measurements of LAI versus $\delta^{13}\text{C}_{\text{SOM}}$ (Fig. 1a) and the MODIS-derived LAI values versus $\delta^{13}\text{C}_{\text{SOM}}$ (Fig. 1b) shows that despite technological advances, ground truthing remains important. The global compilation and up-scaling of isotope data, that is. the concept of isoscapes currently being pioneered in North America (Bowen 2010; Powell, Yoo & Still 2012), is a promising initiative that could be useful for validating the next generation of remote sensing products, that is lidar and radar remote sensing (Ladd & Peri 2013). Our results demonstrate that an isoscape for $\delta^{13}\text{C}_{\text{SOM}}$ values could provide spatially explicit estimates of LAI and therefore key aspects of ecosystem functioning, such as productivity at the ecosystem scale. As $\delta^{13}\text{C}_{\text{SOM}}$ is essentially a productivity-weighted signal of net photosynthetic production, it may also be considered a community-weighted estimate of the range of plant functional traits expressed within a given plant community (see also van Wijk, Williams & Shaver 2005; Reich 2012). Our results thus support the recent suggestion (Wang *et al.* 2012; Ali *et al.* 2013) that plant functional traits scale up to determine the attributes of entire ecosystems.

APPLICATIONS TO PALAEOECOSYSTEMS

Constraining all the possible independent variables that can influence ^{13}C values in palaeosols presents a formidable challenge. Constraining diagenetic effects, for example, may prove unachievable (Tippie, Meyers & Pagani 2010). Nevertheless, establishing correlations between the natural abundance of stable isotopes and ecosystem-scale parameters in extant ecosystems could contribute to a more nuanced interpretation of palaeosol ^{13}C values (Drucker *et al.* 2008; Cerling *et al.* 2011). The use of plant isotope composition for

quantifying aridity in palaeohabitats is one example (Diefendorf *et al.* 2010; Kohn 2010). Historically, $\delta^{13}\text{C}_{\text{SOM}}$ values have been used to quantify the relative abundance of C_3 versus C_4 species. Our results suggest that $\delta^{13}\text{C}_{\text{SOM}}$ can do more than this. The fact that $\delta^{13}\text{C}_{\text{SOM}}$ is correlated to LAI across a broad range of ecosystems is consistent with the idea that $\delta^{13}\text{C}_{\text{SOM}}$ relates to a range of ecosystem functions such as productivity and stand water use. However, $\delta^{13}\text{C}_{\text{SOM}}$ values were also affected by climatic variables, which jointly explained over 35% of the variance in LAI (see Table 1). Therefore, use of $\delta^{13}\text{C}_{\text{SOM}}$ as a proxy for canopy density in palaeoecosystems requires us to disentangle the potentially confounding and interacting effects of climate and tree cover on $\delta^{13}\text{C}_{\text{SOM}}$ in extant ecosystems. In this respect, measurement of other isotopic signals, for example the coupled use of $\delta^{18}\text{O}$, δD , $\delta^{15}\text{N}$ and $\delta^{13}\text{C}$ (Brader *et al.* 2010), hold promise for better describing both palaeo- and extant ecosystems.

Acknowledgements

We thank the City of Onkaparinga, the University of New South Wales, the University of Bonn and the Swedish University of Agricultural Sciences for financial support and the anonymous reviewers and editorial team at *Journal of Ecology* for helpful advice and observations on the manuscript.

References

- Ali, A.A., Medlyn, B.E., Crous, K.Y. & Reich, P.B. (2013) A trait-based ecosystem model suggests that long-term responsiveness to rising atmospheric CO_2 concentration is greater in slow-growing than fast-growing plants. *Functional Ecology*, **27**, 1011–1022.
- Asner, G.P., Scurlock, J.M.O. & Hicke, J.A. (2003) Global synthesis of leaf area index observations: implications for ecological and remote sensing studies. *Global Ecology and Biogeography*, **12**, 191–205.
- Baisden, W., Amundson, R., Cook, A. & Brenner, D. (2002) Turnover and storage of C and N in five density fractions from California annual grassland surface soils. *Global Biogeochemical Cycles*, **16**, 1117.
- Baldocchi, D. & Bowling, D. (2003) Modelling the discrimination of ^{13}C above and within a temperate broad-leaved forest canopy on hourly to seasonal time scales. *Plant, Cell & Environment*, **26**, 231–244.
- Bowen, G.J. (2010) Isoscapes: spatial pattern in isotopic biogeochemistry. *Annual Review of Earth and Planetary Sciences*, **38**, 161–187.
- Brader, A.V., van Winden, J.F., Bohncke, S.J.P., Beets, C.J., Reichart, G.-J. & de Leeuw, J.W. (2010) Fractionation of hydrogen, oxygen and carbon isotopes in n-alkanes and cellulose of three Sphagnum species. *Organic Geochemistry*, **41**, 1277–1284.
- Cerling, T.E., Wynn, J.G., Andanje, S.A., Bird, M.I., Korir, D.K., Levin, N.E., Mace, W., Macharia, A.N., Quade, J. & Remien, C.H. (2011) Woody cover and hominin environments in the past 6 million years. *Nature*, **476**, 51–56.
- Clemmensen, K., Bahr, A., Ovaskainen, O., Dahlberg, A., Ekblad, A., Wallander, H., Stenlid, J., Finlay, R., Wardle, D. & Lindahl, B. (2013) Roots and associated fungi drive long-term carbon sequestration in boreal forest. *Science*, **339**, 1615–1618.
- Diefendorf, A.F., Mueller, K.E., Wing, S.L., Koch, P.L. & Freeman, K.H. (2010) Global patterns in leaf ^{13}C discrimination and implications for studies of past and future climate. *Proceedings of the National Academy of Sciences*, **107**, 5738–5743.
- Drucker, D.G., Bridault, A., Hobson, K.A., Szuma, E. & Bocherens, H. (2008) Can carbon-13 in large herbivores reflect the canopy effect in temperate and boreal ecosystems? Evidence from modern and ancient ungulates. *Palaeogeography, Palaeoclimatology, Palaeoecology*, **266**, 69–82.
- Eamus, D., Hatton, T., Cook, P. & Colvin, C. (2006) *Ecohydrology: Vegetation Function, Water and Resource Management*. CSIRO Publishing, Melbourne, Vic., Australia.
- Farquhar, G.D., Ehleringer, J.R. & Hubick, K.T. (1989) Carbon isotope discrimination and photosynthesis. *Annual Review of Plant Physiology and Plant Molecular Biology*, **40**, 503–537.

- Fox, J. (2002) *An R and S-Plus Companion to Applied Regression*. Sage Publications, Thousand Oaks, CA.
- Hijmans, R.J., Cameron, S.E., Parra, J.L., Jones, P.G. & Jarvis, A. (2005) Very high resolution interpolated climate surfaces for global land areas. *International Journal of Climatology*, **25**, 1965–1978.
- Hobbie, E.A. (2005) Assessing functions of soil microbes with isotopic measurements. *Microorganisms in Soils: Roles in Genesis and Functions* (eds F. Buscot & A. Varma), pp. 383–402. Springer-Verlag, New York, NY.
- Jarvis, A., Reuter, H.L., Nelson, A. & Guevara, E. (2008) *Hole-filled Seamless SRTM Data V4*, International Centre for Tropical Agriculture (CIAT), available at <http://srtm.csi.cgiar.org> (accessed 27/05/2011).
- Koch, G.W., Sillett, S.C., Jennings, G.M. & Davis, S.D. (2004) The limits to tree height. *Nature*, **428**, 851–854.
- Kohn, M.J. (2010) Carbon isotope compositions of terrestrial C3 plants as indicators of (paleo)ecology and (paleo)climate. *Proceedings of the National Academy of Sciences*, **107**, 19691–19695.
- Ladd, B. & Peri, P.L. (2013) REDD+ en Latinoamérica: el caso de Perú. *Bosque (Valdivia)*, **34**, 125–128.
- Ladd, B., Bonser, S.P., Peri, P.L., Larsen, J.R., Laffan, S.W., Pepper, D.A. & Cendón, D.I. (2009) Towards a physical description of habitat: quantifying environmental adversity (abiotic stress) in temperate forest and woodland ecosystems. *Journal of Ecology*, **97**, 964–971.
- Ladd, B., Laffan, S.W., Amelung, W., Peri, P.L., Silva, L.C.R., Gervassi, P., Bonser, S.P., Navall, M. & Sheil, D. (2013) Estimates of soil carbon concentration in tropical and temperate forest and woodland from available GIS data on three continents. *Global Ecology and Biogeography*, **22**, 461–469.
- Lloyd, J., Bird, M.I., Vellen, L., Miranda, A.C., Veenendaal, E.M., Djagbletey, G., Miranda, H.S., Cook, G. & Farquhar, G.D. (2008) Contributions of woody and herbaceous vegetation to tropical savanna ecosystem productivity: a quasi-global estimate. *Tree Physiology*, **28**, 451–468.
- Macfarlane, C., Hoffman, M., Eamus, D., Kerp, N., Higginson, S., McMurtrie, R. & Adams, M. (2007) Estimation of leaf area index in eucalypt forest using digital photography. *Agricultural and Forest Meteorology*, **143**, 176–188.
- Ometto, J., Ehleringer, J.R., Domingues, T.F., Berry, J.A., Ishida, F.Y., Mazzi, E., Higuchi, N., Flanagan, L.B., Nardoto, G.B. & Martinelli, L.A. (2006) The stable carbon and nitrogen isotopic composition of vegetation in tropical forests of the Amazon Basin, Brazil. *Biogeochemistry*, **79**, 251–274.
- Peri, P.L., Ladd, B., Pepper, D.A., Bonser, S.P., Laffan, S.W. & Amelung, W. (2012) Carbon ($\delta^{13}\text{C}$) and nitrogen ($\delta^{15}\text{N}$) stable isotope composition in plant and soil in Southern Patagonia's native forests. *Global Change Biology*, **18**, 311–321.
- Powell, R.L., Yoo, E.-H. & Still, C.J. (2012) Vegetation and soil carbon-13 isoscapes for South America: integrating remote sensing and ecosystem isotope measurements. *Ecosphere*, **3**, art109.
- Quinn, G.P. & Keough, M.J. (2002) *Experimental Design and Data Analysis for Biologists*, 1st edn. Cambridge University Press, Cambridge, UK.
- Reich, P.B. (2012) Key canopy traits drive forest productivity. *Proceedings of the Royal Society B: Biological Sciences*, **279**, 2128–2134.
- Silva, L.C. & Anand, M. (2013) Probing for the influence of atmospheric CO_2 and climate change on forest ecosystems across biomes. *Global Ecology and Biogeography*, **22**, 83–92.
- Silva, L.C. & Horwath, W.R. (2013) Explaining global increases in water use efficiency: why have we overestimated responses to rising atmospheric CO_2 in natural forest ecosystems? *PLoS One*, **8**, e53089.
- Silva, L.C.R., Sternberg, L.S.L., Haridasan, M., Hoffmann, W.A., Miralles-Wilhelm, F. & Franco, A.C. (2008) Expansion of gallery forests into central Brazilian savannas. *Global Change Biology*, **14**, 2108–2118.
- Silva, L.C.R., Haridasan, M., Sternberg, L.S.L., Franco, A.C. & Hoffmann, W.A. (2010) Not all forests are expanding over central Brazilian savannas. *Plant and Soil*, **333**, 431–442.
- Silva, L.C., Corrêa, R.S., Doane, T.A., Pereira, E.I. & Horwath, W.R. (2013) Unprecedented carbon accumulation in mined soils: the synergistic effect of resource input and plant species invasion. *Ecological Applications*, **23**, 1345–1356.
- Song, C. (2013) Optical remote sensing of forest leaf area index and biomass. *Progress in Physical Geography*, **37**, 98–113.
- Song, M., Duan, D., Chen, H., Hu, Q., Zhang, F., Xu, X., Tian, Y., Ouyang, H. & Peng, C. (2008) Leaf d^{13}C reflects ecosystem patterns and responses of alpine plants to the environments on the Tibetan Plateau. *Ecography*, **31**, 499–508.
- Staddon, P.L. (2004) Carbon isotopes in functional soil ecology. *Trends in Ecology & Evolution*, **19**, 148–154.
- Sternberg, L.D.S., Moreira, M.Z., Martinelli, L.A., Victoria, R.L., Barbosa, E.M., Bonates, L. & Nepstad, D.C. (1997) Carbon dioxide recycling in two Amazonian tropical forests. *Agricultural and Forest Meteorology*, **88**, 259–268.
- Tans, P., De Jong, A. & Mook, W. (1979) Natural atmospheric ^{14}C variation and the Suess effect. *Nature*, **280**, 826–828.
- Tipple, B.J., Meyers, S.R. & Pagani, M. (2010) Carbon isotope ratio of Cenozoic CO_2 : a comparative evaluation of available geochemical proxies. *Paleoceanography*, **25**, PA32020.
- Voelker, S.L., Meinzer, F.C., Lachenbruch, B., Brooks, J.R. & Guyette, R.P. (2014) Drivers of radial growth and carbon isotope discrimination of bur oak (*Quercus macrocarpa* Michx.) across continental gradients in precipitation, vapour pressure deficit and irradiance. *Plant, Cell & Environment*, **37**, 766–779.
- Wang, Q., Tenhunen, J., Dinh, N.Q., Reichstein, M., Otieno, D., Granier, A. & Pilegard, K. (2005) Evaluation of seasonal variation of MODIS derived leaf area index at two European deciduous broadleaf forest sites. *Remote Sensing of Environment*, **96**, 475–484.
- Wang, Y.P., Lu, X.J., Wright, I.J., Dai, Y.J., Rayner, P.J. & Reich, P.B. (2012) Correlations among leaf traits provide a significant constraint on the estimate of global gross primary production. *Geophysical Research Letters*, **39**, L19405.
- van Wijk, M.T., Williams, M. & Shaver, G.R. (2005) Tight coupling between leaf area index and foliage N content in arctic plant communities. *Oecologia*, **142**, 421–427.
- Williams, M., Shimabukuro, Y.E., Herbert, D.A., Pardi Lacruz, S., Renno, C. & Rastetter, E.B. (2002) Heterogeneity of soils and vegetation in an Eastern Amazonian Rain Forest: implications for scaling up biomass and production. *Ecosystems*, **5**, 692–704.
- Wynn, J.G. & Bird, M.I. (2007) C4-derived soil organic carbon decomposes faster than its C3 counterpart in mixed C3/C4 soils. *Global Change Biology*, **13**, 2206–2217.

Received 10 April 2014; accepted 4 August 2014

Handling Editor: Hans Cornelissen

Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix S1. Raw data.

Appendix S2. Summary statistics from the multiple regression analysis.