



Leaf to canopy upscaling approach affects the estimation of canopy traits

Tawanda W. Gara, Andrew K. Skidmore, Roshanak Darvishzadeh & Tiejun Wang

To cite this article: Tawanda W. Gara, Andrew K. Skidmore, Roshanak Darvishzadeh & Tiejun Wang (2019) Leaf to canopy upscaling approach affects the estimation of canopy traits, GIScience & Remote Sensing, 56:4, 554-575, DOI: [10.1080/15481603.2018.1540170](https://doi.org/10.1080/15481603.2018.1540170)

To link to this article: <https://doi.org/10.1080/15481603.2018.1540170>



© 2018 The Author(s). Published by Informa UK Limited, trading as Taylor & Francis Group.



Published online: 30 Oct 2018.



Submit your article to this journal [↗](#)



Article views: 1339



View related articles [↗](#)







View Crossmark data [↗](#)



Citing articles: 6 View citing articles [↗](#)



Leaf to canopy upscaling approach affects the estimation of canopy traits

Tawanda W. Gara ^{*a}, Andrew K. Skidmore ^{a,b}, Roshanak Darvishzadeh ^a
and Tiejun Wang ^a

^aFaculty of Geo-Information Science and Earth Observation (ITC), University of Twente, P.O. Box 6, 7500 Enschede, The Netherlands; ^bDepartment of Environmental Science, Macquarie University, 2106, Sydney, Australia

(Received 1 May 2018; accepted 16 October 2018)

In remote sensing applications, leaf traits are often upscaled to canopy level using sunlit leaf samples collected from the upper canopy. The implicit assumption is that the top of canopy foliage material dominates canopy reflectance and the variability in leaf traits across the canopy is very small. However, the effect of different approaches of upscaling leaf traits to canopy level on model performance and estimation accuracy remains poorly understood. This is especially important in short or sparse canopies where foliage material from the lower canopy potentially contributes to the canopy reflectance. The principal aim of this study is to examine the effect of different approaches when upscaling leaf traits to canopy level on model performance and estimation accuracy using spectral measurements (*in-situ* canopy hyperspectral and simulated Sentinel-2 data) in short woody vegetation. To achieve this, we measured foliar nitrogen (N), leaf mass per area (LMA), foliar chlorophyll and carbon together with leaf area index (LAI) at three vertical canopy layers (lower, middle and upper) along the plant stem in a controlled laboratory environment. We then upscaled the leaf traits to canopy level by multiplying leaf traits by LAI based on different combinations of the three canopy layers. Concurrently, *in-situ* canopy reflectance was measured using an ASD FieldSpec-3 Pro FR spectrometer, and the canopy traits were related to *in-situ* spectral measurements using partial least square regression (PLSR). The PLSR models were cross-validated based on repeated *k-fold*, and the normalized root mean square errors (nRMSE_{cv}) obtained from each upscaling approach were compared using one-way analysis of variance (ANOVA) followed by *Tukey's post hoc* test. Results of the study showed that leaf-to-canopy upscaling approaches that consider the contribution of leaf traits from the exposed upper canopy layer together with the shaded middle canopy layer yield significantly ($p < 0.05$) lower error (nRMSE_{cv} < 0.2 for canopy N, LMA and carbon) as well as high explained variance ($R^2 > 0.71$) for both *in-situ* hyperspectral and simulated Sentinel-2 data. The widely-used upscaling approach that considers only leaf traits from the upper illuminated canopy layer yielded a relatively high error (nRMSE_{cv} > 0.2) and lower explained variance ($R^2 < 0.71$) for canopy N, LMA and carbon. In contrast, canopy chlorophyll upscaled based on leaf samples collected from the upper canopy and total canopy LAI exhibited a more accurate relationship with spectral measurements compared with other upscaling approaches. Results of this study demonstrate that leaf to canopy upscaling approaches have a profound effect on canopy traits estimation for both *in-situ* hyperspectral measurements and simulated Sentinel-2 data in short woody vegetation. These findings have implications for field sampling protocols of leaf traits measurement as well as upscaling leaf traits to canopy level especially in short and less foliated vegetation where leaves from the lower canopy contribute to the canopy reflectance.

*Corresponding author. Tawanda W. Gara Email: t.w.gara@utwente.nl

Keywords: leaf traits; canopy traits; vertical heterogeneity; leaf-to-canopy upscaling; *in-situ* hyperspectral; Sentinel-2

Introduction

Essential biodiversity variables (EBVs) such as leaf traits play a key role in ecosystem structure, functioning and parameterization of dynamic biogeochemical models and nutrient budget simulations (Scheiter, Langan, and Higgins 2013). For example, leaf chlorophyll content is a critical leaf trait in assessing plant physiological status (plant health and/or phenological stage) as well as a plant's photosynthetic capacity (Malenovsky et al. 2013). Foliar nitrogen (N) is a key element in chlorophyll (~6% by weight) (Kokaly et al. 2009) and in enzymes responsible for carbon fixation i.e. ribulose-1.5-biphosphate (RuBP) carboxylase and phenolpyruvate (PEP) essential for photosynthesis in C₃ and C₄ plants, respectively (Schlemmer et al. 2013; Cho et al. 2013; Gibson 2008). In addition, leaf mass per area (LMA) and carbon content mirror the plant economic spectrum with regard to nutrients uptake, light harvesting and carbon sequestration (Poorter et al. 2009; Martin and Thomas 2011). An improved understanding of leaf traits is critical in characterizing, monitoring and simulating ecosystem biogeochemical fluxes over space and time. In this regard, leaf traits are a critical component of ecosystem functional and structural diversity – proxies of essential biodiversity variables (EBVs) (Skidmore et al. 2015). Therefore, estimating leaf traits improves the conservation and monitoring efforts of EBVs fluxes towards the Aichi Biodiversity Targets (Pereira et al. 2013).

Remote sensing provides a cost-effective and practical means of estimating and monitoring leaf traits for biodiversity conservation (Kissling et al. 2017). Field spectroscopy and satellite multispectral data such as Sentinel-2 are critical primary data sources that can improve quantitative estimation and monitoring of foliar traits (Chemura et al. 2018). Essentially, two modelling approaches [i.e. empirical (statistical) and physical models (radiative transfer models-RTM)] are employed to estimate field measured leaf traits from spectral data. Empirical models explore parametric and non-parametric statistical relationships between spectral data or indices and field measured leaf traits using statistical techniques such as stepwise regression and partial least squares regression (Verrelst et al. 2015). Physical models, on the other hand, apply radiation transfer laws to explicitly simulate light absorption, transmittance and scattering inside vegetation canopies by accounting for leaf traits content, canopy structural properties and soil background (Yi et al. 2014; Croft et al. 2015). Quantitative leaf traits that are related or matched to spectral measurements based on these modelling approaches are often determined from leaf samples collected from dominant and co-dominant species within sampling units (Homolová et al. 2013). The leaf traits expressed either in mass (concentration) or area (content) based units are then upscaled to the canopy level using two techniques, i.e. the direct extrapolation and canopy integrated approaches (He and Mui 2010). The direct extrapolation approach applies the relationship between leaf traits and reflectance or vegetation indices observed at leaf level directly to the canopy level (Yoder and Pettigrew-Crosby 1995). The basic assumption of the direct extrapolation method is that trait content or concentration of leaves across the vertical canopy profile is homogeneous (Peterson et al. 1988). However, the direct extrapolation approach is not commonly used because it does not account for canopy structure – a key variable that drives canopy reflectance (Knyazikhin et al. 2013). The canopy integrated method on the other hand upscale leaf

traits to canopy level by accounting for LAI or foliage biomass. The plant traits at canopy level are then regressed against canopy reflectance measured using a field spectroradiometer or airborne and satellite multispectral sensors. In both upscaling approaches, leaf samples for determining traits are often collected from the sunlit top-of-the canopy layer, especially in highly foliated forest canopies. The assumption underlying this widely used approach is that the vertical variation in leaf traits across the canopy is small and leaves at the top of the canopy dominate canopy reflectance and thus represents the whole canopy (Thomas et al. 2008). However, in short woody vegetation the foliage material across the vertical profile significantly contributes to the canopy spectral signal (Roelofsen et al. 2013). This phenomenon complicates leaf sampling protocols because “top of canopy” sampling becomes less valid. To this end, establishing an ecologically meaningful canopy trait value that corresponds to the overall canopy spectral signal is critical when estimating and mapping canopy traits.

The distribution of leaf trait content across vegetation canopies is complex and often varies across the vertical canopy profile (Hirose, Werger, and van Rheenen 1989; Gara et al. 2018). Plants often exhibit higher nutrient content in the upper canopy and on illuminated leaves that receive increased radiation amounts (Chen et al. 1993; Weerasinghe et al. 2014). The variation in leaf traits across the canopy vertical profile is an established plant physiological mechanism designed to maintain an equilibrium between the RuBisCo-limited rate of carboxylation and electron transport limited rate of carboxylation (Chen et al. 1993). Recent research on multiple-layer radiative transfer models (MRTM) have shown that the vertical heterogeneity in foliar traits (LMA, chlorophyll and water) have a significant influence on simulated canopy reflectance and subsequent retrieval of canopy traits especially in low to moderately foliated canopies (Li and Wang 2013; Wang and Li 2013). Moreover, previous studies demonstrated that canopies of similar foliage material (e.g., LAI or biomass) can yield significantly different canopy reflectance across the whole spectrum (Darvishzadeh et al. 2008). This observation has been linked to variation in confounding factors such as leaf trait heterogeneity within a canopy (Luo et al. 2016; Li et al. 2013). Related studies have also demonstrated that the “big leaf” (sunlit leaves only) upscaling approach underestimates canopy gross primary production by up to 70 % in highly clumped forest stands compared to the “two leaf” (sunlit plus shaded leaves) approach (Sprintsin et al. 2012). The “big leaf” approach does not account for the vertical variability in leaf traits and assumes that a sunlit leaf in the upper canopy represent the whole canopy, while the “two leaf” approach requires information on the vertical variability in leaf traits based on both sunlit and shaded leaves (Mercado et al. 2006). Within the framework of the “big leaf” and “two leaf” upscaling approaches, we explored different combinations of leaf trait-LAI upscaling scenarios from three vertical (upper, middle and bottom) canopy layers in short woody vegetation in a laboratory setup. To the best of our knowledge, the effect of these upscaling conceptualizations are poorly understood and remain untested in canopy traits estimation using *in-situ* hyperspectral measurements or simulated satellite data. To this end, we, therefore hypothesize that leaf to canopy upscaling approaches have a significant effect on the relationship between canopy traits and spectral measurements and subsequently estimation accuracy of canopy traits.

The principal aim of this study is to evaluate the effect of different approaches of upscaling leaf traits (foliar N, LMA, chlorophyll, and carbon) from leaf to canopy level on model performance and estimation accuracy using spectral measurements (*in-situ* canopy hyperspectral and simulated Sentinel-2 multispectral data) and partial least squares regression. A number of upscaling approaches based on different LAI and leaf

trait mathematical combinations were computed and used to calibrate a PLSR model, with the intention of assessing the accuracy of canopy traits estimation. Prior to ascertaining the effect of upscaling approaches, we tested whether leaf traits significantly vary across the canopy vertical profile.

2. Materials and methods

2.1. Species description

Four plant species of different leaf form and canopy structure, representing tropical and temperate biomes, were used to evaluate the effect of leaf to canopy upscaling approaches on model performance and estimation accuracy using *in-situ* hyperspectral and simulated Sentinel-2 multispectral data. We selected the following plant species; *Camellia japonica* (n = 24, mean height 83.71 ± 4.75 cm), *Ficus benjamina* (n = 24; mean height 82.07 ± 4.27 cm), *Chamaedorea elegans* (n = 24; mean height 88.5 ± 4.75 cm) and *Fatsyhedera lizei* (n = 24; mean height 88.93 ± 1.53 cm). Further description of the plant species used in this study can be found in Gara et al. (2018). All the plants used in this study were purchased from a local nursery.

2.2. Experimental setup

Since the plants purchased from the nursery were assumed to be pretreated with fertilizer, we changed the pot-soil to a new homogenized mixture of seven parts of nutrient-poor sand soil to two parts of fertile loamy soil. After changing the potting mix, we administered three soil nitrogen treatments (high, medium and low) to the ninety-six (96) plants used in this study. For the high (n = 32) and medium (n = 32) treatment groups, 2.9 g and 0.9 g per pot were supplied respectively, while no fertilizer was applied to the low treatment (n = 32). Further details on the experimental setup is provided in Gara et al. (2018)

2.3 Canopy spectral measurements

Canopy spectral reflectance from 350 to 2500 nm were measured in a controlled remote sensing laboratory using an ASD FieldSpec-3 Pro FR spectrometer. The walls and ceiling of the laboratory were coated with black material in order to minimize any ambient light or reflection, thus minimizing the effect of diffuse radiation and lateral flux (Darvishzadeh et al. 2009). Three pots of the same species and soil treatment were used to form a canopy as shown in a schematic presentation in Figure 1A. In order to create a canopy, the three pots were placed in fixed positions within a 60 cm by 60 cm soil bed. A fiber optic probe with a field of view of 25° was mounted on a tripod at nadir and positioned 90 cm above the soil bed, thus creating a field of view (FOV) with a diameter of 40 cm on the soil surface. A halogen light bulb (235 W) positioned 1.2 m from the canopy was used to supply illumination on the canopy. All canopy spectral measurements were calibrated with a Spectralon white reference panel. The sensor's FOV was completely covered with foliage material; hence the effect of the background soil was minimized. After each canopy spectral measurement, the soil bed was rotated 45° in order to average out the differences in canopy orientation and to minimize the possible bidirectional reflectance distribution function (BRDF) effects (Darvishzadeh et al. 2008, 2009). Consequently, the spectral reflectance of 32 canopies were measured. To minimize noise in the canopy reflectance spectra, a moving second order Savitzky-

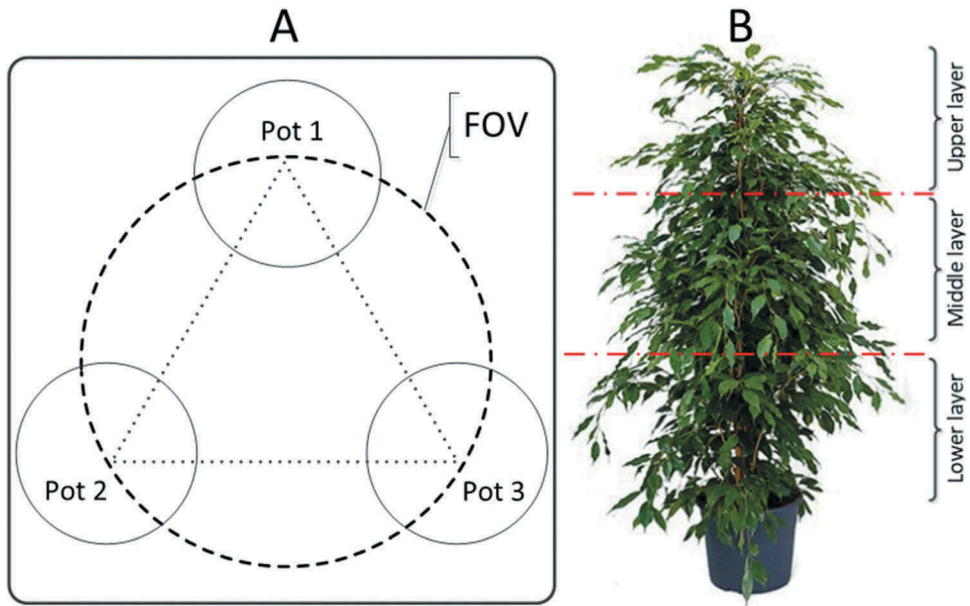


Figure 1. Positioning of plant vessels in the field of view (A) and the demarcation of the three canopy layers considered in the experiment (B).

Golay filter (Savitzky and Golay 1964) with a frame size of 11 was applied to each sample reflectance spectra. Wavelengths before 400 nm and after 2200 nm were too noisy and were thus removed from the dataset. Therefore, 1801 spectral bands were retained for subsequent analysis.

The canopy reflectance spectra measured using the ASD spectroradiometer were convolved to the spectral band configurations of Sentinel-2 multispectral instrument based on the sensor spectral response function. To account for environmental and instrument uncertainties in natural satellite operating system, we added a random Gaussian (white) noise component of 20% to the convolved spectra (Verrelst et al. 2014; Richter et al. 2009). Sentinel-2 is a Multi-Spectral Instrument (MSI) operated by the European Space Agency (ESA). The Multi-Spectral Instrument (MSI) on board Sentinel-2 is composed of 13 spectral bands ranging from 400 to 2400 nm with a grain size ranging from 10–60 m and a swath width of 290 km (European Space Agency 2010; Hill 2013). The mission has two identical multispectral sensors (Sentinel 2A and 2B) in orbit delivering a revisit time of three to five days (Aschbacher and Milagro-Pérez 2012)

2.4 Determining LAI and leaf traits

After measuring canopy reflectance spectra, the canopy was divided into three vertical layers, i.e. upper, middle and lower, as shown in Figure 1B. The three layers were determined based on the height of a canopy along the stem. We divided the canopy into three layers for three reasons. First, the plants used in this study were relatively short (mean height ~ 85 cm) in order to clearly identify more than three canopy layers. Second, identifying less than three canopy layers could have been insufficient to understand the effect of leaf traits vertical heterogeneity

on canopy traits estimation. Finally, three canopy layers match forestry and agronomic standards in reporting research in canopy layers (Whitehurst et al. 2013; Wilkes et al. 2016). Foliage material belonging to each canopy layer were harvested. The LAI for each canopy layer was measured as the cumulative leaf surface area of leaves making up a canopy layer divided by the field of view. Care was taken to eliminate leaves outside the field of view. The total surface area of the leaves were measured using a LI-3100C area meter. The calibration of the LI-3100C area meter was routinely checked against a metal surface of known surface area. Representative leaves (approximately 3 g) from foliage material of each canopy layer were randomly sampled for leaf traits measurement. Leaf chlorophyll content (C_{ab} $\mu\text{g}/\text{cm}^2$) was measured using a CCM-300 chlorophyll content meter (Opti-Sciences 2011). After measuring chlorophyll, a digital scale at an accuracy of ~ 0.01 g was used to determine the fresh weight for each sample. We also scanned the leaf surface area (LA cm^2) of each sample using an AMH 350 area meter (ADC-BioScientific 2013). The samples were then oven-dried at 65°C until at constant weight was attained after approximately 72 hours after which dry weight was measured. The leaf mass per area (LMA, g/cm^2) was determined by dividing dry weight by fresh leaf area.

After determining LMA, leaf samples were prepared for nitrogen and carbon analysis by grinding them to a fine and homogeneous powder using a mortar and pestle to pass through a $180\ \mu\text{m}$ sieving and mesh screen. Approximately 2 mg of each sample powder was placed in aluminium capsules for nitrogen and carbon analysis using the Perkin Elmer 2400 CHNS/O Elemental Analyzer (Perkin-Elmer 2005). We duplicated 25% of samples and ran an acetanilide standard after every ten to fifteen samples to constantly monitor the system calibration and reliability. The nitrogen and carbon results from the elemental analyzer were obtained on a dry mass ash-included basis (Meerdink et al. 2016). We therefore multiplied the mass-based nitrogen and carbon by LMA of each sample to obtain area based nitrogen and carbon content (Wang et al. 2015).

2.5 Upscaling leaf traits to canopy level

Using the LAI of each canopy layer, we explored different approaches for upscaling foliar nitrogen, LMA, chlorophyll and carbon to canopy level as described in Table 1. It is important to note that although the study aimed to assess five primary upscaling approaches (i.e., A-E, Table 1), we also used several secondary upscaling approaches (F-L) to explore the relative effect of each layer on the spectral signal observed at the top of the canopy, even if these secondary approaches are not necessarily practical in terms of field sampling. For example, upscaling option F requires measuring LAI of the top and bottom canopy layers, which is a challenge especially in environments characterized by tall trees. However, such an upscaling approach assists in assessing the contribution of the middle layer.

2.6 Statistical analysis

The effect of different leaf to canopy upscaling approaches on canopy traits estimation were assessed using partial least squares regression (PLSR) and validated using repeated *k-fold* cross validation. In this study we used PLSR for the following reasons; 1) it is more stable and suitable for high dimensional and collinear datasets such as a hyperspectral dataset (Wold, Sjöström, and Eriksson 2001); 2) it reduces model overfitting by decomposing spectral data into non-collinear latent variables especially when the number

Table 1. Leaf to canopy upscaling approaches.

Upscaling approach ID	Description	Formula
A	mean leaf traits across the three canopy layers × total canopy LAI	$\left(\frac{1}{n} \sum_{i=1,2,3} trait\right) * \sum_{1,2,3} LAI$
B	mean leaf traits of the top two canopy layers × total canopy LAI	$\left(\frac{1}{n} \sum_{i=1,2} trait\right) * \sum_{1,2,3} LAI$
C	leaf traits of the top canopy layer × total canopy LAI	$trait_1 * \sum_{1,2,3} LAI$
D	canopy traits weighted by LAI of each of the three canopy layers	$\sum_{i=1,2,3} LAI_i * trait_i$
E	canopy traits weighted by LAI of the top two layers	$\sum_{i=1,2} LAI_i * trait_i$
F	canopy traits weighted by the LAI of the top and bottom canopy layers	$\sum_{i=1,3} LAI_i * trait_i$
G	canopy traits weighted by LAI of the middle and bottom canopy layer	$\sum_{i=2,3} LAI_i * trait_i$
H	leaf traits for the top canopy layer × LAI of the top canopy layer	$LAI_1 * trait_1$
I	leaf traits for the middle canopy layer × LAI of the middle canopy layer	$LAI_2 * trait_2$
J	leaf traits of the bottom canopy layer × LAI of the bottom layer	$LAI_3 * trait_3$
K	mean leaf traits of the top and bottom layers × total canopy LAI	$\left(\frac{1}{n} \sum_{i=1,3} trait\right) * \sum_{1,2,3} LAI$
L	mean leaf traits of the middle and bottom canopy layers × total canopy LAI	$\left(\frac{1}{n} \sum_{i=2,3} trait\right) * \sum_{1,2,3} LAI$

1, 2, and 3 represents upper, middle and lower canopy layers respectively, trait = leaf traits

of predictor variables is more than the number of observations (Carrascal, Galván, and Gordo 2009); 3) it provides an opportunity to generate an error matrix for the internal validation subset (Wakeling and Morris 1993; Kuhn 2008); 4) it is computationally fast and statistically efficient and is widely used in leaf traits estimation using spectral data (Ramoelo et al. 2011; Neinavaz et al. 2016; Roelofsen et al. 2013; Serbin et al. 2014; Shiklomanov et al. 2016; Ullah et al. 2014).

PLSR projects the explanatory variables (canopy reflectance spectra) into new orthogonal latent variables that explain the most variance in the original predictors (Geladi and Kowalski 1986). The dependent variable (canopy traits) is then regressed against the optimal number of latent variables (Wold, Sjöström, and Eriksson 2001). The number of latent factors selected for the PLSR model was determined by minimizing the cross validated root mean square error ($RMSE_{cv}$) generated from the repeated *k-fold* cross validation. In order to avoid overfitting and maintain model parsimony, we restricted the number of latent factors to a maximum of 10% (three) of the sample size i.e. 32 canopies (Bian et al. 2010; Wold, Sjöström, and Eriksson 2001; Marcoulides and Saunders 2006). Repeated *k-fold* cross validation was used to proficiently exploit on our small dataset ($n = 32$). Five folds repeated 20 times (100 iterations) were used in the cross validation procedure. The repeated *k-fold* cross validation procedure iteratively splits the data set ($n = 32$) into five semi-equal partitions or blocks randomly. At each iteration, $k-1$ partitions were used to train the PLSR model while the left out partition were used for validation. This process was repeated iteratively k times (100 times in our case) until all the partitions were used for validation as well for calibration. In order to minimize bias on the estimate based on the composition of samples making up each partition the resampling procedure of the dataset was repeated twenty times. In the end, a total of 100 model runs were performed for each canopy trait.

The cross validated R^2_{cv} , and normalized RMSE ($nRMSE_{cv} = RMSE_{cv}/range$) between the predicted and measured canopy trait values were used to evaluate the performance of each PLSR model for each leaf to canopy upscaling approach. For canopy chlorophyll content modelling we used canopy reflectance spectra within the visible and red edge region (400-790nm) as this spectral region is sensitive to variation in chlorophyll content (Kumar et al. 2001). However, for the other canopy traits (N, LMA and carbon) we used the whole spectrum (400-2200nm) as these traits do not have a defined spectral feature and are related to other compounds distributed across the whole spectrum (Curran 1989).

To evaluate the effect of different leaf to canopy upscaling approaches on canopy traits estimation, we compared the mean $nRMSE_{cv}$ of the 100 model runs for each upscaling approach using one-way ANOVA with the upscaling approach as a fixed factor. We then used Tukey's HSD *post hoc* test to perform a pairwise comparison of the upscaling approaches. Prior to the PLSR modelling process, independent variables (spectra) and dependent (leaf traits) variables were mean-centered. All PLSR analyses were performed in R 3.3.3 for Windows using the classification and regression (caret) package (Kuhn 2008).

3. Results

3.1 Exploratory data analysis of foliar traits and in-situ hyperspectral data across the canopy vertical profile

Foliar N, chlorophyll and carbon content significantly ($p < 0.05$) increased from the lower to the upper canopy layer, while the LAI of the middle canopy layer was significantly ($p < 0.05$) higher compared to LAI of the lower and upper canopy layers

(Figure 2). LMA generally increased from lower to upper layers. However, LMA did not significantly ($p > 0.05$) vary across the three canopy layers.

Table 2 shows the descriptive statistics of the four canopy traits based on the different leaf to canopy upscaling approaches described in Table 1. Canopy N ($F = 6.21$, $p = 0.00$), canopy LMA ($F = 10.24$, $p = 0.00$), canopy chlorophyll ($F = 16.5$, $p = 0.00$), canopy carbon ($F = 13.48$, $p = 0.00$) significantly varied across the upscaling approaches based on a one-way ANOVA test. Moreover, the range of canopy traits computed from each of the upscaling approaches were different (Table 2). For example, the canopy traits based on top of canopy trait content (upscaling approach C – in bold, Table 2) had a limited range compared to the range of the other primary upscaling approaches especially for canopy N, LMA and carbon. The range of canopy traits could potentially affect the strength of the relationship between canopy spectral measurements and canopy traits. The mean canopy spectral reflectance flanked by the standard deviation for the thirty-two canopies is shown in Figure 3. Increased variations in the canopy spectral reflectance can be observed in the NIR (750–1350 nm) and SWIR (1400–1850 nm)

3.2 Effect of upscaling approach on model prediction using in-situ canopy hyperspectral measurements

Figure 4 shows prediction accuracies ($nRMSE_{cv}$) of the twelve upscaling approaches for canopy N, LMA, chlorophyll and carbon based on the repeated k -fold PLSR modelling for both in-situ hyperspectral measurements and simulated Sentinel-2 multispectral data. An analysis of the primary upscaling approaches indicate that upscaling approach A

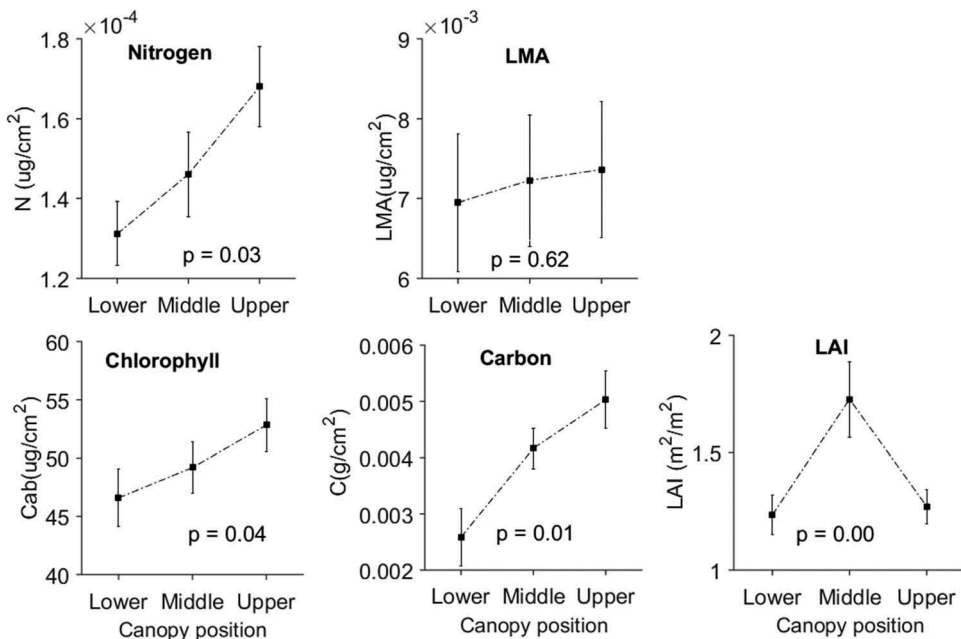


Figure 2. Variation in leaf traits and LAI across the canopy vertical profile.

Table 2. Summary statistics of the leaf to canopy upscaling approaches.

		Leaf to canopy upscaling approach											
		A	B	C	D	E	F	G	H	I	J	K	L
Canopy N (g/m ²)	min	0.79	0.97	1.04	0.69	0.48	0.5	0.29	0.28	0.12	0.09	0.8	0.49
	max	20.24	21.9	16.92	22.88	19.53	9.23	19.2	5.75	15.88	6.47	16.61	22.21
	mean	6.95	7.55	6.95	7.33	5.69	3.65	5.31	2.01	3.68	1.63	6.3	6.95
	SD	5.6	6.2	5.16	6.06	5.21	2.63	4.8	1.59	3.93	1.53	4.74	6.12
Canopy LMA (g/m ²)	min	9.62	10.77	12.25	9.83	6.13	6.88	5.14	3.36	2.18	2.11	9.41	8.3
	max	78.47	79.24	71.41	81.53	66.33	37.39	65.41	19.86	50.21	17.53	74.17	82
	mean	30.23	30.89	30.31	30.69	22.77	16.85	21.75	8.93	13.83	7.92	26.61	30.19
	SD	19.13	19.85	17.83	19.7	16.54	8.59	15.4	5.26	12	4.95	17.8	30.11
Canopy Cab (g/m ²)	min	1.14	1.15	1.13	1.14	0.71	0.77	0.65	0.26	0.33	0.14	1.13	1.15
	max	5.13	5.17	5.31	5.12	4.04	2.66	3.53	1.59	2.56	1.11	5.18	5.03
	mean	2.18	2.19	2.17	2.19	1.58	1.24	1.56	0.63	0.95	0.61	2.17	2.19
	SD	1.08	1.10	1.13	1.08	0.91	0.45	0.87	0.28	0.69	0.26	1.08	1.05
Canopy Carbon (g/m ²)	min	1.21	1.58	1.68	1.11	0.77	0.8	0.46	0.46	0.2	0.14	1.28	0.74
	max	79.21	83.87	61.17	89.07	75.26	32.05	75.26	18.13	61.45	14.19	65.54	88.23
	mean	18.64	20.09	17	19.76	15.69	9.1	14.72	5.03	10.65	4.07	16.36	19.45
	SD	22.63	24.73	18.53	24.45	20.71	9.5	19.37	5.77	15.55	4.64	18.46	25.16

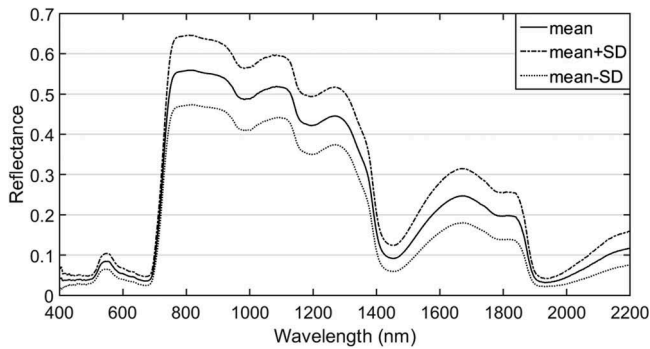


Figure 3. The mean and standard deviation of canopy spectral reflectance for the thirty-two canopies used for analysis.

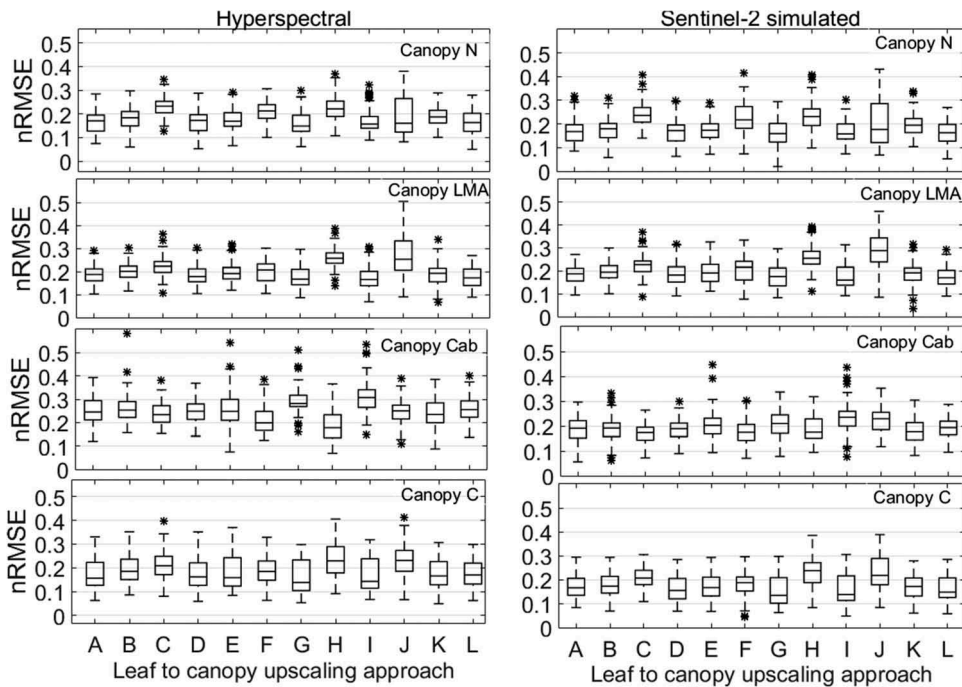


Figure 4. Prediction accuracies ($nRMSE_{cv}$) of each upscaling approach for the four traits. The boxplots are based on the repeated k -fold cross validation PLSR 100 iterations.

(mean leaf traits across the three canopy layers \times total canopy LAI), B (mean traits of the top two canopy layers \times total canopy LAI), D (leaf traits weighted by the LAI of each canopy layer) and E (leaf traits weighted by the LAI of the top two canopy layers) consistently yielded higher retrieval accuracy ($nRMSE_{cv} < 0.2$) and higher explained variance (Table 3) compared to upscaling approach C (upper of the canopy traits \times total canopy LAI; $nRMSE_{cv} = 0.23, 0.22,$ and 0.22 for canopy N, LMA and carbon, respectively). It is worthwhile to note that upscaling approach “A” exhibited relatively higher

Table 3. Performance of PLSR reflectance models based on *in-situ* hyperspectral measurements.

Upscaling approach	Canopy N		Canopy LMA		Canopy chlorophyll		Canopy Carbon	
	nlv	R ² _{cv} ±SD	nlv	R ² _{cv} ±SD	nlv	R ² _{cv} ±SD	nlv	R ² _{cv} ±SD
A	2	0.78 ± 0.15	2	0.66 ± 0.18	2	0.65 ± 0.27	3	0.76 ± 0.11
B	2	0.76 ± 0.16	2	0.65 ± 0.19	2	0.64 ± 0.28	3	0.77 ± 0.12
C	2	0.60 ± 0.22	2	0.58 ± 0.17	2	0.68 ± 0.25	2	0.71 ± 0.14
D	2	0.79 ± 0.15	2	0.67 ± 0.17	2	0.64 ± 0.27	3	0.79 ± 0.14
E	2	0.73 ± 0.14	2	0.63 ± 0.16	3	0.57 ± 0.28	3	0.74 ± 0.1
F	2	0.64 ± 0.18	2	0.6 ± 0.21	2	0.61 ± 0.3	3	0.76 ± 0.17
G	2	0.74 ± 0.2	2	0.65 ± 0.22	2	0.64 ± 0.26	3	0.78 ± 0.18
H	2	0.57 ± 0.24	2	0.48 ± 0.2	2	0.38 ± 0.27	2	0.64 ± 0.2
I	2	0.68 ± 0.16	2	0.63 ± 0.24	2	0.56 ± 0.25	3	0.74 ± 0.16
J	2	0.49 ± 0.23	1	0.51 ± 0.29	2	0.5 ± 0.26	3	0.66 ± 0.24
K	2	0.62 ± 0.19	2	0.54 ± 0.16	2	0.64 ± 0.23	3	0.62 ± 0.11
L	2	0.7 ± 0.16	2	0.6 ± 0.16	2	0.6 ± 0.27	3	0.72 ± 0.15

nlv: number of latent variables, SD standard deviation

model stability based on the limited range of the yielded nRMSE_{cv}. A Tukey HSD *post hoc* test demonstrated that the nRMSE_{cv} distribution obtained from upscaling approach C (leaf traits of the upper canopy layer × total canopy LAI) was significantly ($p < 0.05$) higher compared to the nRMSE_{cv} obtained from the other primary upscaling approaches especially for canopy N, LMA and canopy carbon (Figure 5). The nRMSE_{cv} generated from upscaling approaches A, B, D and E for canopy N, LMA and carbon were not significantly ($p > 0.05$) different from each other demonstrating that they yield comparable retrieval accuracies and ultimately outperformed upscaling approach C.

By contrast, canopy chlorophyll content exhibited a different pattern compared to the other canopy traits estimation models. Canopy chlorophyll content upscaled based on leaf samples collected from the upper canopy and total canopy LAI (upscaling approach C) exhibited a better relationship ($R^2_{cv} = 0.68$, nRMSE_{cv} = 0.24) against *in-situ* canopy hyperspectral measurements compared to other canopy chlorophyll upscaling approaches (Figure 4, Table 3). It is important to note that although upscaling approach C produced a higher retrieval accuracy in canopy chlorophyll estimation; it yielded comparable prediction errors to other primary upscaling approaches i.e. the generated (nRMSE_{cv}) were not statistically different ($p > 0.05$, Figure 5). Although there were no statistical differences in the generated nRMSE_{cv} among the canopy chlorophyll estimations, it is worthwhile to note that all leaf to canopy upscaling approaches generated high errors (nRMSE_{cv} > 0.23) compared to canopy N, LMA and carbon using *in-situ* hyperspectral measurements.

Upscaling approaches that included both LAI or traits of the middle layer (i.e. A, B, D, E, G, I and L) yielded low nRMSE_{cv} especially for canopy N, LMA and carbon. Although the middle canopy layer demonstrated a strong influence on retrieval accuracy of canopy traits, a combination of the middle and upper canopy layers (upscaling approach R²_{cv}: A = 0.78, B = 0.76, D = 0.79 and E = 0.73) generally outperforms the combination of middle and bottom canopy layer (R²_{cv}: G = 0.74, L = 0.7) for canopy N estimations (Table 3) using hyperspectral measurements. However, upscaling approaches that excludes the middle canopy layer yielded lower explained variance e.g. (R²_{cv}: F = 0.64, K = 0.62). We observed that upscaling approach B (mean traits of the top

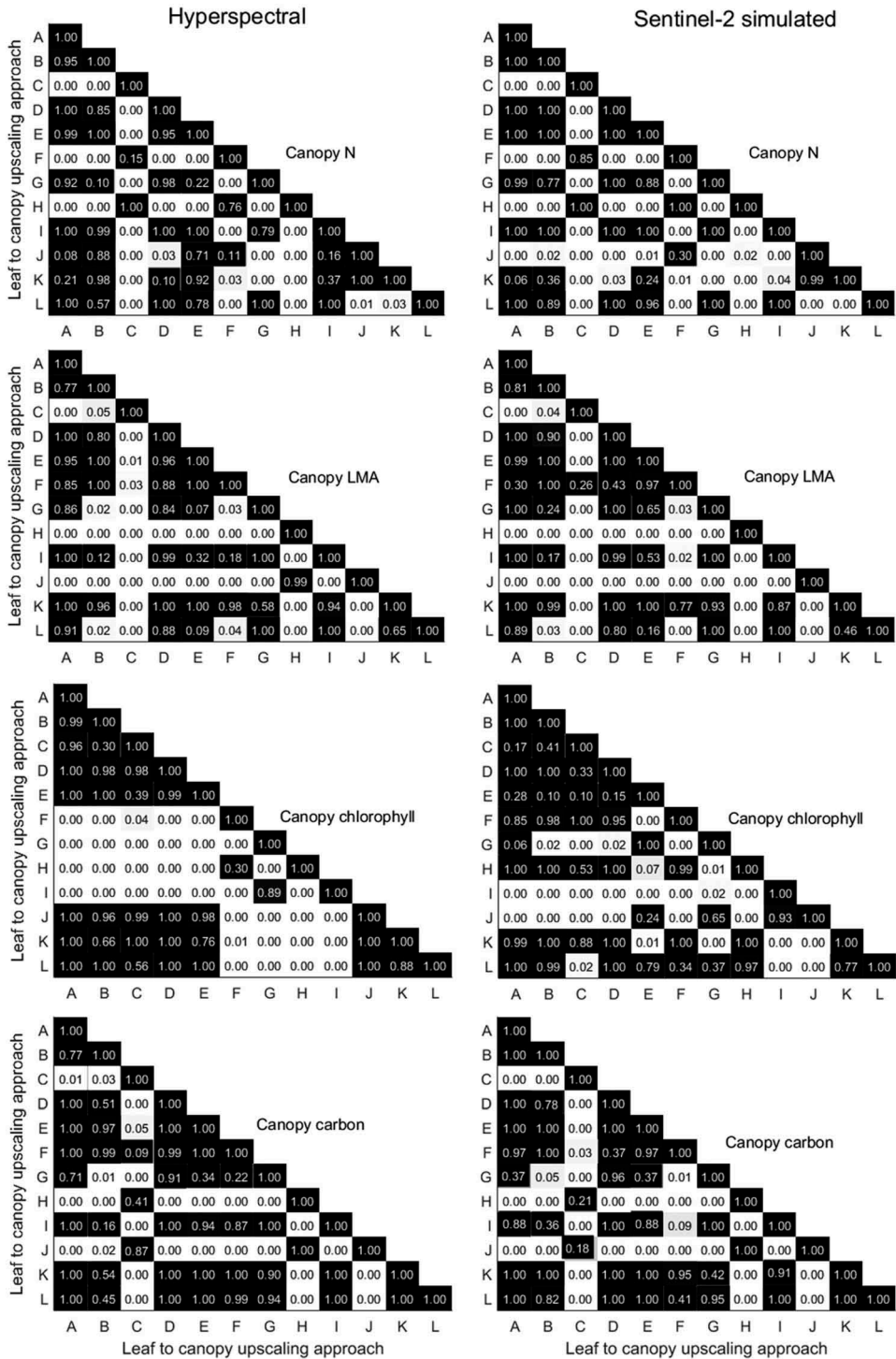


Figure 5. Pairwise comparison of the upscaling approaches based on the *Tukey's HSD post hoc* test. Values in each cell indicate the p-value of each pairwise comparison. White and black cells represent significant and non-significant pairwise comparison, respectively ($\alpha = 0.05$).

two layers × total canopy LAI), K (mean traits of the top and bottom layers × total canopy LAI) and L (mean traits of the middle and bottom layers × total canopy LAI), – all computed based on total canopy LAI, yielded different explained variance (R^2_{cv} : B = 0.65, K = 0.64 and L = 0.7) and prediction errors (nRMSE_{cv}: B = 0.2, K = 0.19 and L = 0.17) for canopy LMA estimations. A similar pattern can also be observed regarding the relationship between canopy N or carbon and *in-situ* hyperspectral measurements. For practical purposes, this implies that leaf traits and LAI of the top two canopy layers are key and contribute significantly to canopy spectral reflectance.

3.3 Effect of upscaling approach on model prediction using simulated sentinel 2 data

Generally, upscaling option C yielded the lowest retrieval accuracy for canopy N (nRMSE_{cv} = 0.23), LMA (nRMSE_{cv} = 0.22) and C (nRMSE_{cv} = 0.21) compared to the other primary upscaling approaches (A, B, D and E) (see Figure 4) using simulated Sentinel-2 dataset. The nRMSE_{cv} generated from upscaling approach C were significantly ($p < 0.05$) different from nRMSE_{cv} generated the other primary upscaling approaches (A, B, D and E) in canopy N, LMA and carbon estimations (Figure 5). No significant difference ($p > 0.05$) in nRMSE_{cv} were observed between upscaling options A, B, D and E implying they yield comparable results for canopy N, LMA and carbon estimations. The generated R^2_{cv} , confirmed that upscaling approaches A, B, D and E outperformed upscaling approach C for canopy N, LMA and carbon estimations (Table 4). In contrast, upscaling approach C yielded the highest retrieval accuracy for canopy chlorophyll estimation compared to upscaling approaches A, B, D and E. On average, upscaling approach C yielded the lowest nRMSE (nRMSE_{cv} = 0.17; Figure 4) and highest R^2 ($R^2_{cv} = 0.62$; Table 4) compared to other upscaling options for canopy chlorophyll estimation. However, the generated nRMSE_{cv} were not statistically significantly different ($p > 0.05$) between upscaling approach C and the other primary upscaling approaches i.e. A, B, D and E (Figure 5).

Similar to the results observed for canopy trait estimation using *in-situ* hyperspectral measurements, functional attributes (leaf traits and LAI) of the top two canopy layers

Table 4. Performance of PLSR reflectance models calibrated based simulated Sentinel-2.

Upscaling approach	Canopy N		Canopy LMA		Canopy chlorophyll		Canopy Carbon	
	nlv	$R^2_{cv} \pm SD$	nlv	$R^2_{cv} \pm SD$	nlv	$R^2_{cv} \pm SD$	nlv	$R^2_{cv} \pm SD$
A	3	0.73 ± 0.15	3	0.62 ± 0.17	3	0.59 ± 0.24	2	0.73 ± 0.19
B	3	0.73 ± 0.16	3	0.62 ± 0.19	3	0.56 ± 0.28	2	0.73 ± 0.15
C	3	0.53 ± 0.2	3	0.53 ± 0.21	3	0.62 ± 0.2	2	0.67 ± 0.17
D	3	0.71 ± 0.18	3	0.59 ± 0.23	3	0.57 ± 0.23	2	0.75 ± 0.18
E	3	0.68 ± 0.16	2	0.57 ± 0.21	3	0.5 ± 0.24	2	0.70 ± 0.15
F	2	0.52 ± 0.27	3	0.51 ± 0.27	3	0.55 ± 0.24	2	0.7 ± 0.18
G	3	0.67 ± 0.24	3	0.59 ± 0.25	3	0.57 ± 0.24	2	0.75 ± 0.16
H	3	0.49 ± 0.22	2	0.43 ± 0.25	3	0.35 ± 0.26	2	0.6 ± 0.19
I	3	0.64 ± 0.17	3	0.57 ± 0.21	3	0.5 ± 0.27	2	0.71 ± 0.18
J	2	0.39 ± 0.32	2	0.36 ± 0.26	3	0.42 ± 0.25	2	0.61 ± 0.25
K	3	0.6 ± 0.17	3	0.5 ± 0.2	3	0.57 ± 0.25	2	0.52 ± 0.18
L	3	0.70 ± 0.15	3	0.56 ± 0.19	3	0.53 ± 0.27	3	0.66 ± 0.15

nlv: number of latent variables, SD standard deviation

imposed a strong influence on canopy N, LMA and carbon estimations using simulated Sentinel-2 data. For example, upscaling approach I (leaf traits and LAI of the middle canopy layer) outperformed upscaling approaches H (leaf traits and LAI of the top canopy layer) and J (leaf traits of the bottom canopy layer and \times LAI of the bottom canopy layer) for canopy N, LMA and carbon estimation (Figure 4). For canopy chlorophyll estimation upscaling approach H ($nRMSE_{cv} = 0.18$) outperformed upscaling approach I ($nRMSE_{cv} = 0.23$) demonstrating that the top layer imposes a strong influence on canopy chlorophyll estimation from simulated Sentinel-2 data (Figure 4).

4. Discussion

This study set out to examine the effect of different approaches of upscaling foliar N, LMA, chlorophyll and carbon from leaf to canopy level on model performance and estimation accuracy using *in-situ* canopy hyperspectral measurements and simulated Sentinel-2 data. Results of this study demonstrate that leaf to canopy upscaling approaches have a profound effect on the estimation of canopy traits. In comparison to other upscaling approaches the widely-used product of top of canopy traits and total canopy LAI (upscaling approach C) consistently underperformed ($nRMSE_{cv} > 0.2$) compared to other primary upscaling approaches ($nRMSE_{cv} < 0.2$) that consider the contribution of leaf traits content from the shaded middle and lower canopy layers (Figure 4). This demonstrates that functional attributes (LAI and leaf traits) of the top canopy layer did not completely control the spectral reflectance observed by the sensor. Upscaling approaches that include functional attributes of the top and middle canopy layers (upscaling approaches B, E) significantly, ($p < 0.05$) improved the estimation accuracies of the canopy traits (Figure 5). However, upscaling approaches that considered functional attributes of the three layers (upscaling approach A and D) yielded comparable results to upscaling approaches that considered traits of the top two layers (upscaling B and E; Figures 4 and 5). The inclusion of functional attributes (leaf traits and LAI) of the lower canopy layer did not significantly improve the estimation accuracy of canopy traits. This demonstrates that canopy reflectance observed by a sensor is not necessarily generated by the entire canopy. The obscured foliage material of the lower canopy contributed less to the canopy spectral signal (Roelofsen et al. 2013). This observation can be linked to the problem of saturation in reflectance and vegetation indices with increasing amount of vegetation (Mutanga and Skidmore 2004; Prabhakara, Hively, and McCarty 2015). Saturation occurs when spectral reflectance or indices reach an asymptotic level beyond which any further increase in vegetation biomass or LAI does not result in a significant change on the spectral signal or index (Liang et al. 2015). This problem may lead to inaccurate and underestimation of canopy traits in high LAI or biomass environments (Thenkabail, Smith, and De Pauw 2000).

In contrast to observations made on canopy N, LMA and carbon, canopy chlorophyll estimations exhibited an improved relationship with upscaling approach C (leaf traits of the top canopy layer and total LAI) (Figure 4). Upscaling approach C yielded the highest R^2_{cv} and lowest $nRMSE_{cv}$ in canopy chlorophyll estimations for both *in-situ* hyperspectral measurements ($R^2_{cv} = 0.68$ and $nRMSE_{cv} = 0.24$) and simulated Sentinel-2 data ($R^2_{cv} = 0.62$ and $nRMSE_{cv} = 0.17$). The top layer proved to have a strong influence on canopy chlorophyll estimation for both spectral datasets. This observation concurs with previous studies (Verrelst et al. 2010; Gitelson et al. 2005) that demonstrate that canopy reflectance (especially in the visible spectrum) is strongly influenced by chlorophyll

content of the upper canopy layer due to strong chlorophyll sensitivity within the visible spectrum. However, the explained variance and model accuracy generated from upscaling approach C for canopy chlorophyll estimations for both spectral datasets were not significantly different ($p > 0.05$) from the other primary upscaling approaches that consider the contribution of the exposed top canopy layer together with the shaded middle and bottom layers (Figure 5). In this regard, estimation of both dry matter related traits (i.e. N, LMA and carbon) and leaf pigments such as chlorophyll need to consider the vertical variation in leaf traits for improved prediction and mapping of these traits at landscape and regional landscapes. It is important to note that a number of studies report a wide range of explained variances in leaf traits estimation ranging from as low as 46% for canopy N (Ramoelo et al. 2015) to as high as 92% for canopy chlorophyll (Clevers and Gitelson 2013) using in-situ hyperspectral measurements and simulated Sentinel-2 multispectral data. Most of these studies do not generally provide detailed description of how the leaf traits were upscaled to canopy level. Our results suggest that the approach used to upscale foliar traits from leaf-to-canopy level is a potential source of uncertainty in canopy trait estimation especially in less foliated vegetation biomes where foliage material from the lower canopy contributes to the canopy reflectance.

To understand the relative influence of leaf traits and LAI for each canopy layer on model performance and estimation accuracy, different upscaling combinations of foliar traits and LAI were explored (Table 1). Our results indicate that the middle layer had a key effect on the estimation of N, LMA and carbon from *in-situ* canopy hyperspectral measurements and simulated Sentinel-2 data. Leaf to canopy upscaling approaches that excluded functional attributes of the middle layer (e.g. upscaling approaches F, H and J) resulted in low explained variance ($R^2_{cv} = 0.6, 0.48$ and 0.51) and estimation accuracy ($nRMSE_{cv} = 0.2, 0.21$ and 0.27), compared to upscaling approaches such as G and I ($R^2_{cv} = 0.65$ and 0.63 and $nRMSE_{cv} = 0.15, 0.16$ respectively) that included functional attributes of the middle layer in canopy LMA estimation using in-situ hyperspectral measurements (Figure 4, Tables 3 and 4). A similar pattern was also observed in canopy N; carbon model estimations using either *in-situ* hyperspectral measurements or simulated Sentinel-2 data (Figure 4). This observation can be ascribed to the high LAI values of the middle layer (Figure 2), which ultimately controlled canopy radiation dynamics (Wang and Li 2013).

Results of this study also indicated the effect of leaf traits vertical heterogeneity on canopy traits estimation from spectral data. This was shown when different combination of leaf traits from the three canopy layers were explored whilst LAI remained invariant. For example, upscaling approach B (mean traits of the top two layers \times total canopy LAI), K (mean traits of the top and bottom layers \times total canopy LAI) and L (mean traits of the middle and bottom layers \times total canopy LAI) were all computed based on total canopy LAI, however they generated different explained variances and $nRMSE_{cv}$ for N, LMA and carbon. For instance, upscaling approaches B, K and L yielded R^2_{cv} of 0.76, 0.62 and 0.7 for *in-situ* hyperspectral measurements and 0.73, 0.6 and 0.7 for a simulated Sentinel-2 dataset for canopy N prediction models. The average of leaf traits of the top and middle canopy layers (B) resulted in a higher R^2_{cv} (0.76 and 0.73 for *in-situ* hyperspectral and simulated Sentinel-2 data), while the combination of the middle and bottom (upscaling approach L) resulted in explained variance dropping by 6% and 3% respectively for *in-situ* hyperspectral and simulated Sentinel 2 respectively. However, the exclusion of the leaf traits content of the middle layer (upscaling approach K) resulted in a further decrease in explained variance by 8 and 10% for *in-situ* hyperspectral and simulated Sentinel 2, respectively. A similar pattern can also be observed for canopy

LMA or carbon for both *in-situ* hyperspectral measurements and simulated Sentinel-2 data. This result indicates that leaf traits of the top and middle canopy layers together with their respective LAI drive canopy reflectance observed by the sensor. This result conforms to Wang and Li's (2013) observation that the vertical heterogeneity in leaf chlorophyll, water and dry matter content variation have a significant effect on simulated canopy reflectance. This observation is also in agreement with Luo et al. (2016) who demonstrated that the top three layers of ~ 2 meters high wetland reeds were key in canopy N prediction using vegetation indices computed from *in-situ* hyperspectral measurements. In this regard, the vertical variation in both LAI and leaf traits have an effect on the relationship between canopy reflectance and the canopy traits. This implies that large uncertainties can be presented in canopy parameter estimations if information on the vertical variation of key traits is not incorporated in the modelling approach. However, as LAI is often measured as total canopy LAI (Peng et al. 2017), significant consideration should, therefore be placed on leaf traits that vary across the canopy.

Results obtained in this study are similar to observations of earlier studies (Mercado et al. 2006; Coble et al. 2016; Sprintsin et al. 2012) that reported that the "big leaf" approach underestimates quantification of total canopy processes such as total canopy photosynthesis and gross primary productivity compared to the "two leaf" approach. The "big leaf" approach assumes that unshaded, sunlit leaves in the upper canopy represents the whole canopy metabolic processes, while the "two leaf" approach accounts for canopy metabolic processes based on both sunlit and shaded leaves. Sprintsin et al. (2012) observed that gross primary production models calibrated based on the big leaf upscaling approach consistently yielded low explained variance and accuracy in gross primary productivity modelling across different vegetation biomes against flux tower measurements. Their study demonstrated that the exclusion of the photosynthetic contribution of the shaded leaves could underestimate canopy gross photosynthesis productivity by over 70% in highly clumped vegetation stands. In light of this background, the "big leaf" is closely related to upscaling approach C that only considered leaf traits of the upper layer. The "two leaf" approach closely resembles the other primary upscaling approaches (A, B, D and E) that factor in the contribution of foliage material from both the exposed upper layers and the shaded lower layers. This observation demonstrates that canopy reflectance observed by a sensor constitutes contribution of all foliage material within the canopy.

Importantly, our study examines the effect of upscaling leaf traits from the leaf to canopy level using *in-situ* canopy hyperspectral and simulated Sentinel 2 data. Results obtained in this study can be tested in forests or woodlands where foliage from the lower canopy contributes to canopy reflectance. Partitioning the total canopy LAI into sunlit and shaded layers following methods such as those proposed by Chen et al. (1999) and subsequently collecting leaf samples from both sunlit and shaded layers are critical in examining the contribution of each canopy layer in canopy trait estimations. Hence, the effect of separating LAI and leaf samples into sunlit and shaded on canopy traits estimation using airborne or satellite data need further investigation.

5. Conclusion

In this study, we evaluated the effect of different approaches of upscaling leaf traits to the canopy level on the accuracy of estimation of canopy N, LMA, chlorophyll and carbon from *in-situ* hyperspectral measurements and simulated Sentinel-2 reflectance data.

Through a robust sampling procedure, we determined leaf traits from different vertical canopy positions and applied various weighted averages to examine how various canopy components impact the estimation of canopy traits from *in-situ* canopy hyperspectral and simulated Sentinel 2 data. Based on the results we conclude that:

- (i) Leaf-to-canopy upscaling approaches yield significantly different canopy traits values. The range (max-min) of the canopy traits varies depending on the upscaling approach used.
- (ii) Leaf-to-canopy upscaling approaches that consider the contribution of both the exposed upper canopy leaves together with the shaded lower canopy leaves results in improved prediction of canopy nitrogen, LMA and carbon from both *in-situ* canopy hyperspectral measurements and simulated Sentinel-2 data. However, the same pattern does not hold true for canopy chlorophyll.
- (iii) The widely used upscaling approach that considers leaf traits from the exposed top of the canopy yields a better accuracy for canopy chlorophyll estimation from *in-situ* canopy hyperspectral measurements. However, the prediction errors obtained among the canopy chlorophyll upscaling approaches were not significantly different ($p > 0.05$).

We therefore, conclude that sampling methods that intend to use remote sensing measurements to upscale leaf traits, especially dry matter related leaf traits, need to account for the vertical heterogeneity in leaf traits across plant canopies for improved canopy traits estimation and mapping. As this study was conducted in the laboratory setup with relatively short shrubs, it is important to ascertain whether similar results can be obtained in high-foliated forests using airborne or satellite spectral measurements.

Acknowledgements

This study was carried out with the financial assistance of the Faculty of Geo-Information Science and Earth Observation (ITC), University of Twente. The authors extend their gratitude for the technical support during laboratory measurements from the staff of the Geosciences Laboratory at ITC faculty, University of Twente.

Disclosure statement

No potential conflict of interest was reported by the authors.

ORCID

Tawanda W. Gara  <http://orcid.org/0000-0001-8134-4849>

Andrew K. Skidmore  <http://orcid.org/0000-0002-7446-8429>

Roshanak Darvishzadeh  <http://orcid.org/0000-0001-7512-0574>

Tiejun Wang  <http://orcid.org/0000-0002-1138-8464>

References

ADC-BioScientific. 2013. *AM350 Portable Leaf Area Meter*. Hoddesdon: ADC BioScientific.

- Aschbacher, J., and M. P. Milagro-Pérez. 2012. "The European Earth Monitoring (GMES) Programme: Status and Perspectives." *Remote Sensing of Environment* 120: 3–8. doi:10.1016/j.rse.2011.08.028.
- Bian, M., A. K. Skidmore, M. Schlerf, T. Fei, Y. Liu, and T. Wang. 2010. "Reflectance Spectroscopy of Biochemical Components as Indicators of Tea (*Camellia Sinensis*) Quality." *Photogrammetric Engineering & Remote Sensing* 76 (12): 1385–1392. doi:10.14358/pers.76.12.1385.
- Carrascal, L. M., I. Galván, and O. Gordo. 2009. "Partial Least Squares Regression as an Alternative to Current Regression Methods Used in Ecology." *Oikos* 118 (5): 681–690. doi:10.1111/j.1600-0706.2008.16881.x.
- Chemura, A., O. Mutanga, J. Odindi, and D. Kutuywayo. 2018. "Mapping Spatial Variability of Foliar Nitrogen in Coffee (*Coffea Arabica* L.) Plantations with Multispectral Sentinel-2 MSI Data." *ISPRS Journal of Photogrammetry and Remote Sensing* 138: 1–11. doi:10.1016/j.isprsjprs.2018.02.004.
- Chen, J.-L., J. F. Reynolds, P. C. Harley, and J. D. Tenhunen. 1993. "Coordination Theory of Leaf Nitrogen Distribution in a Canopy." *Oecologia* 93 (1): 63–69. doi:10.1007/bf00321192.
- Chen, J. M., J. Liu, J. Cihlar, and M. L. Goulden. 1999. "Daily Canopy Photosynthesis Model through Temporal and Spatial Scaling for Remote Sensing Applications." *Ecological Modelling* 124 (2): 99–119. doi:10.1016/S0304-3800(99)00156-8.
- Cho, M. A., A. Ramoelo, P. Debba, O. Mutanga, R. Mathieu, H. van Deventer, and N. Ndlovu. 2013. "Assessing the Effects of Subtropical Forest Fragmentation on Leaf Nitrogen Distribution Using Remote Sensing Data." *Landscape Ecology* 28 (8): 1479–1491. doi:10.1007/s10980-013-9908-7.
- Clevers, J. G. P. W., and A. A. Gitelson. 2013. "Remote Estimation of Crop and Grass Chlorophyll and Nitrogen Content Using Red-Edge Bands on Sentinel-2 and -3." *International Journal of Applied Earth Observation and Geoinformation* 23: 344–351. doi:10.1016/j.jag.2012.10.008.
- Coble, A. P., B. VanderWall, A. Mau, and M. A. Cavaleri. 2016. "How Vertical Patterns in Leaf Traits Shift Seasonally and the Implications for Modeling Canopy Photosynthesis in a Temperate Deciduous Forest." *Tree Physiology* 36: 1077–1091. doi:10.1093/treephys/tpw043.
- Croft, H., J. M. Chen, Y. Zhang, A. Simic, T. L. Noland, N. Nesbitt, and J. Arabian. 2015. "Evaluating Leaf Chlorophyll Content Prediction from Multispectral Remote Sensing Data within a Physically-Based Modelling Framework." *ISPRS Journal of Photogrammetry and Remote Sensing* 102: 85–95. doi:10.1016/j.isprsjprs.2015.01.008.
- Curran, P. J. 1989. "Remote Sensing of Foliar Chemistry." *Remote Sensing of Environment* 30 (3): 271–278. doi:10.1016/0034-4257(89)90069-2.
- Daniel, K. W., A. Ahumada Jorge, A. Bowser, M. Fernandez, N. Fernández, E. P. Alonso García, G. Robert, et al. 2017. "Building Essential Biodiversity Variables (Ebvs) of Species Distribution and Abundance at a Global Scale." *Biological Reviews* 93 (1): 600–625. doi:10.1111/brv.12359.
- Darvishzadeh, R., C. Atzberger, A. K. Skidmore, and A. A. Abkar. 2009. "Leaf Area Index Derivation from Hyperspectral Vegetation Indices and the Red Edge Position." *International Journal of Remote Sensing* 30 (23): 6199–6218. doi:10.1080/01431160902842342.
- Darvishzadeh, R., A. Skidmore, C. Atzberger, and V. W. Sip. 2008. "Estimation of Vegetation LAI from Hyperspectral Reflectance Data: Effects of Soil Type and Plant Architecture." *International Journal of Applied Earth Observation and Geoinformation* 10 (3): 358–373. doi:10.1016/j.jag.2008.02.005.
- European Space Agency. 2010. "GMES Sentinel-2 Mission Requirements Document: Technical Report Issue 2 Revision 1." Accessed 17 April 2017. http://esamultimedia.esa.int/docs/GMES/Sentinel-2_MRD.pdf
- Gara, T., R. Darvishzadeh, A. Skidmore, and T. Wang. 2018. "Impact of Vertical Canopy Position on Leaf Spectral Properties and Traits across Multiple Species." *Remote Sensing* 10 (2): 346. doi:10.3390/rs10020346.
- Geladi, P., and B. R. Kowalski. 1986. "Partial Least-Squares Regression: A Tutorial." *Analytica Chimica Acta* 185: 1–17. doi:10.1016/0003-2670(86)80028-9.
- Gibson, D. J. 2008. *Grasses and Grassland Ecology*. New York: Oxford University Press.
- Gitelson, A. A., A. Viña, V. Ciganda, D. C. Rundquist, and T. J. Arkebauer. 2005. "Remote Estimation of Canopy Chlorophyll Content in Crops." *Geophysical Research Letters* 32 (8): 1–4. doi:10.1029/2005GL022688.

- He, Y., and A. Mui. 2010. "Scaling up Semi-Arid Grassland Biochemical Content from the Leaf to the Canopy Level: Challenges and Opportunities." *Sensors* 10 (12): 11072–11087. doi:10.3390/s101211072.
- Li, H., C. Zhao, W. Huang, and G. Yang. 2013. "Non-Uniform Vertical Nitrogen Distribution within Plant Canopy and Its Estimation by Remote Sensing: A Review." *Field Crops Research* 142: 75–84. doi:10.1016/j.fcr.2012.11.017.
- Hill, M. J. 2013. "Vegetation Index Suites as Indicators of Vegetation State in Grassland and Savanna: An Analysis with Simulated SENTINEL 2 Data for a North American Transect." *Remote Sensing of Environment* 137: 94–111. doi:10.1016/j.rse.2013.06.004.
- Hirose, T., M. J. A. Werger, and J. W. A. van Rheenen. 1989. "Canopy Development and Leaf Nitrogen Distribution in a Stand of *Carex Acutiformis*." *Ecology* 70 (6): 1610–1618. doi:10.2307/1938094.
- Homolová, L., Z. Malenovský, J. G. P. W. Clevers, G. García-Santos, and M. E. Schaepman. 2013. "Review of Optical-Based Remote Sensing for Plant Trait Mapping." *Ecological Complexity* 15: 1–16. doi:10.1016/j.ecocom.2013.06.003.
- Knyazikhin, Y., M. A. Schull, P. Stenberg, M. Möttus, M. Rautiainen, Y. Yang, A. Marshak, et al. 2013. "Hyperspectral Remote Sensing of Foliar Nitrogen Content." *Proceedings of the National Academy of Sciences* 110 (3): E185–E92. doi:10.1073/pnas.1210196109.
- Kokaly, R. F., G. P. Asner, S. V. Ollinger, M. E. Martin, and C. A. Wessman. 2009. "Characterizing Canopy Biochemistry from Imaging Spectroscopy and Its Application to Ecosystem Studies." *Remote Sensing of Environment* 113 (Supplement 1): S78–S91. doi:10.1016/j.rse.2008.10.018.
- Kuhn, M. 2008. "Building Predictive Models in R Using the Caret Package." *Journal of Statistical Software* 28 (5): 26. doi:10.18637/jss.v028.i05.
- Kumar, L., K. Schmidt, S. Dury, and A. Skidmore. 2001. "Imaging Spectrometry and Vegetation Science." In: *Imaging Spectrometry: Basic Principles and Prospective Applications*, edited by F. D. der van Meer and S. M. De Jong, 111–155. Dordrecht: Springer Netherlands.
- Li, P., and Q. Wang. 2013. "Developing and Validating Novel Hyperspectral Indices for Leaf Area Index Estimation: Effect of Canopy Vertical Heterogeneity." *Ecological Indicators* 32: 123–130. doi:10.1016/j.ecolind.2013.03.025.
- Liang, L., L. Di, L. Zhang, M. Deng, Z. Qin, S. Zhao, and H. Lin. 2015. "Estimation of Crop LAI Using Hyperspectral Vegetation Indices and a Hybrid Inversion Method." *Remote Sensing of Environment* 165: 123–134. doi:10.1016/j.rse.2015.04.032.
- Luo, J., R. Ma, H. Feng, and X. Li. 2016. "Estimating the Total Nitrogen Concentration of Reed Canopy with Hyperspectral Measurements considering a Non-Uniform Vertical Nitrogen Distribution." *Remote Sensing* 8 (10): 789. doi:10.3390/rs8100789.
- Malenovský, Z., L. Homolová, R. Zurita-Milla, P. Lukeš, V. Kaplan, J. Hanuš, J.-P. Gastellu-Etchegorry, and M. E. Schaepman. 2013. "Retrieval of Spruce Leaf Chlorophyll Content from Airborne Image Data Using Continuum Removal and Radiative Transfer." *Remote Sensing of Environment* 131: 85–102. doi:10.1016/j.rse.2012.12.015.
- Marcoulides, G. A., and C. Saunders. 2006. "Editor's Comments: PLS: A Silver Bullet?" *MIS Quarterly* 30 (2): iii–ix. doi:10.2307/25148727.
- Martin, A. R., and S. C. Thomas. 2011. "A Reassessment of Carbon Content in Tropical Trees." *PLoS ONE* 6 (8): e23533. doi:10.1371/journal.pone.0023533.
- Meerdink, S. K., D. A. Roberts, J. Y. King, K. L. Roth, P. E. Dennison, C. H. Amaral, and S. J. Hook. 2016. "Linking Seasonal Foliar Traits to VSWIR-TIR Spectroscopy across California Ecosystems." *Remote Sensing of Environment* 186: 322–338. doi:10.1016/j.rse.2016.08.003.
- Mercado, L., J. Lloyd, F. Carswell, Y. Malhi, P. Meir, and A. D. Nobre. 2006. "Modelling Amazonian Forest Eddy Covariance Data: A Comparison of Big Leaf versus Sun/Shade Models for the C-14 Tower at Manaus I. Canopy Photosynthesis." *Acta Amazonica* 36: 69–82. doi:10.1590/S0044-59672006000100009.
- Mutanga, O., and A. K. Skidmore. 2004. "Narrow Band Vegetation Indices Overcome the Saturation Problem in Biomass Estimation." *International Journal of Remote Sensing* 25 (19): 3999–4014. doi:10.1080/01431160310001654923.
- Neinavaz, E., A. K. Skidmore, R. Darvishzadeh, and T. A. Groen. 2016. "Retrieval of Leaf Area Index in Different Plant Species Using Thermal Hyperspectral Data." *ISPRS Journal of Photogrammetry and Remote Sensing* 119: 390–401. doi:10.1016/j.isprsjprs.2016.07.001.
- Opti-Sciences. 2011. *CCM-300 Chlorophyll Content Meter*. Hudson: Opti-Sciences.

- Peng, Y., A. Nguy-Robertson, T. Arkebauer, and A. Gitelson. 2017. "Assessment of Canopy Chlorophyll Content Retrieval in Maize and Soybean: Implications of Hysteresis on the Development of Generic Algorithms." *Remote Sensing* 9 (3): 226. doi:10.3390/rs9030226.
- Pereira, H. M., S. Ferrier, M. Walters, G. N. Geller, R. H. G. Jongman, R. J. Scholes, M. W. Bruford et al. 2013. "Essential Biodiversity Variables." *Science* 339 (6117): 277. doi:10.1126/science.1229931.
- Perkin-Elmer. 2005. *2400 Series II CHNS/O Elemental Analysis*. Waltham: Perkin Elmer.
- Peterson, D. L., J. D. Aber, P. A. Matson, D. H. Card, N. Swanberg, C. Wessman, and M. Spanner. 1988. "Remote Sensing of Forest Canopy and Leaf Biochemical Contents." *Remote Sensing of Environment* 24 (1): 85–108. doi:10.1016/0034-4257(88)90007-7.
- Poorter, H., Ü. Niinemets, L. Poorter, I. J. Wright, and R. Villar. 2009. "Causes and Consequences of Variation in Leaf Mass per Area (LMA): A Meta-Analysis." *New Phytologist* 182 (3): 565–588. doi:10.1111/j.1469-8137.2009.02830.x.
- Prabhakara, K. W., D. Hively, and G. W. McCarty. 2015. "Evaluating the Relationship between Biomass, Percent Groundcover and Remote Sensing Indices across Six Winter Cover Crop Fields in Maryland, United States." *International Journal of Applied Earth Observation and Geoinformation* 39: 88–102. doi:10.1016/j.jag.2015.03.002.
- Wang, Q., and P. Li. 2013. "Canopy Vertical Heterogeneity Plays a Critical Role in Reflectance Simulation." *Agricultural and Forest Meteorology* 169: 111–121. doi:10.1016/j.agrformet.2012.10.004.
- Ramoelo, A., M. Cho, R. Mathieu, and A. K. Skidmore. 2015. "Potential of Sentinel-2 Spectral Configuration to Assess Rangeland Quality." *Journal of Applied Remote Sensing* 9 (1): 094096. doi:10.1117/1.jrs.9.094096.
- Ramoelo, A., A. K. Skidmore, M. Schlerf, R. Mathieu, and I. M. A. Heitkönig. 2011. "Water-Removed Spectra Increase the Retrieval Accuracy When Estimating Savanna Grass Nitrogen and Phosphorus Concentrations." *ISPRS Journal of Photogrammetry and Remote Sensing* 66 (4): 408–417. doi:10.1016/j.isprsjprs.2011.01.008.
- Richter, K., C. Atzberger, F. Vuolo, P. Weihs, and G. D'Urso. 2009. "Experimental Assessment of the Sentinel-2 Band Setting for RTM-based LAI Retrieval of Sugar Beet and Maize." *Canadian Journal of Remote Sensing* 35 (3): 230–247. doi:10.5589/m09-010.
- Roelofs, H., P. van Bodegom, L. Kooistra, and J.-P. Witte. 2013. "Trait Estimation in Herbaceous Plant Assemblages from in Situ Canopy Spectra." *Remote Sensing* 5 (12): 6323. doi:10.3390/rs5126323.
- Savitzky, A., and M. J. E. Golay. 1964. "Smoothing and Differentiation of Data by Simplified Least Squares Procedures." *Analytical Chemistry* 36 (8): 1627–1639. doi:10.1021/ac60214a047.
- Scheiter, S., L. Langan, and S. I. Higgins. 2013. "Next-Generation Dynamic Global Vegetation Models: Learning from Community Ecology." *New Phytologist* 198 (3): 957–969. doi:10.1111/nph.12210.
- Schlemmer, M., A. Gitelson, J. Schepers, R. Ferguson, Y. Peng, J. Shanahan, and D. Rundquist. 2013. "Remote Estimation of Nitrogen and Chlorophyll Contents in Maize at Leaf and Canopy Levels." *International Journal of Applied Earth Observation and Geoinformation* 25: 47–54. doi:10.1016/j.jag.2013.04.003.
- Serbin, S. P., A. Singh, B. E. McNeil, C. C. Kingdon, and P. A. Townsend. 2014. "Spectroscopic Determination of Leaf Morphological and Biochemical Traits for Northern Temperate and Boreal Tree Species." *Ecological Applications* 24 (7): 1651–1669. doi:10.1890/13-2110.1.
- Shiklomanov, A. N., M. C. Dietze, T. Viskari, P. A. Townsend, and S. P. Serbin. 2016. "Quantifying the Influences of Spectral Resolution on Uncertainty in Leaf Trait Estimates through a Bayesian Approach to RTM Inversion." *Remote Sensing of Environment* 183: 226–238. doi:10.1016/j.rse.2016.05.023.
- Skidmore, A. K., N. Pettorelli, N. C. Coops, G. N. Geller, M. Hansen, R. Lucas, C. A. Múcher et al. 2015. "Environmental Science: Agree on Biodiversity Metrics to Track from Space." *Nature* 523 (7561): 403–405. doi:10.1038/523403a.
- Sprintsin, M., J. M. Chen, A. Desai, and C. M. Gough. 2012. "Evaluation of Leaf-To-Canopy Upscaling Methodologies against Carbon Flux Data in North America." *Journal of Geophysical Research: Biogeosciences* 117 (G01023): 1–17. doi:10.1029/2010jg001407.

- Thenkabail, P. S., R. B. Smith, and E. De Pauw. 2000. "Hyperspectral Vegetation Indices and Their Relationships with Agricultural Crop Characteristics." *Remote Sensing of Environment* 71 (2): 158–182. doi:10.1016/S0034-4257(99)00067-X.
- Thomas, V., P. Treitz, J. H. McCaughey, T. Noland, and L. Rich. 2008. "Canopy Chlorophyll Concentration Estimation Using Hyperspectral and Lidar Data for a Boreal Mixedwood Forest in Northern Ontario, Canada." *International Journal of Remote Sensing* 29 (4): 1029–1052. doi:10.1080/01431160701281023.
- Ullah, S., A. K. Skidmore, A. Ramoelo, T. A. Groen, M. Naeem, and A. Ali. 2014. "Retrieval of Leaf Water Content Spanning the Visible to Thermal Infrared Spectra." *ISPRS Journal of Photogrammetry and Remote Sensing* 93: 56–64. doi:10.1016/j.isprsjprs.2014.04.005.
- Verrelst, J., J. P. Rivera, G. Leonenko, L. Alonso, and J. Moreno. 2014. "Optimizing LUT-Based RTM Inversion for Semiautomatic Mapping of Crop Biophysical Parameters from Sentinel-2 and -3 Data: Role of Cost Functions." *IEEE Transactions on Geoscience and Remote Sensing* 52 (1): 257–269. doi:10.1109/TGRS.2013.2238242.
- Verrelst, J., G. Camps-Valls, J. Muñoz-Mari, J. P. Rivera, F. Veroustraete, J. G. P. W. Clevers, and J. Moreno. 2015. "Optical Remote Sensing and the Retrieval of Terrestrial Vegetation Biophysical Properties – A Review." *ISPRS Journal of Photogrammetry and Remote Sensing* 108: 273–290. doi:10.1016/j.isprsjprs.2015.05.005.
- Verrelst, J., M. E. Schaepman, Z. Malenovsky, and J. G. P. W. Clevers. 2010. "Effects of Woody Elements on Simulated Canopy Reflectance: Implications for Forest Chlorophyll Content Retrieval." *Remote Sensing of Environment* 114 (3): 647–656. doi:10.1016/j.rse.2009.11.004.
- Wakeling, I. N., and J. J. Morris. 1993. "A Test of Significance for Partial Least Squares Regression." *Journal of Chemometrics* 7 (4): 291–304. doi:10.1002/cem.1180070407.
- Wang, Z., A. K. Skidmore, R. Darvishzadeh, U. Heiden, M. Heurich, and T. Wang. 2015. "Leaf Nitrogen Content Indirectly Estimated by Leaf Traits Derived from the PROSPECT Model." *IEEE Journal of Selected Topics in Applied Earth Observations and Remote Sensing* 8 (6): 3172–3180. doi:10.1109/jstars.2015.2422734.
- Weerasinghe, L. K., D. Creek, K. Y. Crous, S. Xiang, M. J. Liddell, M. H. Turnbull, and O. K. Atkin. 2014. "Canopy Position Affects the Relationships between Leaf Respiration and Associated Traits in a Tropical Rainforest in Far North Queensland." *Tree Physiology* 34 (6): 564–584. doi:10.1093/treephys/tpu016.
- Whitehurst, A., A. Swatantran, J. Blair, M. Hofton, and R. Dubayah. 2013. "Characterization of Canopy Layering in Forested Ecosystems Using Full Waveform Lidar." *Remote Sensing* 5 (4): 2014. doi:10.3390/rs5042014.
- Wilkes, P., S. D. Jones, L. Suarez, A. Haywood, A. Mellor, W. Woodgate, M. Soto-Berelov, and A. K. Skidmore. 2016. "Using Discrete-Return Airborne Laser Scanning to Quantify Number of Canopy Strata across Diverse Forest Types." *Methods in Ecology and Evolution* 7 (6): 700–712. doi:10.1111/2041-210x.12510.
- Wold, S., M. Sjöström, and L. Eriksson. 2001. "PLS-regression: A Basic Tool of Chemometrics." *Chemometrics and Intelligent Laboratory Systems* 58 (2): 109–130. doi:10.1016/S0169-7439(01)00155-1.
- Yi, Q., F. Wang, A. Bao, and G. Jiapaer. 2014. "Leaf and Canopy Water Content Estimation in Cotton Using Hyperspectral Indices and Radiative Transfer Models." *International Journal of Applied Earth Observation and Geoinformation* 33: 67–75. doi:10.1016/j.jag.2014.04.019.
- Yoder, B. J., and R. E. Pettigrew-Crosby. 1995. "Predicting Nitrogen and Chlorophyll Content and Concentrations from Reflectance Spectra (400–2500 Nm) at Leaf and Canopy Scales." *Remote Sensing of Environment* 53 (3): 199–211. doi:10.1016/0034-4257(95)00135-N.