ORIGINAL ARTICLE



# Field spectroscopy and radiative transfer modelling to assess impacts of petroleum pollution on biophysical and biochemical parameters of the Amazon rainforest

Paul Arellano<sup>1,2,3</sup> · Kevin Tansey<sup>3</sup> · Heiko Balzter<sup>3,4</sup> · Doreen S. Boyd<sup>5</sup>

Received: 9 March 2016/Accepted: 28 February 2017 © The Author(s) 2017. This article is published with open access at Springerlink.com

Abstract Biophysical and biochemical plant foliage parameters play a key role in assessing vegetation health. Those plant parameters determine the spectral reflectance and transmittance properties of vegetation; therefore, hyperspectral remote sensing, particularly imaging spectroscopy, can provide estimates of leaf and canopy chemical properties. Based on the relationship between spectral response and biochemical/biophysical properties of the leaves and canopies, the PROSPECT radiative transfer model simulates the interaction of light with leaves. In this study, more than 1100 leaf samples from the Amazon

**Electronic supplementary material** The online version of this article (doi:10.1007/s12665-017-6536-6) contains supplementary material, which is available to authorized users.

☑ Paul Arellano parellano@yachaytech.edu.ec; pa134@le.ac.uk

- <sup>1</sup> Present Address: School of Geological Sciences and Engineering, Yachay Tech University, Hacienda San Jose s/n, San Miguel de Urcuqui, Imbabura, Ecuador
- <sup>2</sup> Centre for Earth Observation (CEO), Yachay Tech University, Hacienda San José s/n, San Miguel de Urcuqui, Imbabura, Ecuador
- <sup>3</sup> Department of Geography, Centre of Landscape and Climate Research, Leicester Institute of Space and Earth Observation, University of Leicester, University Road, Leicester LE1 7RH, UK
- <sup>4</sup> National Centre for Earth Observation, University of Leicester, University Road, Leicester LE1 7RH, UK
- <sup>5</sup> School of Geography, University of Nottingham, Nottingham NG7 2RD, UK

forest of Ecuador were collected at several study sites, some of which are affected by petroleum pollution, and across the vertical profile of the forest. For every sample, field spectroscopy at leaf level was conducted with a spectroradiometer. The goal of this study was to assess leaf optical properties of polluted and unpolluted rainforest canopies across the vertical profile and identify vegetation stress expressed in changes of biophysical and biochemical properties of vegetation. An ANOVA followed by Holme's multiple comparisons of means and a principal component analysis showed that photosynthetic pigments, chlorophyll and carotenoids have significantly lower levels across the vertical profile of the forest, particularly in sites affected by petroleum pollution. On the other hand, foliar water content showed significantly higher levels in the polluted site. Those findings are symptoms of vegetation stress caused by reduced photosynthetic activity and consequently decreased transpiration and water-use efficiency of the plants. Cross-comparison between SPAD-502 chlorophyll content meter index and chlorophyll content showed strong positive correlation coefficients (r = 0.71 and  $r^2 = 0.51$ ) which suggests that using the SPAD-502 chlorophyll index itself is sensitive enough to detect vegetation stress in a multispecies tropical forest. Therefore, the SPAD-502 can be used to assess chlorophyll content of vegetation across polluted and non-polluted sites at different canopy layers. The results presented in this paper contribute to the very limited literature on field spectroscopy and radiative transfer models applied to the vertical profile of the Amazon forest.

**Keywords** Amazon forest · Chlorophyll content · PROSPECT model · SPAD-502 · Yasuni National Park · Petroleum pollution

#### Introduction

Primary productivity in vegetation is principally determined by leaf photosynthetic rate and leaf life duration, as well as the availability of such factors as nitrogen, water and temperature (Bindi et al. 2002). It is the foliar biochemistry of a leaf that is closely related to maximum photosynthetic rates (Martin and Aber 1997; Bacour et al. 2006) and in particular the photochemical chlorophyll. At canopy level, chlorophyll content is positively correlated with net primary productivity (NPP) (Dash and Curran 2007; Gitelson et al. 2006). NPP is impacted on by vegetation stress and health (Peñuelas and Filella 1998; Richardson et al. 2002; Bannari et al. 2007), with chlorophyll content directly related to plant stress physiology as its levels decrease under stress. This provides vital information regarding the response of vegetation to changes in its environment (Clevers et al. 2002; Ollinger and Smith 2005; Kumar et al. 2006).

When a plant is under stress, changes in chlorophyll content are evident in the initial stages, and as the stress increases, chlorophyll content decreases more quickly than the other photosynthetic pigments. Moreover, this decrease is likely to occur prior to observed physiological changes, such as leaf area loss and chlorosis. Since different plant species respond differently to a particular stressor and the nature, intensity and length of exposure are factors that define the stress level on the vegetation, the ability to determine this photosynthetic pigment content yields important information about both vegetation vigour and environmental quality (Noomen et al. 2012; Carter and Spiering 2002).

It is not clear how the biophysical and biochemical parameters of tropical forest change under stress conditions. A recent paper found that tropical forest exposed to petroleum pollution showed reduced levels of chlorophyll content, higher levels of foliar water content and leaf structural changes (Arellano et al. 2015). Another study carried out in central Sumatra (western Indonesia) identified vegetation stress in densely vegetated primary tropical forest caused by metal contamination of the soil (Cu, Pb and Zn) and mineralised areas. The detected stress symptoms are expressed as a reduction in chlorophyll content and leaf structural deformation (Hede et al. 2015). In both studies, remote sensing methods were suitable to identify vegetation stress caused by pollution in tropical forest environments. This capability is useful in addition to several other methods that have been successfully applied to detect soil, sediment and water pollution from disposal of coal waste piles emplaced in old mine sites and adjacent areas (Cutruneo et al. 2014; Saikia et al. 2014; Hower et al. 2013; Oliveira et al. 2012a, b, 2013; Ribeiro et al. 2010, 2013; Silva et al. 2012a, b; Quispe et al. 2012).

There is considerable evidence that hydrocarbon gases in the soil are a source of vegetation stress (Gustafson 1944, 1950; Horvitz 1972; Lang 1985a, b; Melo et al. 1996; Yang et al. 1999, 2000; Smith et al. 2004; 2005b; Noomen 2007; Noomen and Skidmore 2009). Spectroscopy studies of stressed vegetation have shown an increased value in visible wavelengths due to the chlorophyll content reduction and a decreased reflectance in the NIR due to the structural damage of the plants. Most of these studies have been developed under controlled experimental conditions in the laboratory. Field spectroscopy studies of stressed vegetation in the Amazon forest are rare. Given the remote location faced by these environments (Malhi et al. 2008; Phillips et al. 2009; Davidson et al. 2012) and the recent contention around ecosystem functioning in tropical forests (Morton et al. 2014), it is important that where possible field data are collected which is for purpose and made readily available.

High spectral resolution remote sensing, particularly imaging spectroscopy, provides estimates of leaf and canopy chemical properties (Asner and Martin 2008). Spectral reflectance and transmittance of vegetation are the result of biophysical and biochemical parameters of plants. Based on the relationship between reflectance and the biochemical and biophysical properties of the leaves and canopies, models have been created in order to simulate the interaction of the light with the plant leaves through radiative transfer theory.

The leaf optical properties spectra (PROSPECT) model describes radiative transfer within a broadleaf with a plate model. Plate models treat internal leaf structure as sheets or plates and calculate multiple reflections of diffuse radiation between these interfaces (Kumar et al. 2006). PROSPECT is based on the representation of the leaf as one or several absorbing plates with a rough surface giving rise to isotropic scattering. The model estimates the directional-hemispherical reflectance and transmittance of leaves across the solar spectrum from 400 to 2500 nm (Jacque-moud 2009).

A leaf structure parameter of the model is represented by *N* which is the number of compact layers specifying the average number of air/cell wall interfaces within the mesophyll. The leaf biophysical parameters of the model are represented by chlorophyll a + b content ( $C_{ab}$ ), equivalent water thickness ( $C_w$ ), dry matter content ( $C_m$ ) and brown pigments content ( $C_{bp}$ ). Inversion of PRO-SPECT revealed good agreement between measured and predicted leaf chlorophyll concentrations (Kumar et al. 2006; Jacquemoud 2009; Féret et al. 2008).

In this study, we assess foliar biophysical and biochemical parameters of tropical forests in remote areas of the western Amazon region. More than one thousand leaf samples were collected from both primary and secondary forests plots (thus having different primary productivities); some of the secondary forests have a history of petroleum pollution. Field spectroscopy was conducted to sample the spectral response of the collected leaves. The objectives of this study were to understand the effects of petroleum pollution on the vegetation of the Amazon forest by investigating the biophysical and biochemical parameters in the vertical profile of the forest to stress conditions caused by hydrocarbon pollution.

#### Materials and methods

#### Study area and sites

The study area was selected to include three forest plots in the Amazon rainforest of Ecuador, including two secondary forest sites, one of which was polluted by an oil spill (Site 1) and the other unpolluted (Site 2), and one pristine rainforest site at Yasuni National Park (Site 3). The secondary forest plots are located just hundreds of meters distance from each other, therefore sharing the same environmental conditions (soil type, weather, human influence). This is a lowland secondary forest, having been disturbed by selective logging and agricultural activities which have, however, diminished over the past 20 years leading to forest regeneration. In addition, the polluted site has also been exposed to petroleum pollution as a result of oil industry activity. This provided vegetation with lower expected chlorophyll content than the unpolluted site. Controlled experiments have demonstrated that plants exposed to hydrocarbons experience reduced levels of chlorophyll content (Smith et al. 2004, 2005a, b; Noomen 2007; Noomen and Skidmore 2009; Yang 1999). Moreover, the interaction between petroleum and the soil reduces the amount of soil oxygen and increases the CO<sub>2</sub> concentration, causing soils to turn acidic, and minerals are mobilised affecting plant health (Yang et al. 1999; Shumacher 1996; Noomen et al. 2006; van der Meer et al. 2006). The third study site is a lowland evergreen pristine forest where the Pontifical Catholic University of Ecuador established and manages a permanent forest dynamics plot of 50 hectares where over 150,000 mapped trees >1 cm in diameter at breast height (dbh) from over 1100 species have been identified (Valencia 2004). Studies consider that the plant species richness in this area is among the highest in the world (Tedersoo et al. 2010). Location maps are shown in Figs. 1 and 2. Table 1 describes the locations of each study site.

### **Field sampling**

Fieldwork was undertaken from April to June 2012. At each of the three sites, well-developed branches were

carefully selected and collected at different levels of the vertical profile of the forest by using a telescopic pruner, by tree-climbing techniques and by canopy towers. The branches were sealed in large polyethylene bags to maintain the moisture content and then stored in ice coolers. The foliar material was transported to a local site, and fully expanded mature leaves with no damage by herbivores or pathogens were selected for analysis. A total of 1137 samples were collected across the study sites and vertical profile of the forest.

The sampling process determined that a wide range of vegetation heterogeneity related to species distribution, phenological stage and leaf structure were selected. Lists of vegetation species sampled in the study sites are detailed in Supporting Information Tables S1 and S2. In each case, depending on the size and shape of the leaf, different cork borers of variable size between 2.5 and 8.5 cm diameter were used to clip a leaf disc from the central and widest portion of the leaf blade, avoiding the major veins. All leaf discs were clipped from the midpoint of the leaves since it has been documented that it is the best position from which to take chlorophyll readings (Hoel 1998).

In addition to the leaf samples, soil samples were taken from all sites and analysed in certified laboratories. Parameters related to physical properties, petroleum hydrocarbons, metals and soil nutrients were considered. Table 2 shows the results of laboratory analysis of soil in the three study sites. Higher levels of TPH (total petroleum hydrocarbons) of Site 1 confirmed that it is an area affected by petroleum pollution.

#### Ground truth instruments

Field spectroscopy at leaf level was conducted with an ASD FieldSpec HandHeld-2 spectroradiometer (Analytical Spectral Devices Inc., Boulder, Colorado). This instrument measures a wavelength range from 325 to 1075 nm and sampling interval of 1 nm. The spectrometer was attached to a plant probe with an internal 4.05-W halogen light source and a leaf clip that includes a rotating head with both white and black reference panels. This mechanism holds the leaves, excludes ambient direct and scattered light and ensures a constant field of view (FOV) of 10 mm for the target sample. Reflectance was estimated by recording the radiance reflected from the leaf with the black reference panel, and double transmittance was estimated by recording the radiance reflected from the leaf with the white panel.

Spectrum averages of ten scans were taken for each leaf disc and to the white reference radiance panel. The appropriate number of scans was established based on a compromise between the time required for each spectrum collection and noise reduction. The measured differences were tested in the specific wavelength range used by

400000

Cuyabeno Ecological Reserve

Yasuni National Park

400000



Fig. 1 Study area maps. a Study area located in the north-east Ecuador. b Polluted and unpolluted sites (Sites 1 and 2) in the secondary forest. Pristine Amazon rainforest site (Site 3). *Source*:

ESRI/Data&Maps 2000 (CD 1), WorldSat Color Shaded Relief Image, WorldSat International, Inc.



Fig. 2 Location of sampled areas in secondary forest. *Black polygons* polluted site (Site 1); *red polygons* unpolluted secondary forest (Site 2). Ikonos image as background

reflectance indices for the estimation of chlorophyll content. The difference between 10 and 20 scans in the range of 700–750 nm was 2.0%, and in the range 751–800 nm it was 0.9%. Differences between 10 and 30 scans in the same ranges were 3.3 and 1.8%, respectively. These small differences were considered acceptable for the reflectance index calculation. This is an advantage of the plant probe and leaf clip mechanism which provides a constant light source in a closed environment allowing us to reduce the numbers of scans and keep a comparable signal-to-noise ratio. These spectroradiometer measurements (reflectance and double transmittance) were used to estimate chlorophyll content by applying the inversion process of the PROSPECT model.

Table 1	Description	of the	e three	sites	in	the	study a	area
---------	-------------	--------	---------	-------	----	-----	---------	------

	Site 1	Site 2		Site 3			
Forest classification	A seasonal lowland evergree	en rainforest					
Province	Sucumbios/Tarapoa	Sucumbios/Tarapoa					
Forest type	Secondary/regrowth following	Secondary/regrowth following abandoning farming					
Pollution	Petroleum pollution	Petroleum pollution Not affected by pollution					
Coordinates system	UTM-WGS84-Z18N						
Latitude (m)	9,979,706	9,980,060	9,978,875	9,924,894			
Longitude (m)	348,523	348,694	347,525	344,220			
Altitude (m. above sea level)	232	238	229	216-248 (Valencia 2004)			
Mean annual temperature (°C)	23 (Fitton 2000)			24-34 (Valencia 2004)			
Mean annual rainfall (mm)	3300 (Fitton 2000)			3081 (Valencia 2004)			
Soil type	Histosols/fibrits/tropofibrits			Inceptisols/tropots/distrops			

Table 2 Results of laboratory analysis of soil samples for the three study sites (Arellano et al. 2015)

Parameters	Standard method	Units	Site 1 polluted	Site 2 unpolluted		Site 3 Pristine forest—Yasuni National Park				
Hydrocarbons										
Hydrocarbons aromatic polycyclic-HAPs	CP-PEE-S001	mg/kg	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5	<0.5
Total petroleum hydrocarbons—TPH	CP-PEE-S003	mg/kg	8876.6	<200	<200	<200	<200	<200	<200	<200
Metals										
Iron	EPA 3051/7000A	mg/kg	18,905	17,635	12,598	15,460	15,108	14,569	13,692	18,916
Aluminium	EPA 3051/APHA 350	mg/kg	36	36	36	36	<10	<10	<10	<10
Zinc	EPA 3051/7000A	mg/kg	28.7	47	50	31.5	38.1	80.9	34.2	>100
Magnesium	EPA 3051/7000A	mg/kg	372.5	>500	>500	445	386.7	383.3	380.3	389.2
Manganese	EPA 3051/7000A	mg/kg	116.3	>500	>500	90.5	836.7	836.1	156.6	232.5
Barium	EPA 3051/Hach 801	mg/kg	<50	83.2	87.6	113	<50	<50	53.5	88.8
Physical properties										
Electrical conductivity	SM 2010 B	uS/cm	32	56.6	62.6	85.6	38.4	34.4	32	28.9
рН	CP-PEE-S004		4.6	5.5	5.5	<4.0	5.3	4.8	4.6	4.2
Nutrients										
Organic matter	Gravimetric	%	43.3	62.1	52.6	41.9	60.3	64.6	60.4	64
Total N	SM 4500-N C	mg/kg	22	50	110	46	27	36	78	44
Total P	SM 4500 P B-C	mg/kg	>450	400	>450	>450	>500	>500	>500	150
Potassium (K)	EPA 3051/7000A	mg/kg	280	>500	>500	420	189	188.1	201.2	201.5

The laboratory has the follow accreditations: National Accreditation Office from Spain (ENAC No. 415/LE 929). Ecuadorian Accreditation Office (OAE No. LE2 C 04-001 and OAE LE C 10-011)

Bold value indicates high levels beyond the environmental regulation

A SPAD-502 chlorophyll meter (Konica Minolta, Osaka, Japan), which offers an easy, rapid and portable method for an indirect estimation of chlorophyll content, was used. The SPAD-502 bases its measurements on the light that is transmitted by the leaf in two wavelength regions. The two wavelengths used by the meter are located in the red region at 650 nm which corresponds to the chlorophyll absorption

peak unaffected by carotene and the infrared region at 940 nm where chlorophyll absorption is extremely low. The light emitted by the instrument and transmitted by the leaf is measured by the receptor and converted into electrical signals. Finally, a chlorophyll index is calculated by using the ratio of the intensity of the transmitted light (Konica Minolta 2009). Chlorophyll meters have been used extensively in

agriculture to estimate chlorophyll and nitrogen in different species (Monje and Bugbee 1992; Marwell et al. 1995; Torres-Netto et al. 2002, 2005; Hawkins et al. 2009) and also in forest studies (Richardson et al. 2002, Richardson 2002, Castro-Esau et al. 2006). Furthermore, chlorophyll meters have been used in the indirect assessment of foliar nitrogen (Hoel 1998; Torres-Netto et al. 2002, 2005) and carotenoid content (Torres-Netto et al. 2002; Poorter et al. 2000). However, the use of chlorophyll meters in the Amazon forest is extremely rare (Coste et al. 2010; Marenco et al. 2009; Cerovic et al. 2012). Three readings at different position of the leaf disc were taken using the portable SPAD-502 chlorophyll meter, and a mean index value was computed. Here we use the SPAD chlorophyll index to correlate with chlorophyll content estimations from the radiative transfer PROSPECT model.

### **PROSPECT** model

Biophysical and biochemical parameters of the vegetation layer were estimated for each leaf sample. First of all, the spectral data were pre-processed, followed by the inversion of the PROSPECT model. Figure 3 illustrates this procedure. During the pre-processing, the Savitzky–Golay filter (SGF) was applied to the reflectance and double-transmittance data in order to smooth the signal and increase the signal-to-noise ratio. SGF performs a least squares fit of a small set of consecutive data points to a polynomial and takes the calculated central point of the fitted polynomial curve a the new smoothed data point. Mathematically, it operates as follows:

$$X_{j} = \frac{1}{N} \sum_{h=-k}^{k} C_{h} X_{j} + h$$
(1)

where  $X_j$  is the new value, N is a normalising coefficient, k is the number of neighbour values at each site of j and  $C_h$  are pre-computed coefficients that depend on the chosen order of the polynomial. A MATLAB© script was applied to the whole reflectance and double-reflectance foliage samples with a third-order fitted polynomial and a frame size of 31.

An automatic procedure to estimate transmittance based on double transmittance was implemented in R (R Core Team 2012) based on the Kubelka–Munk theory of light scattering and light absorption by the following equation (International Standard Organization 2012, Jacquemoud and Marcq 2012):

$$T = \sqrt{\left[R(\lambda) - R_0(\lambda)\right] \left[\frac{1}{R_w(\lambda)} - R_0(\lambda)\right]}$$
(2)

where *T* is the estimated transmittance,  $R(\lambda)$  is the luminance factor of the leaf using the white background disc,  $R_0(\lambda)$  is the luminance factor of the leaf using the black

background disc of the leaf clip and  $R_w(\lambda)$  is the luminance factor of the white background disc of the leaf clip.

Reflectance and transmittance were used to perform the inversion of the PROSPECT model and obtain the following parameters: leaf structure parameter represented by N, chlorophyll a + b content ( $C_{ab}$ ), equivalent water thickness ( $C_w$ ), dry matter content ( $C_m$ ) and brown pigments content ( $C_{bp}$ ).

## **Results and discussion**

Soil analysis revealed increased levels of total petroleum hydrocarbons (TPH) of nearly 9000 mg/kg in the secondary forests affected by petroleum pollution (Site 1), which confirms that the open pits containing petroleum are still active sources of pollution. Soil samples from the other sites, the unpolluted secondary forest (Site 2) and pristine forest (Site 3) had less than 200 mg/kg TPH which confirms that these two sites are not affected by petroleum pollution (Arellano et al. 2015).

# Biophysical and biochemical vegetation parameters across sites and vertical forest canopy profile

Figure 4 illustrates the average reflectance response of the 1137 samples from the three study sites. Differences between sites are manifest in the visible (VIS, 400–680 nm) and near-infrared (NIR, 750–1100 nm) domains. Figure 4a shows that light absorption in the VIS is higher in the pristine forest and lower in the polluted forest. This suggests greater photosynthetic activity in the pristine forest, therefore higher content of photosynthetic pigments (specially chlorophyll), followed by secondary forest (non-polluted), and lower values in the polluted forest. Figure 4b illustrates that mean reflectance in the NIR decreases in the polluted forest which indicates increased levels of foliar water content.

The vertical profile of the tropical forest is a key determinant of photosynthetic activity along the upper, medium and understory. The high diversity of the tropical forest and its complex structure plays a particular role in the interaction with light which is the principal source of energy in the forest. The light is reflected, absorbed and transmitted at different levels in the vertical profile. The upper layer receives 25-100% of the relative irradiance, lower down the irradiance decreases until it reaches 1-3% in the lower layers of the forest (Longman 1987; Chazdon and Fetcher 1984). Canopy high and vegetation density cause steep vertical gradients of the microclimate specially related to temperature and humidity and a consequently differentiation in carbon dioxide (CO<sub>2</sub>) concentrations (Roberts et al. 1990; Grace 1999). Figure 5 shows the mean



Fig. 4 Average reflectance for the 1139 samples in the three study sites. **a** Reflectance representation in the visible range only (400-700 nm), **b** reflectance response in the visible and NIR ranges (400-1100 nm)

and standard error reflectance in the VIS and NIR for the three study sites and three levels of the vertical profile of the tropical forest. In all sites and vertical layers of the forest, there are significant differences in the VIS range which may lead to significant differences in plant pigments content; in the same way, significant differences in the NIR may suggest structural damages on leaves from the polluted site.

In order to obtain specific indicators of biophysical and biochemical parameters of the forest, an inversion process of the PROSPECT model was constructed for the whole dataset containing 1109 samples after removing outliers. Table 3 shows the descriptive statistics of PROSPECT parameters and the SPAD index.

Figure 6 illustrates the strongest correlation coefficients between parameters (correlations between other parameters were shown to be weak and are nor presented in this study).

Chlorophyll content and SPAD index show strong correlations (r = 0.71,  $r^2 = 0.51$ ) which demonstrate that the portable SPAD-502 chlorophyll meter is an effective method to assess photosynthetic activity and plant pigments in the tropical forest. The correlation between the light-harvesting pigments, chlorophyll and carotenoids is strong as well (r = 0.72,  $r^2 = 0.52$ ). In senescent leaves, the relationship between those two pigments is negative since chlorophyll degrades faster than carotenoids. When carotenoids become the dominant pigment, then leaves appear yellow. Our study sites are evergreen forest; therefore, the relationship between those pigments is relatively constant across sites and canopy layers (see Fig. 7e).

Tables S3 and S4 in Supporting Information present the descriptive statistics across study sites and canopy layers, and Fig. 7 displays mean and 95% confidence interval for each parameter across sites and canopy layers. All



Fig. 5 Average reflectance per site and canopy layer. The line represents the mean, and line width represents the standard error

of the derived PROSPECT model parameters and SPAD	PROSPECT parameters	Units	Mean	Min	Max	SD	SE
	N		1.8428	1.2320	2.9500	0.3327	0.0365
index	Chlorophyll $(a + b)$	µg/cm <sup>2</sup>	55.7483	19.2120	105.3600	17.0960	1.8765
	Carotenoids	µg/cm <sup>2</sup>	12.2677	0.9534	44.7160	7.5868	0.8328
	Brown pigments	µg/cm <sup>2</sup>	0.1650	0.0005	0.6980	0.1415	0.0155
	Water content	g/cm <sup>2</sup>	0.0106	0.0008	0.0431	0.0078	0.0009
	Organic matter	g/cm <sup>2</sup>	0.0067	0.0002	0.0191	0.0036	0.0004

Index

56.2980

36.1000

parameters show minor differences between upper and medium canopy probably because light and environmental factors at the highest canopy levels remain relatively constant. Figure 7a (chlorophyll content), Fig. 7b (SPAD chlorophyll index), Fig. 7c (N parameter) as expected illustrate lower levels in the understorey due the vertical gradient of the forest; however, a much lower level for these parameters is observed in the polluted site which demonstrates that petroleum pollution may be the principal

SPAD

factor that affects biophysical and biochemical process of the vegetation. Carotenoid content (Fig. 7d) also shows reduced levels in the understorey, especially in the nonpolluted site.

77.6000

10.0284

1.1008

The relationship between carotenoids and chlorophyll was discussed above (Fig. 7e). Organic matter content (Fig. 7f) shows lower levels in the pristine forest across the vertical profile of the forest. Brown pigments (Fig. 7g) show lower levels in pristine forest and higher levels in the

Fig. 6 Correlation coefficient between a chlorophyll content versus SPAD chlorophyll index; b chlorophyll content versus carotenoids



secondary forest. Finally, it is interesting to observe that foliar water content reports consistently higher values across the vertical profile of the forest in vegetation growing in the polluted site (see Fig. 7h) which suggests structural changes in the leaf caused by vegetation stress as illustrated in the lower reflectance values in the NIR region (Fig. 4b). This finding endorses the results of a recent study which found that plants exposed to contaminants exhibit stomata closure which reduces photosynthetic activity and consequently decrease transpiration and water-use efficiency of the plants (Hede et al. 2015). Therefore, higher levels of water content are closely related to chlorophyll content reduction in plants affected by pollution in the tropical forest.

In order to determine the significance of the differences of the vegetation parameters between sites and vertical canopy layers of the forest, an ANOVA was conducted at 99.9% confidence level (p < 0.001), followed by a post hoc pairwise comparison using the type I error adjustment method of Holme (see Table 4). The ANOVA identified significant differences between sites and between canopy layers for chlorophyll ( $C_{ab}$  and SPAD) and organic matter content ( $C_m$ ). Differences of brown pigments ( $P_{brown}$ ) and foliar water content ( $C_w$ ) are found only between the sites. Differences in N parameter and carotenoids ( $C_{ar}$ ) are only found between vertical canopy layers.

The Holme's pairwise comparisons indicate that chlorophyll ( $C_{ab}$  and SPAD) and foliar water content ( $C_w$ ) are the vegetation parameters exhibiting significant differences between the polluted site and the non-polluted sites. This finding confirms that plants exposed to higher levels of pollution have experienced reduced photosynthetic activity and consequently decrease transpiration and water-use efficiency of the plants, specifically chlorophyll and foliar water content. These results confirm the findings of recently published studies (Arellano et al. 2015; Hede et al. 2015) which reached the same conclusions using

different ground data and methods to estimate biophysical and biochemical parameters in tropical forest.

Pairwise comparisons across the vertical profile of the forest reveal no differences between upper and medium canopy layers. Chlorophyll ( $C_{ab}$  and SPAD), N parameter and organic matter content ( $C_m$ ) show significant differences between the understorey and the other levels of the canopy which suggest that vegetation growing in the understorey (near to the source of pollution) is more affected by the petroleum on the ground than the layers several meters above the ground.

In order to identify patterns in data and highlight similarities and differences between leaf parameters, a principal component analysis (PCA) was conducted. The first two principal components (PC1 and PC2) contain 69% of the variance (Tables 5, 6).

Figure 8 shows the relationship of PC1 and PC2 for the different leaf parameters. The vectors of  $C_{ab}$  and  $C_{ar}$  have the same direction and length which means those two variables are positively correlated; therefore, these plant pigments decrease in areas affected by petroleum pollution. On the other hand,  $C_w$  is pointing in an opposite direction which suggests that its values increase in polluted sites.

The analysis of biophysical and biochemical parameters of vegetation has revealed that the concentration of photosynthetic pigments,  $C_{ab}$  and  $C_{ar}$ , decreases significantly in areas affected by petroleum pollution in spite of the natural gradient of the vertical profile of the tropical forest. This finding is supported by ground data collected by SPAD-502 chlorophyll index which exhibit the same behaviour. Foliar water content,  $C_w$ , increased at the polluted site.

# Conclusions

In this study, field spectroscopy measurements and the PROSPECT radiative transfer model were applied to assess the effects of petroleum pollution in three study sites of the



Fig. 7 Mean and 95% confidence interval for each derived vegetation parameter per site and vertical canopy layer

Table 4ANOVA test andHolme's pairwise comparisonsof vegetation parametersbetween study sites and verticalcanopy layers

	Ν	C <sub>ab</sub>	$P_{\rm car}$	P <sub>brown</sub>	$C_{ m w}$	C <sub>m</sub>	SPAD
ANOVA							
Site	0.0607	< 0.0001	0.15	< 0.0001	0.0004	< 0.0001	< 0.0001
Layers	< 0.0001	< 0.0001	0.0089	0.492	0.738	< 0.0001	< 0.0001
Holms pairwise comparison							
Polluted—unpolluted	0.054	0.022	0.18	0.004	0.0317	0.0877	0.0013
Polluted-pristine forest	0.363	< 0.0001	0.47	0.054	0.0005	0.0018	< 0.0001
Unpolluted—pristine forest	0.134	0.26	0.47	< 0.0001	0.9899	0.0003	0.7049
Holme's pairwise comparison							
Upper-medium	0.41	0.17	0.2043	0.72	1	0.7635	0.059
Upper-understorey	< 0.0001	< 0.0001	0.0869	0.97	1	0.0001	< 0.0001
Medium-understorey	< 0.0001	< 0.0001	0.0065	0.97	1	0.0001	< 0.0001
ANOVA							
Site		***		***	***	***	***
Layers	***	***	**			***	***
Holme's pairwise comparison							
Polluted—unpolluted		*		**	*		**
Polluted-pristine forest		***			***	**	***
Unpolluted—pristine forest				***		***	
Holme's pairwise comparison							
Upper-medium							
Upper-understorey	***	***				***	***
Medium-understorey	***	***	**			***	***

Table 5 Importance of the
strongest six principal
components from the principal
component analysis of the leaf
parameters

	Importance of components								
	PC1	PC2	PC3	PC4	PC5	PC6			
Standard deviation	1.6215	1.2288	0.9858	0.6523	0.5228	0.4359			
Proportion of variance	0.4382	0.2517	0.1620	0.0709	0.0456	0.0317			
Cumulative proportion	0.4382	0.6899	0.8518	0.9228	0.9683	1.0000			

**Table 6** Eigenvalues forstrongest six principalcomponents from the principalcomponent analysis of the leafparameters

	PC1	PC2	PC3	PC4	PC5	PC6
V	0.4227	-0.2934	0.4821	-0.5758	0.0969	0.4024
Cab	0.5668	-0.0543	0.1655	0.0383	0.0877	-0.7995
Car	0.5303	0.0168	0.0204	0.7192	0.1325	0.4280
3P	-0.2371	-0.6840	-0.1415	0.1164	0.6611	-0.0728
$C_{w}$	-0.3165	-0.3799	0.6478	0.3622	-0.4423	-0.0957
C <sub>m</sub>	0.2499	-0.5464	-0.5479	-0.0700	-0.5768	0.0342

Amazon forest in Ecuador. More than 1100 samples were collected at different levels of the vertical profile of the forest in three study sites. The first two sites are secondary tropical forest, one of them is affected by petroleum pollution, and the third study site is a pristine forest in Yasuni National Park. ANOVA test, Holme's pairwise comparison and principal component analysis (PCA) revealed vegetation stress symptoms across the vertical canopy layers, particularly significantly lower levels of photosynthetic pigments, chlorophyll and carotenoid content and significantly higher levels of foliar water content, which suggests a decline of photosynthetic activity in polluted areas. Strongly positive correlation was found between chlorophyll content and the SPAD-502 index which demonstrates that the SPAD-502 chlorophyll meter can assess vegetation stress in a multispecies tropical forest.



Fig. 8 Principal components analysis (PCA) for biophysical parameters of vegetation

The results presented in this paper contribute to the very limited literature on assessing vegetation stress caused by pollution in tropical forests and this unprecedented dataset, covering a range of biophysical and biochemical leaf parameters across sites and the vertical profiles of the forest, afforded an evaluation of ecophysiological status of tropical forest, as well as the relative sensitivity of these field-deployed optical-based methods, thus informing how well these "off-the-peg" approaches might transfer between each other and support scientific investigations in these environments that are challenging but crucial to understanding and managing environmental change.

Acknowledgements The data of this paper are available upon request to the corresponding author. We acknowledge people and institutions that supported this study: Fieldwork assistants during the challenging and exhausting data collection in the Amazon: Ivan Becerra, Angel Donoso, Elias. Yasuni Research Station–Pontifical Catholic University of Ecuador: Dr. Renato Valencia, Dr. Hugo Romero, David Lasso, Carlos Padilla, Milton Pavon, Pablo Alvia and Milton Zambrano. Biologist Tatiana Avila for her valuable contribution during tree species identification. In Ecuador to Andes Petroleum Ltd., Environmental Ministry and Non-Renewable Resources Ministry. This research has been funded by the principal author and had an important contribution of the SENESCYT (National Secretariat for Science and Technology of Ecuador). H. Balzter was supported by the Royal Society Wolfson Research Merit Award, 2011/R3 and the NERC National Centre for Earth Observation.

**Open Access** This article is distributed under the terms of the Creative Commons Attribution 4.0 International License (http://crea tivecommons.org/licenses/by/4.0/), which permits unrestricted use, distribution, and reproduction in any medium, provided you give appropriate credit to the original author(s) and the source, provide a link to the Creative Commons license, and indicate if changes were made.

#### References

- Arellano P, Tansey K, Balzter H, Boyd DS (2015) Detecting the effects of hydrocarbon pollution in the Amazon forest using hyperspectral satellite images. Environ Pollut 205:225–239
- Asner GP, Martin RE (2008) Spectral and chemical analysis of tropical forests: scaling from leaf to canopy levels. Remote Sens Environ 112(10):3958–3970
- Bacour C, Baret F, Béal D, Weiss M, Pavageau K (2006) Neural network estimation of LAI, fAPAR, fCover and LAI × Cab, from top of canopy MERIS reflectance data: Principles and validation. Remote Sens Environ 105(4):313–325
- Bannari A, Khurshid KS, Staenz K, Schwarz JW (2007) A comparison of hyperspectral chlorophyll indices for wheat crop chlorophyll content estimation using laboratory reflectance measurements. IEEE Trans Geosci Remote Sens 45(10):3063–3074
- Bindi M, Hacour A, Vandermeiren K, Ojanperä K, Selldén G, Högy P, Finnan J, Fibbi L (2002) Chlorophyll concentration of potatoes grown under elevated carbon dioxide and/or ozone concentrations. Eur J Agron 17(4):319–335
- Carter GA, Spiering BA (2002) Optical properties of intact leaves for estimating chlorophyll concentration. J Environ Qual 31(5):1424–1432
- Castro-Esau KL, Sánchez-Azofeifa GA, Rivard B, Wright SJ, Quesada M (2006) Variability in leaf optical properties of mesoamerican trees and the potential for species classification. Am J Bot 93(4):517–530
- Cerovic ZG, Masdoumier G, Ghozlen NB, Latouche G (2012) A new optical leaf-clip meter for simultaneous non-destructive assessment of leaf chlorophyll and epidermal flavonoids. Physiol Plant 146(3):251–260
- Chazdon RL, Fetcher N (1984) Photosynthetic light environments in a lowland tropical rain forest in Costa Rica. J Ecol 72(2):553–564
- Clevers JGPW, De Jong SM, Epema GF, Van der Meer FD, Bakker WH, Skidmore AK, Scholte KH (2002) Derivation of the red edge index using the MERIS standard band setting. Int J Remote Sens 23(16):3169–3184
- Coste S, Baraloto C, Leroy C, Marcon É, Renaud A, Richardson AD, Roggy J, Schimann H, Uddling J, Hérault B (2010) Assessing foliar chlorophyll contents with the SPAD-502 chlorophyll meter: a calibration test with thirteen tree species of tropical rainforest in French Guiana. Ann For Sci 67(6):607p1–607p5
- Cutruneo CMNL, Oliveira MLS, Ward CR, Hower JC, De Brum IAS, Sampaio CH, Kautzmann RM, Taffarel SR, Teixeira EC, Silva LFO (2014) A mineralogical and geochemical study of three Brazilian coal cleaning rejects: demonstration of electron beam applications. Int J Coal Geol 130:33–52
- Dash J, Curran PJ (2007) Evaluation of the MERIS terrestrial chlorophyll index (MTCI). Adv Space Res 39(1):100–104
- Davidson EA, De Araüjo AC, Artaxo P, Balch JK, Brown IFC, Bustamante MM, Coe MT, Defries RS, Keller M, Longo M, Munger JW, Schroeder W, Soares-Filho BS, Souza CM Jr, Wofsy SC (2012) The Amazon basin in transition. Nature 481(7381):321–328
- Féret J, François C, Asner GP, Gitelson AA, Martin RE, Bidel LPR, Ustin SL, le Maire G, Jacquemoud S (2008) PROSPECT-4 and 5: advances in the leaf optical properties model separating photosynthetic pigments. Remote Sens Environ 112(6):3030–3043
- Gitelson AA, Keydan GP, Merzlyak MN (2006) Three-band model for noninvasive estimation of chlorophyll, carotenoids, and anthocyanin contents in higher plant leaves. Geophys Res Lett 33(11):L11402

- Grace J (1999) Environmental controls of gas exchange in tropical rain forests. In: Press MC, Scholes JD, Barker MG (eds) Physiological plant ecology. British Ecological Society, pp 367–389
- Gustafson F (1944) Is natural gas injurious to flowering plants? Plant Physiol 19:551–558
- Gustafson F (1950) Is the American Elm (*Ulmus americana*) injured by natural gas? Plant Physiol 25(422):440
- Hawkins TS, Gardiner ES, Comer GS (2009) Modeling the relationship between extractable chlorophyll and SPAD-502 readings for endangered plant species research. J Nat Conserv 17(2):125–129
- Hede ANH, Kashiwaya K, Koike K, Sakurai S (2015) A new vegetation index for detecting vegetation anomalies due to mineral deposits with application to a tropical forest area. Remote Sens Environ 171:83–97
- Hoel BO (1998) Use of a hand-held chlorophyll meter in winter wheat: evaluation of different measuring positions on the leaves. Acta Agric Scand Sect B Soil Plant Sci 48(4):222–228
- Horvitz L (1972) Vegetation and geochemical prospecting for petroleum. Am Assoc Pet Geol Bull 56(5):925–940
- Hower JC, O'Keefe JMK, Henke KR, Wagner NJ, Copley G, Blake DR, Garrison T, Oliveira MLS, Kautzmann RM, Silva LFO (2013) Gaseous emissions and sublimates from the Truman Shepherd coal fire, Floyd County, Kentucky: a re-investigation following attempted mitigation of the fire. Int J Coal Geol 116–117:63–74
- International Standard Organization-ISO (2012) Determination of transmittance by diffuse reflectance measurement, ISO 22891:2012(E)
- Jacquemoud S (2009) PROSPECT SAIL models: a review of use for vegetation characterization. Remote Sens Environ 113(1):S56– S66
- Jacquemoud S, Marcq S (2012) Personal comunication. Institut de Physique du Globe de Paris & Université Paris Diderot
- Konica Minolta (2009) Chlorophyll meter SPAD-502Plus. Instruction manual, Manual edn, Konica Minolta Sensing, Inc, Japan
- Kumar L, Schmidt K, Dury S (2006) Imaging spectrometry and vegetation science. In: van del Meer F, de Jong S (eds) Imaging spectrometry. Basic principles and prospective applications, 4th edn. Springer, Dordrecht, pp 111–155
- Lang HR, Aldman WH, Sabins FF (1985a) Patrick draw, Wyomingpetroleum test case report. American Association of Petroleum Geologists (AAPG), Tulsa
- Lang HR, Curtis JB, Kavacs JC (1985b) Lost River, West Virginia petroleum test site report. American Association of Petroleum Geologists (AAPG), Tulsa
- Longman KA, Jeník J (1987) Tropical forest and its environment. Tropical ecology series, 2nd edn. Longman Scientific and Technical, Harlow, Essex (co-published by John Wiley, New York), p 347
- Malhi Y, Roberts JT, Betts RA, Killeen TJ, Li W, Nobre CA (2008) Climate change, deforestation, and the fate of the Amazon. Science 319(5860):169–172
- Marenco RA, Antezana-Vera SA, Nascimento HCS (2009) Relationship between specific leaf area, leaf thickness, leaf water content and SPAD-502 readings in six Amazonian tree species. Photosynthetica 47(2):184–190
- Martin ME, Aber JD (1997) High spectral resolution remote sensing of forest canopy lignin, nitrogen, and ecosystem processes. Ecol Appl 7(2):431–443
- Marwell J, Osterman J, Mitchell J (1995) Calibration of the Minolta SPAD-502 leaf chlorophyll meter. Photosynth Res 46:467–472
- Melo MR, Concalves FT, Babinski NA, Miranda FP (1996) Hydrocarbon prospecting in the Amazon rainforest: application of surface geochemical, microbiological, and remote sensing methods. In: Schumecher D, Abrams MA (eds) Hydrocarbon

migration and its near-surface expressions. American Association of Petroleum Geologists (AAPG), Tulsa, pp 401–411

- Monje OA, Bugbee B (1992) Inherent limitations of nondestructive chlorophyll meters: a comparison of two types of meters. HortScience 27(1):69–71
- Morton DC, Nagol J, Carabajal CC, Rosette J, Palace M, Cook BD, Vermote EF, Harding DJ, North PRJ (2014) Amazon forests maintain consistent canopy structure and greenness during the dry season. Nature 506(7487):221–224
- Noomen MF (2007) Hyperspectral reflectance of vegetation affected by underground hydrocarbon gas seepage, PhD edn. International Institute for Geo-Information Science & Earth Observation-ITC, Enschede
- Noomen MF, Skidmore AK (2009) The effects of high soil CO<sub>2</sub> concentrations on leaf reflectance of maize plants. Int J Remote Sens 30(2):481–497
- Noomen MF, Skidmore AK, van der Meer FD, Prins HHT (2006) Continuum removed band depth analysis for detecting the effects of natural gas, methane and ethane on maize reflectance. Remote Sens Environ 105(3):262–270
- Noomen MF, van der Werff HMA, van der Meer FD (2012) Spectral and spatial indicators of botanical changes caused by long-term hydrocarbon seepage. Ecol Inform 8:55–64
- Oliveira MLS, Ward CR, French D, Hower JC, Querol X, Silva LFO (2012a) Mineralogy and leaching characteristics of beneficiated coal products from Santa Catarina, Brazil. Int J Coal Geol 94:314–325
- Oliveira MLS, Ward CR, Izquierdo M, Sampaio CH, de Brum IAS, Kautzmann RM, Sabedot S, Querol X, Silva LFO (2012b) Chemical composition and minerals in pyrite ash of an abandoned sulphuric acid production plant. Sci Total Environ 430:34–47
- Oliveira MLS, Ward CR, Sampaio CH, Querol X, Cutruneo CMNL, Taffarel SR, Silva LFO (2013) Partitioning of mineralogical and inorganic geochemical components of coals from Santa Catarina, Brazil, by industrial beneficiation processes. Int J Coal Geol 116–117:75–92
- Ollinger SV, Smith M (2005) Net primary production and canopy nitrogen in a temperate forest landscape: an analysis using imaging spectroscopy, modeling and field data. Ecosystems 8(7):760–778
- Peñuelas J, Filella L (1998) Technical focus: visible and near-infrared reflectance techniques for diagnosing plant physiological status. Trends Plant Sci 3(4):151–156
- Phillips OL, Aragão LEOC, Lewis SL, Fisher JB, Lloyd J, López-González G, Malhi Y, Monteagudo A, Peacock J, Quesada CA, Van Der Heijden G, Almeida S, Amaral I, Arroyo L, Aymard G, Baker TR, Bánki O, Blanc L, Bonal D, Brando P, Chave J, De Oliveira ÁCA, Cardozo ND, Czimczik CI, Feldpausch TR, Freitas MA, Gloor E, Higuchi N, Jiménez E, Lloyd G, Meir P, Mendoza C, Morel A, Neill DA, Nepstad D, Patiño S, Peñuela MC, Prieto A, Ramírez F, Schwarz M, Silva J, Silveira M, Thomas AS, Steege HT, Stropp J, Vásquez R, Zelazowski P, Dávila EA, Andelman S, Andrade A, Chao K, Erwin T, Di Fiore A, Honorio EC, Keeling H, Killeen TJ, Laurance WF, Cruz AP, Pitman NCA, Vargas PN, Ramírez-Angulo H, Rudas A, Salamão R, Silva N, Terborgh J, Torres-Lezama A (2009) Drought sensitivity of the amazon rainforest. Science 323(5919):1344-1347
- Poorter L, Kwant R, Hernández R, Medina E, Werger MJA (2000) Leaf optical properties in Venezuelan cloud forest trees. Tree Physiol 20(8):519–526
- Quispe D, Pérez-López R, Silva LFO, Nieto JM (2012) Changes in mobility of hazardous elements during coal combustion in Santa Catarina power plant (Brazil). Fuel 94:495–503

- R Core Team (2012) R: a language and environment for statistical computing, Software edn. R Foundation for Statistical Computing, Vienna
- Ribeiro J, DaBoit K, Flores D, Ward CR, Silva LFO (2010) Identification of nanominerals and nanoparticles in burning coal waste piles from Portugal. Sci Total Environ 408(23):6032–6041
- Ribeiro J, Taffarel SR, Sampaio CH, Flores D, Silva LFO (2013) Mineral speciation and fate of some hazardous contaminants in coal waste pile from anthracite mining in Portugal. Int J Coal Geol 109–110:15–23
- Richardson AD (2002) Changes in foliar spectral reflectance and chlorophyll fluorescence of four temperate species following branch cutting. Tree Physiol 22(7):499–506
- Richardson AD, Duigan SP, Berlyn GP (2002) An evaluation of noninvasive methods to estimate foliar chlorophyll content. New Phytol 153(1):185–194
- Roberts J, Cabral OMR, Ferreira De Aguiar L (1990) Stomatal and boundary-layer conductances in an Amazonian terra firme rain forest. J Appl Ecol 27(1):336–353
- Saikia BK, Ward CR, Oliveira MLS, Hower JC, Baruah BP, Braga M, Silva LF (2014) Geochemistry and nano-mineralogy of two medium-sulfur northeast Indian coals. Int J Coal Geol 121:26–34
- Shumacher D (1996) Hydrocarbon-induced alteration of soils and sediments. In: Shumacher D, Abrams MA (eds) Hydrocarbon migration and its near-surface expression: APPG Memoir 66, pp 71–89
- Silva LFO, Oliveira MLS, Philippi V, Serra C, Dai S, Xue W, Chen W, O'Keefe JMK, Romanek CS, Hopps SG, Hower JC (2012a) Geochemistry of carbon nanotube assemblages in coal fire soot, Ruth Mullins fire, Perry County, Kentucky. Int J Coal Geol 94:206–213
- Silva LFO, Sampaio CH, Guedes A, Fdez-Ortiz De Vallejuelo S, Madariaga JM (2012b) Multianalytical approaches to the characterisation of minerals associated with coals and the diagnosis of their potential risk by using combined instrumental microspectroscopic techniques and thermodynamic speciation. Fuel 94:52–63

- Smith KL, Steven MD, Colls JJ (2004) Remote sensing techniques for monitoring plant stress responses to gas leaks. In: International gas research conference proceedings
- Smith KL, Colls JJ, Steven MD (2005a) A facility to investigate effects of elevated soil gas concentration on vegetation. Springer, Dordrecht
- Smith KL, Steven MD, Colls JJ (2005b) Plant spectral responses to gas leaks and other stresses. Int J Remote Sens 26(18):4067
- Tedersoo L, Sadam A, Zambrano M, Valencia R, Bahram M (2010) Low diversity and high host preference of ectomycorrhizal fungi in Western Amazonia, a neotropical biodiversity hotspot. ISME J 4(4):465–471
- Torres-Netto A, Campostrini E, Gonçalves de Oliveira J, Yamanishi O (2002) Portable chlorophyll meter for the quantification of photosynthetic pigments, nitrogen and the possible use for assessment of the photochemical process in *Carica papaya* L. Braz J Plant Physiol 14(3):203–210
- Torres-Netto A, Campostrini E, Gonçalves de Oliveira J, Bressan-Smith R (2005) Photosynthetic pigments, nitrogen, chlorophyll *a* fluorescence and SPAD-502 readings in coffee leaves. Sci Hortic 104(2):199–209
- Valencia R (2004) Yasuni forest dynamic plot, Ecuador. In: Losos E, Leigh J, Giles E (eds) Tropical forest diversity and dynamism: findings from a large-scale plot network. University of Chicago Press, Chicago, pp 609–628
- van der Meer F, Yang H, Kroonenberg S (2006) Imaging spectrometry and petroleum geology. In: van der Meer F, de Jong SM (eds) Imaging spectrometry. Springer, Dordrecht, pp 219–241
- Yang H (1999) Imagining spectrometry for hydrocarbon microseepage. Delft University of Technology, Delft
- Yang H, Zhang J, Van Der Meer F, Kroonenberg SB (1999) Spectral characteristics of wheat associated with hydrocarbon microseepages. Int J Remote Sens 20(2–4):807–813
- Yang H, Zhang J, Van Der Meer F, Kroonenberg SB (2000) Imaging spectrometry data correlated to hydrocarbon microseepage. Int J Remote Sens 21(1):197–202