# Structure of the author's response

•	Reply to the interactive comment by the reviewer 1 on "Regional detection of canopy nitrogen in Mediterranear
	forests using the spaceborne MERIS Terrestrial Chlorophyll Index" by Yasmina Loozen et alp.2
•	Reply to the interactive comment by the reviewer 2 on "Regional detection of canopy nitrogen in Mediterranear
	forests using the spaceborne MERIS Terrestrial Chlorophyll Index" by Yasmina Loozen et al
•	Revised manuscript with track change version: "Regional detection of canopy nitrogen in Mediterranean forests using
	the spaceborne MERIS Terrestrial Chlorophyll Index"p.27

Please note that the line numbers indicated in the replies correspond to the line numbers in the track change version of the manuscript

# Interactive comment on "Regional detection of canopy nitrogen in Mediterranean forests using the spaceborne MERIS Terrestrial Chlorophyll Index" by Yasmina Loozen et al.

### **Anonymous Referee #1**

Received and published: 5 September 2017

We would like to thank the reviewer for reviewing our manuscript.

The manuscript shows an interesting study on the use of MERIS data to analyse empirical relationships between MTCI and ground measurements of forest canopy N content and concentration. Foliar N influences a variety of important ecosystem processes so it is clear the interest of exploring the capacity for remote detection of canopy N at regional scales from space-based platforms and the potential of new generation of sensors such as those included in the Copernicus program. However, direct estimation of N in fresh vegetation using remote sensing data is challenging due to its weak effect on leaf reflectance so the influence of structural properties of the canopy and other potential confounding factors related with the input data are key issues to be explored.

### C1

The paper is well written and also well-structured and the research questions addressed are relevant and clearly fall within the scope of Biogeosciences.

### Thank you for your nice comment.

However, my main concern about this work is that, at some point, the paper could be read as a search for correlations without a thoughtful discussion on the different **confounding factors** that could potentially affect to the observed relationship between satellite and ground data and how these factors could impact the results.

We understand your concern about this paper being a search for correlations and would like to stress that we did a directed search rather than a random search. Remote sensing of canopy N (especially handheld and airborne) has already been extensively investigated and vegetation indices based on the red-edge region, on which MTCI is based, have been repeatedly used (Schlemmer et al., 2013;Li et al., 2014;Cho et al., 2013;Clevers and Gitelson, 2013;Dash and Curran, 2004). This can be explained by the link between foliar nitrogen and chlorophyll content. This is mentioned in the Introduction part of the manuscript (Line 78 – 81):

Detection of foliage N status with vegetation indices is attributed to the strong link between foliar nitrogen and chlorophyll content (Schlemmer et al., 2013) and is often based on the NIR and red-edge region of the spectrum, hence similar to the ones used for chlorophyll detection (Filella and Penuelas, 1994;Dash and Curran, 2004;Clevers and Gitelson, 2013).

In this study, we want to extend on the existing analyses by including spaceborne remote sensing. In the introduction part of the manuscript, we gave more details about the complexity of the reflectance signal and we clarified why canopy structure was found to be a confounding factor for remote sensing of canopy N at canopy level (Line 82 - 93):

At canopy level, however, spectral reflectance is a complex function of vegetation cover, plant activity, water content, illumination angle, viewing angle and atmospheric composition (Kumar et al., 2006) and it is not straightforward to disentangle

the influence of nitrogen from the other contributions in the spectra. It is thus not clear how the relationships observed at the leaf level translate at the canopy level. The mechanisms possibly modifying the remote detection of foliage N status at the canopy scale are still not clearly understood (Ollinger, 2011). High correlation between canopy N and both NIR reflectance and albedo has been reported in boreal forests (Ollinger et al., 2008). However, the mechanism behind these findings is still controversial. Knyazikhin et al. (2013) argued that the observed correlation solely resulted from canopy structural differences between broad and needleleaf forests and was thus spurious. Other authors, although agreeing that canopy structure was a confounding factor to account for, stated that the NIR – canopy N relationship was not necessarily spurious and stemmed from an association between canopy N and structural traits (Ollinger et al., 2013; Townsend et al., 2013). Canopy traits are interrelated (Wright et al., 2004) and have been known to covary due to evolutionary convergence, as stated by Ollinger (2011).

In the discussion part of the manuscript, Section 4.4 "Possible confounding factors of the MTCI canopy N relationship", we discussed the possible confounding factors that could affect the MTCI – canopy N relationship, including canopy structure, biomass, and climatic conditions (Line 392 – 411):

The relationships between MTCI and both canopy N[%] and canopy N[area] were influenced by the PFT of the plots. The relationship between MTCI and canopy N[%] was stronger for DBF and mixed plots compared to EBF and ENF plots while the opposite was true for the MTCI-canopy N[area] relationship. In the ongoing discussion about the mechanisms underlying the remote detection of canopy N, some authors argued that the difference in structural properties between different PFTs was a confounding factor of the observed relationship between canopy N and remote sensing data, rendering it spurious (Knyazikhin et al., 2013). Other authors, , suggested that the role of canopy structure as confounding factor can be explained by an indirect association between canopy N and canopy structure resulting from convergent adaptive processes (Ollinger et al., 2013; Townsend et al., 2013). In this context, our analysis showed that the PFTs of the plots had an influence on the MTCI canopy N relationship in a specific type of ecosystem, namely Mediterranean forests. Other confounding factors associated with N availability that might affect the observed relationship possibly include biomass, biomass allocation, leaf area index (LAI). water availability, soil type, etc. The data from the forest inventory used in this analysis, i.e. the Catalonian National Forest Inventory, were extensively studied, showing that water availability was the most limiting factor in this region. Water availability was positively correlated with both the N[area] and N[%] in leaves, as well as with foliar and total above-ground biomass through MAP (Sardans, 2011, Sardans, 2013). The MAP also influenced the PFT distribution as DBF plots were located in wetter areas than EBF plots, which were found in wetter sites than ENF plots. Regarding the influence of PFT on the foliar biomass, DBF plots had on average 45% less foliar biomass than EBF or ENF plots (Sardans, 2013). This shows that canopy N[%] and canopy N[area] were interrelated to biomass, PFT and MAP.

A key element in this study is related with the intrinsic limitations of the input data: spatial (1) and temporal mismatch (2) but also, for example, the method used to scale from leaf to canopy N using field sampling strategies (3).

1) The reviewer is correct, since the beginning of this project we were aware of the limitation of the dataset included. We chose to use the data from the Catalonia National Forest Inventory because it includes many plots that are well spread over the forested region of Catalonia. The spatial mismatch has been addressed (in our original and revised manuscripts) by resampling both the MTCI product and the canopy N ground measurements to the same and lower spatial resolution. Then, we analysed the relationship between both datasets, taking the spatial discrepancy into account. The results showed that the correlation between the resampled canopy N and MTCI were significant regardless of the resampled pixel size, as described in the result part section 3.2 "Relationship between MTCI and canopy N data at lower spatial resolution". Moreover, a previous analysis investigated the influence of the spatial resolution on the remote sensing of canopy nitrogen. They could show that, even though the proportion of explained variance was reduced by going from high spatial resolution product to a low spatial resolution one (500 m), it was still possible to observe significant relationship between coarse spatial resolution remote sensing data and ground measurements (Lepine et al., 2016). This is mentioned in the discussion part of the manuscript in Section 4.5 (Line 424 – 426).

2) We addressed (in our original and revised manuscript) the temporal mismatch by averaging the MTCI product by month over the 10 years acquisition period, and selecting only the summer months, i.e. May-October, which corresponds to the growing season. By doing this, we decrease the influence of annual anomaly on the results. Moreover, the different selection criteria applied on the dataset, ensured that the plots that had undergone a land cover change were removed from the analysis. The consequence of this is that among the 846 plots included in the analysis, 625 were measured between 2000 and 2001, i.e. closer in time to the MTCI acquisition period (2002 – 2012), as shown in Table 1 below. Additionally, in response to a later comment in your review report, an analysis of the inter-annual variation of canopy N data has been included in the revised manuscript (figure A1, p.4 of this rebuttal).

Year	1988	1989	1990	1991	1992	1993	1994	1995	2000	2001
Number of plot	8	47	46	35	44	29	9	3	304	321
measured										

Table 1. Number of plots included in the analysis by sampling year.

3) To scale from leaf to canopy N, we use the leaf N value averaged over three individual trees as the plot canopy N value. This methodology, i.e. using leaf N concentration averaged over several individuals as the plot level value, is common (Schlerf et al., 2010). In our study 96% of the plots were monospecific and 4% of the plots contained only two species, therefore we did not weight the average by the species abundance (Smith and Martin, 2001;Townsend et al., 2003;McNeil et al., 2007).

The paragraph about the leaf sampling method (Section 2.2.1 "Canopy N data") has been changed to stress that most of the plots were monospecific (Line 146 - 148):

A proportion of 96% of the plots included in this analysis were monospecific (Sardans, 2015). 4% of the plots (n = 30) had two codominant species. For these plots, two leaf samples were collected, one for each of the codominant species found on the plots.

### We also explain how we scaled up from leaf to canopy level (Line 152 - 154):

To scale from leaf to canopy level, we used the leaf nitrogen concentration averaged over three individuals as the plot level value (Schlerf, 2010). We did not weight the average by species abundance (Smith and Martin, 2001) as only 4% of the plots had two different species.

In this work, allometric equations are used to relate the diameter of the branches to the leaves dry weight in order to estimate canopy N content. It would be interesting to discuss the accuracy of this method compared to others proposed in the literature to estimate canopy foliar mass per species at the stand level.

Thank you for your comment. When analysing this further, we found that the information provided in the original version of the manuscript about biomass calculation was incorrect. The foliar biomass data were calculated using allometric equations based on the diameter at breast height (DBH). The DBH was measured for all the trees present on the plot. This information is provided in two articles that also include data from the Catalonian National Forest Inventory (Vilà et al., 2003;Sardans et al., 2015).

The paragraph in the canopy N data Section 2.2.1 was changed accordingly (Line 155 - 158):

Along with the canopy N[%] data, we used foliar biomass data (g m-2) acquired during the same forest inventory (n=2286). The foliar biomass data were obtained for each plot from allometric equations relating the diameter at breast height to the leave dry weight. These allometric equations were species specific (Sardans et al. (2015), Table A1).

The table with the allometric relationships was included in the Appendix Section (Table A 1):

Table A 1. Allometric relationships between foliar biomass and DBH for the different species included in this analysis. DBH = Diameter at breast height (cm).

Species	Foliar biomass $= a \cdot DBH^b$				
•	a	b	n	r²	
Castanea sativa	0.032	1.669	86	0.49	
Fagus sylvatica	0.026	1.546	285	0.66	
Pinus halepensis	0.037	1.656	2420	0.65	
Pinus nigra	0.022	1.870	1641	0.65	
Pinus pinaster	0.034	1.848	169	0.67	
Pinus pinea	0.014	2.029	335	0.72	
Pinus sylvestris	0.036	1.651	2755	0.66	
Pinus uncinata	0.087	1.410	770	0.62	
Quercus canariensis	0.120	1.322	36	0.57	
Quercus faginea	0.197	0.943	170	0.40	
Quercus humilis	0.047	1.462	595	0.59	
Quercus cerrioides	0.023	1.805	138	0.73	
Quercus ilex	0.063	1.576	2151	0.60	
Quercus petraea	0.014	1.888	121	0.73	
Quercus suber	0.026	1.446	314	0.55	

It would be also interesting to know what is the inter-annual variation of N (ground measurements) in the study region in order to evaluate how this can affect to the discrepancy between timing of ground and satellite data.

We agree with you, the inter-annual variation of the ground measurements of canopy N is indeed essential due to the temporal discrepancy between our two datasets. As we have a large datasets covering the complete sampling period, studying the inter-annual evolution of the canopy N ground measurements was possible. We have mentioned this analysis in the revised manuscript (Line 222 – 224):

The inter-annual variation of canopy  $N_{[\%]}$  data was analysed for each month included in the analysis to ensure that the ground data could be related with MTCI data (Figure A 1).

We have included the inter-annual graph in the Appendix part of the manuscript:

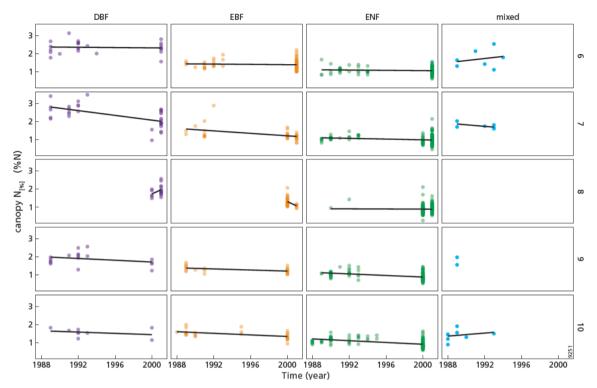


Figure A 1. Inter-annual variation of canopy N[%] (%N) for each month included in the analysis. The numbers 6-10 (right side of the figure, row numbers) refer to the month of June, July, August, September and October, respectively. DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest. Each point represents an observation at a forest plot. Note that the forest plots were not sampled multiple times, hence the inter-annual variation encompasses both temporal variation and spatial variation.

It is important to note that the forest plots were not sampled multiple times. Hence, this inter-annual variation encompasses both temporal and spatial variation and it is not possible to distinguish temporal variation alone. Figure A1 shows that trends in canopy N values over the years are rather small relative to the variation of canopy N values within a year, which includes both spatial and in between month variation. Thus, the temporal discrepancy between both datasets as well as averaging MTCI values over 10 years may have influenced the found correlations between MTCI and canopy N, but we expect the effect to be limited.

Another important issue in this work is the lack of assessment of robustness of empirical models applied using either independent data or statistical techniques (bootstrap). This may be critical when the relationships found could depend on the covariance with other variables as is typically the case in the canopy N estimation from remote sensing.

Thank you, we agree with your comment. In order to assess the robustness of the relationships between MTCI and canopy N, a leave-one-out cross validation was calculated for each of the relationships presented in the analysis. This yielded a Relative Root Mean Square Error (RRMSEcv) value giving information about the prediction error of these relationships.

The calculation of the RRMSEcv was included in the material and method part, Section 2.3.4 Statistical analysis (Line 258 – 263):

We calculated the Relative Root Mean Square Error of cross-validation (RRMSEcv, %) using the leave-one-out cross validation method (Clevers and Gitelson, 2013). Its calculation is presented in Eq. (3) following (Yao et al., 2010):

$$RRMSEcv = \sqrt{\frac{1}{n}} \times \sum_{i=1}^{n} (P_i - O_i)^2 \times \frac{100}{\overline{O}_i}$$
 (3)

where  $P_i$  represents the predicted value,  $O_i$ , the observed value,  $\bar{O}_i$  the mean of all observed value and n the total number of measurement.

The results are showed in the Table 3 and 4 of the revised manuscript, Section 3.3.1 and 3.3.2:

Table 2. Observed linear regression equations between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen concentration ( $CN_{[\%]}$ , %N) for different subgroups. Number of plots (n), determination coefficient (r2), p-value and Relative Root Mean Square Error of cross-validation (RRMEcv). PFT = Plant Functional type, DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest.

group	n	linear regression	95% confidence interval intercept	95% confidence interval slope	$\mathbf{r}^2$	p-value	RRMSEcv
overall	846	$MTCI = 2.18 + 0.79 \log(CN_{[\%]})$	[2.15, 2.20]	[0.71, 0.87]	0.32	< 0.000	17.0
DBF	80	$MTCI = 2.07 + 0.95 log(CN_{[\%]})$	[1.78, 2.36]	[0.59, 1.32]	0.25	< 0.000	12.7
EBF	186	$MTCI = 2.39 + 0.29 log(CN_{[\%]})$	[2.31, 2.48]	[0.04, 0.54]	0.03	0.021	12.4
ENF	564	$MTCI = 2.13 + 0.61 \log(CN_{[\%]})$	[2.10, 2.17]	[0.46, 0.76]	0.10	< 0.000	19.2
mixed	16	$MTCI = 2.05 + 1.35 \log(CN_{[\%]})$	[1.63, 2.46]	[0.53, 2.17]	0.47	0.003	12.4

Table 3. Observed linear regressions equations between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen content  $(CN_{lareal}, g m^2)$  for different subgroups. Number of plots (n), determination coefficient ( $r^2$ ), p-value and Relative Root Mean Square Error of cross-validation (RRMEcv). PFT = Plant Functional type, DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest.

group	n	linear regression	95% confidence interval intercept	95% confidence interval slope	$\mathbf{r}^2$	p-value	RRMSEcv
Overall	841	$MTCI = 2.08 + 0.20 log(CN_{[area]})$	[2.04, 2.12]	[0.17, 0.23]	0.17	< 0.000	18.7
DBF	80	$MTCI = 2.72 + 0.06 log(CN_{[area]})$	[2.58, 2.87]	[-0.04, 0.15]	0.02	0.263	14.7
EBF	186	$MTCI = 2.39 + 0.07 log(CN_{[area]})$	[2.32, 2.46]	[0.02, 0.12]	0.04	0.005	12.4
ENF	563	$MTCI = 1.94 + 0.20 log(CN_{[area]})$	[1.91, 1.99]	[0.17, 0.24]	0.2	< 0.000	18.2
mixed	12	$MTCI = 2.43 + 0.34 log(CN_{[area]})$	[2.05, 2.82]	[-0.26, 0.95]	0.14	0.236	12.8

We also mentioned the RRMSEcv results in the text of the Section 3.3.1 (Line 300 - 301):

The results showed that the linear regression between MTCI and canopy N[%] for the whole dataset (n = 846) was highly significant (p < 0.000) and had an  $r^2$  value of 0.32 and a RRMSEcv value of 18.7 % (Table 3).

Finally, I also miss in the discussion how the authors consider the results could be potentially useful for monitoring canopy N at regional scale considering the strength of the relationships found and the estimation errors (not analyzed in the paper).

Thank you for your comment. The goal of this case-study analysis was to explore the feasibility of canopy N detection at regional scale using MTCI. Although the relationships are modest, our study contributes to the ongoing discussion about how

to remotely sense canopy N over larger area, which could also lead to canopy N monitoring possibilities. This has been explained in the discussion part of the manuscript, Section 4.5 "Perspectives for future applications" (Line 427 – 441):

In this context, the methodology applied in this article could be a valuable alternative to explore canopy N detection at larger scale. Using published data from an extensive field plot inventory, we were able to relate both canopy  $N_{[\%]}$  and canopy  $N_{[area]}$  to MTCI at different spatial resolutions. Although the relationships found were modest, our study contributes to the ongoing discussion about how to remotely sense canopy N over larger area. As MTCI time series are readily and almost globally available, it could eventually be possible to assess our approach at a broader scale in different types of biomes. The results obtained for DBF species and Fagus sylvatica in particular suggest that this method may be efficient at estimating canopy N in temperate forests. If the strength of the relationship between MTCI and canopy N can further be improved, this could lead to canopy N monitoring possibilities at regional scale. In this context, the new sensors OLCI, onboard Sentinel 3 satellite, and especially MSI, onboard Sentinel 2 satellite might be promising due to their higher spatial resolution, from 10 to 60 m for Sentinel 2. They have bands well positioned to compute the MTCI vegetation index. Although the OLCI Terrestrial Chlorophyll Index (OTCI), the successor of the MTCI for the OLCI sensor, is already included in the OLCI level 2b reflectance image, no level 3 product (mosaicked over larger areas and temporally averaged hence similar to the MTCI time series used in this analysis) is available yet. In addition to more detailed remote sensing data, adding additional ground based canopy N observations to the regression models could better constrain these models as well. It would in particular be promising to use canopy N data over larger areas and for more diverse and globally distributed vegetation types.

Regarding your suggestion to include estimation errors, we have calculated the prediction intervals for the MTCI – canopy N relationships. These are shown in the scatterplots Figure 5 and Figure 6 of the revised manuscript:

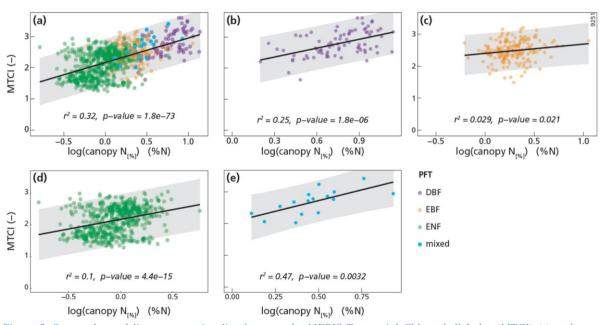


Figure 5. Scatterplot and linear regression line between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen (N) concentration (canopy  $N_{[\%]}$ , % N) for (a) whole dataset (n=846); (b) Deciduous Broadleaf Forest plots (DBF, n=80); (c) Evergreen Broadleaf Forest plots (EBF, n=186); (d) Evergreen Needleleaf Forest plots (ENF, n=564); (e) mixed forest plots (n=16). PFT = Plant functional type. The grey shading represents the prediction intervals (95 %). Canopy  $N_{[\%]}$  variable was log transformed to fulfil linear model assumptions.

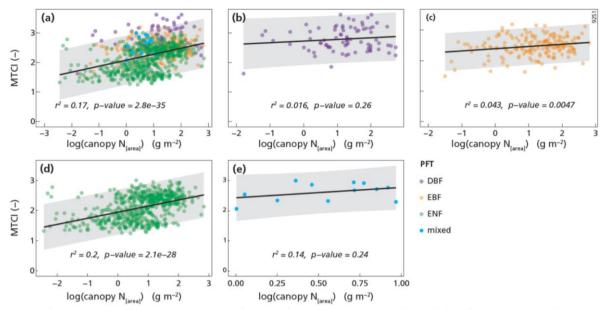


Figure 6. Scatterplot and linear regression line between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy N content (canopy  $N_{lareal}$ , g m-2) for (a) whole dataset (n = 841); (b) Deciduous Broadleaf Forest plots (DBF, n = 80); (c) Evergreen Broadleaf Forest plots (EBF, n = 186); (d) Evergreen Needleleaf Forest plots (ENF, n = 563); (e) mixed forest plots (n = 12). PFT = Plant functional type. The grey shading represents the prediction intervals (95 %). Canopy  $N_{lareal}$  variable was log transformed to fulfil linear models assumptions.

Specific comments addressing particular scientific/technical/formal issues follow:

Page 5 line 139. Complementary o alternative reference on methodology applied?

The explanation on the allometric relationship has been changed (Line 155-):

Along with the canopy N[%] data, we used foliar biomass data (g m-2) acquired during the same forest inventory (n = 2286). The foliar biomass data were obtained for each plot from allometric equations relating the diameter at breast height to the leaves dry weight. The allometric equations were species specific (Sardans et al. (2015), Table A 1).

Page 5 line 143. Correct :::.foliar biomass (N g per square meter:::.

This has been changed in the text (Line 162):

foliar biomass (dry matter g per square meter of ground area, g m<sup>-2</sup>)

### Page 5 line 153. Reword to clarify content and avoid repetitions

The sentence was clarified in the text (Line 146 - 148):

A proportion of 96% of the plots included in this analysis were monospecific (Sardans et al., 2011). 4% of the plots (n = 30) had two codominant species. For these plots, two leaf samples were collected, one for each of the codominant species found on the plots.

Page 6 line 180. Why the MERIS 300m full resolution product was not used instead?

Thank you for your question. We indeed first looked at using the MERIS 300 m full resolution reflectance images. These images were not used for our analysis for several reasons. The 300 m full resolution reflectance images available from the ESA are

not corrected for cloud cover and atmospheric influences. Moreover, there is no temporally averaged product available at full resolution. This means that one image of the 300 m full resolution reflectance data is available every three to four days from 2002 until 2012. Each of the images included in this analysis would thus need to be atmospherically corrected ( $365/4*10 \sim 912$  images). This would have been very time intensive.

In this context, the MTCI 1 km level 3 product presented several advantages. It is a readily usable product that has been corrected for atmospheric influences and cloud cover and was monthly averaged. The availability of the MTCI monthly product made it possible for us to relate the ground canopy N measurements to 10 years monthly averaged without involving time consuming images processing. We believe that this way we could decrease part of the uncertainty of relating ground measurement to any daily remotely sensed reflectance value measured several years later. Finally, the MTCI product is available for the extent of the Catalonia region in one single image, while the MERIS full resolution product can sometimes only partly cover the region and therefore each image would have had to be selected manually.

In the revised manuscript, this is more clearly explained in the Material and Methods part, Section 2.2.2 "MTCI product" of the revised manuscript (Line 200 - 211).

MTCI level 3 imagery was obtained from the NERC Earth Observation Data Centre (NEODC, 2015) for the region of Catalonia between 2002 and 2012. The original data were provided by the European Space Agency and then processed by Airbus Defence and Space. The original MERIS reflectance images, following ENVISAT specifications, have a revisit time of three days and a spatial resolution of 300 m. Compared to the original reflectance images, the MTCI processed imagery has been corrected for atmospheric influences and cloud cover (Curran and Dash, 2005) and is available as an either weekly or monthly averaged product. The spatial resolution of the processed data is approximately 1 km. As there is no temporally averaged product available at full resolution, we chose to carry out this analysis with the MTCI monthly averaged imagery. This was done to decrease the uncertainty resulting from the use of single daily reflectance values. An MTCI 10 time series of 10 years is available almost globally. One MTCI monthly averaged imagery product covering the entire study area was obtained for every month between June 2002 and March 2012, except for October 2003, when no valid product was available.

Page 7 lines 197-199. What about other land cover changes as those caused by forest fires (quite frequent in the study region), where they investigated and filtered?

The land cover changes caused by forest fires were not investigated in a separate way. As Globcover 2009 the land cover map includes a sparse vegetation class, which we believe is how the vegetation appears after a forest fire, the change due to forest fire should be accounted for when excluding sparse vegetation class from the analysis.

Page 7 Sections 2.3.2 and 2.3.3. Would be interesting to know the number of plots per pixel (average, min and max) at the different spatial resolutions.

Thank you, we agree. The number of plots per resampled pixel size are shown in the table 2 of the revised manuscript in the Result part Section 3.2 "Relationship between MTCI and canopy N data at lower spatial resolution" (Line 293 – 296):

Table 2 shows the number of plots per pixel for different pixel sizes (km). As expected, the number of plots per pixel increased with the pixel size, with a mean of 4.1 plots at 20 km spatial resolution. The descriptive statistics of the number of different PFT, species and sampling years per pixel spatial resolution are provided in the Appendix (Table A2 - A4).

Table 4. Descriptive statistics of the number of plots per pixel, for different spatial resolutions (km, pixel length). min = minimum, max = maximum, mean = average,  $sd = standard\ deviation$ .

Spatial resolution (km)	Number of plots per pixel					
Spatial resolution (Kill)	min	max	mean	sd		

5	1	6	1.44	0.77
10	1	11	2.19	1.53
15	1	15	3.11	2.59
20	1	22	4.09	3.74

Page 8 line 238. Foliar biomass is used in the calculation of canopy N content so the correlation is obviously strongest

Thank you, we agree with your comment. This was not intended to be understood as a new finding but we rather wished to be fully explicit about the correlation between the variables. The original sentence was replaced by (Line 277 - 279):

The correlation between each pair of variables was significant and the correlation between canopy  $N_{[area]}$  and foliar biomass was strongest (r = 0.88). This result was expected as the foliar biomass was included in the  $N_{[area]}$  calculation.

# Page 9 line 254. Higher instead of lower

This has been changed.

### Page 9 line 269. R2 for Quercus ilex?

The r2 value for *Quercus ilex* plots has been added in the text (Line 315):

The relationship between MTCI and canopy N[area] was also investigated for 10 individual species and one of them showed significant relationships: Ouercus ilex (r2 = 0.10, n = 160).

Page 9 Section 4.1. Could the authors elaborate here on how this could affect to the regional estimation of canopy N using new generation Sentinel-2 and 3 with improved spatial resolutions?

Due to the higher spatial resolution of the MSI sensor onboard Sentinel 2 and the bands well positioned in the red edge region, remote sensing of canopy N at regional scale might be promising. However, a pre-processed product similar to the MTCI time series should first be made available to reproduce the methodology applied in this study. This has been addressed in the discussion Section 4.5 "Perspective for larger scale applications" (Line 434-439):

In this context, the new sensors OLCI, onboard Sentinel 3 satellite, and especially MSI, onboard Sentinel 2 satellite, might also be promising due to their higher spatial resolution, from 10 to 60 m for Sentinel 2. They have bands well positioned to compute the MTCI vegetation index. Although the OLCI Terrestrial Chlorophyll Index (OTCI), the successor of the MTCI for the OLCI sensor, is already included in the OLCI level 2b reflectance image, no level 3 product similar to the MTCI time series used in this analysis, i.e. mosaicked over larger areas and temporally averaged, is available yet.

Page 11 line 315. Any hypothesis on the stronger relationship found for DBF plots? Further investigation on the proportion of the variance explained by other potential confounding factors would be desirable (same in lines 329 and 341)

The effects of the confounding factors, among which canopy structure and the differences between PFTS, on the MTCI-canopy N relationship was addressed in general in an extensive comment in the Section 4.4 "Possible confounding factors of the MTCI canopy N relationship" (Line 391 – 411):

### 4.4 Possible confounding factors of the MTCI canopy N relationship

The relationships between MTCI and both canopy N[%] and canopy N[area] were influenced by the PFT of the plots. The relationship between MTCI and canopy N[%] was stronger for DBF and mixed plots compared to EBF and ENF plots while

the opposite was true for the MTCI-canopy N[area] relationship. In the ongoing discussion about the mechanisms underlying the remote detection of canopy N, some authors argued that the difference in structural properties between different PFTs was a confounding factor of the observed relationship between canopy N and remote sensing data, rendering it spurious (Knyazikhin et al., 2013). Other authors, , suggested that the role of canopy structure as confounding factor can be explained by an indirect association between canopy N and canopy structure resulting from convergent adaptive processes (Ollinger et al., 2013; Townsend et al., 2013). In this context, our analysis showed that the PFTs of the plots had an influence on the MTCI canopy N relationship in a specific type of ecosystem, namely Mediterranean forests. Other confounding factors associated with N availability that might affect the observed relationship possibly include biomass, biomass allocation, leaf area index (LAI), water availability, soil type, etc. The data from the forest inventory used in this analysis, i.e. the Catalonian National Forest Inventory, were extensively studied, showing that water availability was the most limiting factor in this region. Water availability was positively correlated with both the N[area] and N[%] in leaves, as well as with foliar and total above-ground biomass through MAP (Sardans, 2011, Sardans, 2013). The MAP also influenced the PFT distribution as DBF plots were located in wetter areas than EBF plots, which were found in wetter sites than ENF plots (Sardans, 2013). This shows that canopy N[%] and canopy N[area] were interrelated to biomass, PFT and MAP.

Page 11 lines 332-335. This has been already stated in the results Sections. This apply for other paragraphs in this Section, authors should avoid to repeat the results and focus on the discussion.

Thank you, we agree that repeating this information several times might be unnecessary. In this instance, we wanted to remind the reader what we are going to address in the next paragraph.

Page 12 lines 152-153. I would recommend to include the analysis in this paper using information acquired in the forest inventory used for the study.

Thank you for your comment. We agree that using additional data besides canopy N and foliar biomass would make the analysis stronger. However, only biomass and foliar concentration was measured during the forest inventory. Additional physiological data related to the forest plots is thus not available.

Page 20 FIGURE 1. Please clarify if the plots represented in the map are all the forest inventory plots (2300?) or 1075 (after temporal and spatial filtering) or 846/841 finally used in the analysis. I would recommend including only the plots used in the analysis.

Thank you, the number of plots represented in the figure (n = 846) has been added to the figure caption (Line 664). The number "1075" plots was mistake from a former version of the manuscript and has been changed where it appeared in the text.

Figure 1. Map showing the forest plots (n = 846) location in the region of Catalonia, north eastern Spain. DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest.

#### References:

Cho, M. A., Ramoelo, A., Debba, P., Mutanga, O., Mathieu, R., van Deventer, H., and Ndlovu, N.: Assessing the effects of subtropical forest fragmentation on leaf nitrogen distribution using remote sensing data, Landscape Ecology, 28, 1479-1491, doi:10.1007/s10980-013-9908-7, 2013.

Clevers, J. G. P. W., and Gitelson, A. A.: 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, 2013.

- Dash, J., and Curran, P. J.: The MERIS terrestrial chlorophyll index, International Journal of Remote Sensing, 25, 5403-5413, doi:10.1080/0143116042000274015, 2004.
- Lepine, L. C., Ollinger, S. V., Ouimette, A. P., and Martin, M. E.: Examining spectral reflectance features related to foliar nitrogen in forests: Implications for broad-scale nitrogen mapping, Remote Sensing of Environment, 173, 174-186, doi:10.1016/j.rse.2015.11.028, 2016.
- Li, F., Miao, Y., Feng, G., Yuan, F., Yue, S., Gao, X., Liu, Y., Liu, B., Ustin, S. L., and Chen, X.: Improving estimation of summer maize nitrogen status with red edge-based spectral vegetation indices, Field Crops Research, 157, 111-123, doi:10.1016/j.fcr.2013.12.018, 2014.
- McNeil, B. E., Read, J. M., and Driscoll, C. T.: Foliar nitrogen responses to elevated atmospheric nitrogen deposition in nine temperate forest canopy species, Environmental Science and Technology, 41, 5191-5197, 2007.
- Sardans, J., Janssens, I. A., Alonso, R., Veresoglou, S. D., Rillig, M. C., Sanders, T. G. M., Carnicer, J., Filella, I., Farré-Armengol, G., and Peñuelas, J.: Foliar elemental composition of European forest tree species associated with evolutionary traits and present environmental and competitive conditions, Global Ecology and Biogeography, 24, 240-255, 10.1111/geb.12253, 2015.
- Schlemmer, M., Gitelson, A., Schepers, J., Ferguson, R., Peng, Y., Shanahan, J., and Rundquist, D.: 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, 2013.
- Schlerf, M., Atzberger, C., Hill, J., Buddenbaum, H., Werner, W., and Schüler, G.: Retrieval of chlorophyll and nitrogen in Norway spruce (Picea abies L. Karst.) using imaging spectroscopy, International Journal of Applied Earth Observation and Geoinformation, 12, 17-26, doi:10.1016/j.jag.2009.08.006, 2010.
- Smith, M. L., and Martin, M. E.: A plot-based method for rapid estimation of forest canopy chemistry, Canadian Journal of Forest Research, 31, 549-555, 2001.
- Townsend, P. A., Foster, J. R., Chastain Jr, R. A., and Currie, W. S.: Application of imaging spectroscopy to mapping canopy nitrogen in the forest of the central Appalachian mountains using hyperion and AVIRIS, IEEE Transactions on Geoscience and Remote Sensing, 41, 1347-1354, doi:10.1109/TGRS.2003.813205, 2003.
- Vilà, M., Vayreda, J., Gracia, C., and Ibáñez, J. J.: Does tree diversity increase wood production in pine forests?, Oecologia, 135, 299-303, 2003.

# Interactive comment on "Regional detection of canopy nitrogen in Mediterranean forests using the spaceborne MERIS Terrestrial Chlorophyll Index" by Yasmina Loozen et al.

**Anonymous Referee #2** 

Received and published: 20 October 2017

We would like to thank the reviewer for reviewing our manuscript.

General comments: The paper aims to investigate the potential of using MTCI to map regional variations in canopy nitrogen (N). The study uses field measurements of canopy N for a large number of forest plots situated across Catalonia to derive empirical relationships between N and MTCI data across a range of spatial resolutions (1 - 20 km). The study also aims to identify the influence of plant functional type on the observed relationships. Whilst the premise of the work may be interesting, there are a number of questions and comments, some of which are fairly fundamental, which I feel need addressing before this manuscript can be considered for publication. The comments are provided in the hope that they may help improve the manuscript and its subsequent impact.

Specific comments: I am not entirely convinced of the justification for reducing the spatial resolution of the MTCI data. Why degrade the 1 km product? The MERIS sensor on board ENVISAT is no longer operational (which the author's should note). The authors do note that a variation of the MTCI can be calculated from Sentinel-2 but this is a sensor with a higher spatial resolution then MERIS so what is the justification for making the data worse? Especially since the forest plots were substantially smaller than the original 1 km pixel size in the first instance. Averaging 6 m plots over a 1 km grid would "reduce small-scale variations (line 279)" so why 5, 10, 15 and 20 km also? Without this information the paper appears to be more of an academic exercise as opposed to addressing a tangible issue.

We are thankful that the reviewer finds the premise of the work interesting, and would like to use the opportunity to clarify some of the steps we took in the paper.

We indeed studied the relationship between canopy N and MTCI at different spatial resolutions. In our analysis, we used two datasets: MTCI, which has an original spatial resolution of 1 km, and canopy N ground measurements measured on 6 m diameter forest plots. In a first step, we looked at the relationship between MTCI and canopy N after resampling both datasets to the same, lower spatial resolutions, i.e. 5, 10, 15 and 20 km, removing small scale variability as values were averaged over the larger, new, resolution. The reason for doing this was to overcome the initial spatial discrepancy between our two datasets and to study the relationship between the two variables independently of the initial spatial discrepancy. This step also allowed us to study the influence of the spatial resolution on the relationship between MTCI and canopy N. The results as shown in Section 3.2 "Relationship between MTCI and canopy N data at lower spatial resolution" showed that the relationships between the variables was not strongly affected by the resampling factor. In a second step, we analysed the relationship between MTCI and canopy N at the original spatial resolution, i.e. we related each 6m plot to a 1 km MTCI pixel. During this step, we could analyse the MTCI – canopy N relationship in more detail by looking at the influence of the different PFTs on the relationship. However, during this step, the difference between MTCI and canopy N measurements in spatial resolution was retained and small-scale variation might have had an influence on the found relationship.

To make this clearer we modified the Introduction of the revised manuscript, Section 1 (Line 119 - 123), where we added the resampled pixel size:

Next, both data sets are resampled to the same, lower, spatial resolutions, i.e. 5 km, 10 k, 15 km and 20 km, in order to overcome the initial spatial discrepancy between MTCI spatial resolution (1 km) and the size of the forest plots (6 m). Subsequently, we analyse the relationship between MTCI and both canopy N concentration and canopy N content variables, both at the resampled and initial spatial resolutions. The relationships at the initial spatial resolution are then stratified according to the PFT of the plots.

We adjusted the text to better clarify the steps we took. In Section 4.1, we say that averaging multiple 6 m plots over a lower spatial resolution, i.e. 5, 10, 15 and 20 km (and not 1 plot in a 1 km pixel), would reduce small-scale variations. To make this more clear, we added the resampled pixel size in the text in section 4.1 (Line 324 - 330):

By resampling both datasets to a lower spatial resolution, i.e. 5 km, 10 km, 15 km and 20 km, the obtained values were less impacted by small-scale variations because they were obtained by averaging several values over a larger area. The results showed that the relationship between MTCI and canopy N data was significant and consistent across all spatial resolutions investigated: 5 km, 10 km, 15 km and 20 km. This showed that, when the spatial discrepancy between the original datasets, i.e. 6 m and 1 km, was taken into account, MTCI and canopy N data were linked and that the MTCI-canopy N relationship was not strongly affected by the resampled spatial resolution.

We also more clearly stated the resampled spatial resolutions in the Material and Methods part, Section 2.3.2 "Relationship between MTCI and canopy N data at lower spatial resolution" (Line 233 – 236):

In a first step, the relationships between MTCI and canopy N data values were investigated after resampling both datasets to the same, lower, spatial resolution. The resampled spatial resolutions were 5 km, 10 km, 15 km, and 20 km. This was done because of the initial difference in support size between MTCI spatial resolution and the forest plots size (i.e. 1 km and 6 m, respectively).

Finally, we agree with you that the fact that the MERIS sensor came to an end in 2012 is essential information linked to our analysis. This was also noted in the original version of the manuscript (Line 192):

While the ESA ENVISAT satellite mission producing MERIS data came to an end in 2012

One of the main justifications for the study is that "limited research has been conducted to sense canopy N in Mediterranean ecosystems and even more so in Mediterranean forests", yet there is no discussion of the importance of these ecosystems, or their N content. More information should be included to justify the significance of this sentence.

We have include justification about the ecological importance, especially regarding species diversity (Vilà-Cabrera et al., 2018), of Mediterranean forests in the introduction part of the manuscript (Line 99 - 102):

Moreover, Mediterranean forests have specific functional characteristic due to their great forest ecosystems diversity, influenced by contrasting climatic and topographic conditions, and their high tree species richness (Vilà-Cabrera et al., 2018). However, to our knowledge, limited research has been conducted to sense canopy N in Mediterranean ecosystems (Serrano et al., 2002) and even more so in Mediterranean forests.

More information is required on how the forest plot data are deemed suitable for comparison with the MTCI data. There are several questions here:

1. Is the year of data collection an issue for the correlation? Perhaps colouring the points in figure 4 based on year of in situ collection may be useful e.g. were there any climatically anomalous years that could have influenced the MTCI relationships?

Thank you for your suggestion. In the figure 4, measurements at the vegetation plots were averaged by pixel (20 km) and sampling month (over 10 years). This means that within a particular (resampled) 20 km pixel, the plots were measured in the same month. However, these plots might have been measured in different years. The Material and Methods section 2.3.2 "Relationship between MTCI and canopy N data at lower spatial resolution" (Line 243) has been edited to make this more clear.

Moreover, we agree with you the year of sampling might have an influence on the results given the temporal discrepancy between our two datasets. As we have a large datasets covering the complete sampling period, studying the inter-annual evolution of the canopy N ground measurements was possible. We have mentioned this analysis in the revised manuscript (Line 222 – 224):

The inter-annual variation of canopy  $N_{[\%]}$  data was analysed for each month included in the analysis to ensure that the ground data could be related with MTCI data (Figure A 1).

We have included the inter-annual graph in the Appendix part of the revised manuscript:

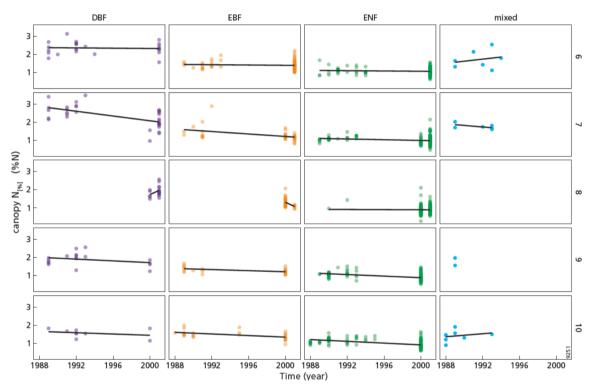


Figure A1. Inter-annual variation of canopy  $N_{[\%]}$  (%N) for each month included in the analysis. The numbers 6-10 (right side of the figure, row numbers) refer to the month of June, July, August, September and October, respectively. DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest. Each point represents an observation at a forest plot. Note that the forest plots were not sampled multiple times, hence the inter-annual variation encompasses both temporal variation and spatial variation.

It is important to note that the forest plots were not sampled multiple times. Hence, the inter-annual variation shown in Figure A1 encompasses both temporal and spatial variation and it is not possible to distinguish temporal variation alone. Figure A1 shows that trends in canopy N values over the years are rather small relative to the variation of canopy N values within a year, which includes both spatial and in between month variation. Thus, the temporal discrepancy between both datasets as well

as averaging MTCI values over 10 years may have influenced the found correlations between MTCI and canopy N, but we expect the effect to be limited.

2. How well do the 6m forest data plots represent the 1, 5, 15 and 20 km grid scales? There isn't any information as to how many points were included in each grid square when the data were resampled at each resolution. What was the distribution of values (mean, SD)?

Thank you for your comment. This is indeed important if we wish to evaluate the effectiveness of the resampling method to overcome the initial difference in support size. A table showing the descriptive statistics (min, max, mean and sd) of the number of plots per pixel, has been added to the manuscript in the Results part, section 3.2 "Relationship between MTCI and canopy N data at lower spatial resolution" (Line 293 - 296):

Table 2 shows the number of plots per pixel resampled spatial resolution (km). As expected, the number of plots per pixel spatial resolution increased with the pixel spatial resolution, with a maximum of 22 plots and a mean of 4.1 plots at 20 km spatial resolution.

Table 2. Descriptive statistics of the number of plots per pixel, by pixel spatial resolution (km). min = minimum, max = maximum, mean = average, sd = standard deviation.

	Number of plots per pixel				
Spatial resolution (km)	min	max	mean	sd	
5	1	6	1.44	0.77	
10	1	11	2.19	1.53	
15	1	15	3.11	2.59	
20	1	22	4.09	3.74	

Even though the number of plots per pixel may be low, we obtained significant correlations for all spatial resolutions studied. Please note that we did not resample to 1 km, as 1 km is the original spatial resolution of the MTCI product.

3. Can homogenous species plots be observed from satellite imagery at 5-20 km resolution? Surely the plots are going to be mixed species at this scale?

Indeed, the plots are likely to be mixed species and mixed PFT too. This is the reason why the analysis by species and PFT was not carried out at this step of the analysis (where we resample both MTCI and plot measurements to the same resolution of 5-20 km), but only in the step where the original MTCI data were used (1 km resolution).

To show the mixing effect at the resampled resolutions, we have incorporated two tables showing the average number of different PFT and species per resampled pixel. These tables are included in the Appendix of the manuscript (Table A 2 and Table A 3 of the revised manuscript):

Table A 2. Descriptive statistics of the number of plant functional types (PFT) per pixel, by pixel spatial resolution (km). min = minimum, max = maximum, mean = average,  $sd = standard\ deviation$ .

Spatial resolution (km)		Number of I	PFT per pixel	
Spatial resolution (km)	min	max	mean	sd
5	1	3	1.08	0.29
10	1	4	1.22	0.48

15	1	4	1.34	0.61
20	1	4	1.45	0.69

Table A 3. Descriptive statistics of the number of species per pixel, by pixel spatial resolution (km). min = minimum, max = maximum, mean = average, sd = standard deviation.

C	Number of species per pixel					
Spatial resolution (km)	min	max	mean	sd		
5	1	4	1.14	0.41		
10	1	4	1.38	0.67		
15	1	4	1.58	0.85		
20	1	6	1.79	1.07		

The results presented, whilst statistically significant have quite low r2 values, which indicates that the precision with which N can be predicted will be low, even though there is a statistically significant relationship between the two variables. The authors do not comment on this but I think they should as this has practical implications for their suggested approach.

It would be useful for the authors to suggest possible reasons why the reported statistically significant regressions are only explain 20 - 30% of the variation at best.

Thank you for your comment. The obtained r2 are indeed low, between 0.10 and 0.40. We would like to stress that other studies report similar or sometimes lower r2, for analyses conducted at higher spatial resolution and in more controlled conditions (Cho et al., 2013; Wang et al., 2016). The obtained results were compared with existing literature in the section 4.2.1 "Canopy N concentration detection". This section was modified to stress the differences in spatial resolution (Line 335 - 344):

The performance of the MTCI vegetation index to detect canopy N[%] in Mediterranean vegetation was similar to the results obtained from previous studies using spaceborne MTCI at higher spatial resolution. For example, using MTCI computed from the spaceborne RapidEye sensor at 5 m spatial resolution, it was possible to detect canopy N[%] in grassland savannah and sub-tropical forest with similar coefficients of determination, r2=0.35 and r2=0.52, respectively (Ramoelo et al., 2012;Cho et al., 2013). However, while there is a consensus regarding MTCI ability for in situ leaf or canopy N[%] detection in a variety of crops using handheld spectrometers (Tian et al., 2011;Li et al., 2014), there is no general agreement about MTCI ability for canopy N[%] detection across vegetation and sensor types at larger scales. For example, MTCI computed from airborne data at 3 m spatial resolution could not be related to canopy N[%] from a mixed temperate forest (Wang et al., 2016). In this context our finding brings new insight into MTCI N[%] sensing capabilities at a much coarser spatial resolution (1 km) compared to what has been done before.

We agree with the reviewer that there is indeed considerable prediction uncertainty. In the revised manuscript we have added prediction intervals for the MTCI – canopy N relationships. These are shown in the scatterplots Figure 5 and Figure 6 of the revised manuscript:

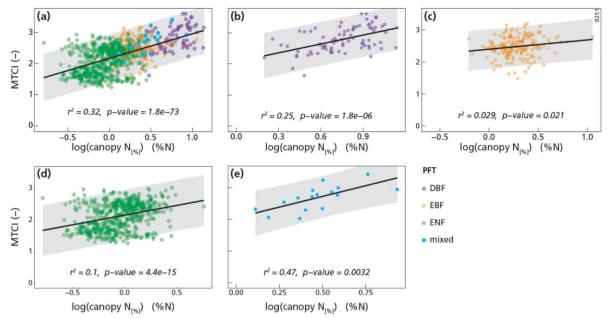


Figure 5. Scatterplot and linear regression line between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen (N) concentration (canopy  $N_{[\%]}$ , % N) for (a) whole dataset (n = 846); (b) Deciduous Broadleaf Forest plots (DBF, n = 80); (c) Evergreen Broadleaf Forest plots (EBF, n = 186); (d) Evergreen Needleleaf Forest plots (ENF, n = 564); (e) mixed forest plots (n = 16). PFT = Plant functional type. The grey shading represents the prediction intervals (95 %). Canopy  $N_{[\%]}$  variable was log transformed to fulfil linear model assumptions.

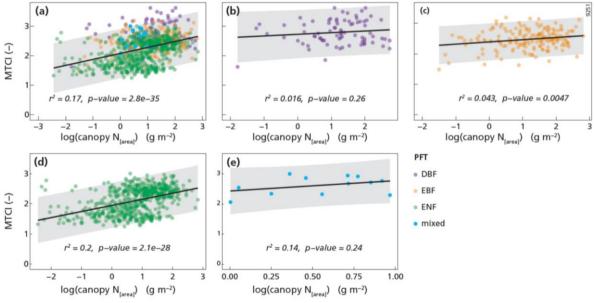


Figure 6. Scatterplot and linear regression line between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy N content (canopy  $N_{lareal}$ , g m-2) for (a) whole dataset (n = 841); (b) Deciduous Broadleaf Forest plots (DBF, n = 80); (c) Evergreen Broadleaf Forest plots (EBF, n = 186); (d) Evergreen Needleleaf Forest plots (ENF, n = 563); (e) mixed forest plots (n = 12). PFT = Plant functional type. The grey shading represents the prediction intervals (95%). Canopy  $N_{lareal}$  variable was log transformed to fulfil linear models assumptions.

The prediction intervals may be wide but still show that the MTCI has considerable information content for estimating canopy N. We would like to stress that the goal of this case-study analysis was to explore the feasibility of canopy N detection at regional scale using MTCI. Although the statistical relationships are modest, the results provide spatio-temporal indicators of canopy N and we believe that this analysis provides information that is valuable to the ongoing discussion about the feasibility of sensing canopy N over larger spatial extent. We expect that the statistical models can be improved in future research using both improved remote sensing data at higher spatial resolution and other plot data from different types of ecosystems. This has been mentioned in the discussion part of the manuscript, Section 4.5 "Perspectives for future applications" (Line 429 – 441):

Although the relationships found were modest, our study contributes to the ongoing discussion about how to remotely sense canopy N over larger area. As MTCI time series are readily and almost globally available, it could eventually be possible to assess our approach at a broader scale in different types of biomes. The results obtained for DBF species and Fagus sylvatica in particular suggest that this method may be efficient at estimating canopy N in temperate forests. If the strength of the relationship between MTCI and canopy N can further be improved, this could lead to canopy N monitoring possibilities at regional scale. In this context, the new sensors OLCI, onboard Sentinel 3 satellite, and especially MSI, onboard Sentinel 2 satellite might be promising due to their higher spatial resolution, from 10 to 60 m for Sentinel 2. They have bands well positioned to compute the MTCI vegetation index. Although the OLCI Terrestrial Chlorophyll Index (OTCI), the successor of the MTCI for the OLCI sensor, is already included in the OLCI level 2b reflectance image, no level 3 product (mosaicked over larger areas and temporally averaged hence similar to the MTCI time series used in this analysis) is available yet. In addition to more detailed remote sensing data, adding additional ground based canopy N observations to the regression models could better constrain these models as well. It would in particular be promising to use canopy N data over larger scale areas and for more diverse and globally distributed vegetation types.

# The authors indicate that these r2 values are somewhat lower than MODIS so why not just use MODIS?

Thank you for your question. We did not use MODIS because our goal was to test the relationship for the MTCI vegetation index. Vegetation indices products available for MODIS are NDVI and EVI, which have showed lower correlation with canopy N compared to MTCI due to saturation problem at high N concentration (Schlemmer et al., 2013; Pacheco-Labrador et al., 2014).

Moreover, the study we referred to in the discussion (Line 424 - 426), did indeed get higher r2 using MODIS images. Their methodology was different as there was no temporal discrepancy between their ground measurements and the satellite image acquisition. They worked with 7 x 7 km MODIS tiles, while the MERIS MTCI level 3 product is available from the ESA for the extend of the whole region (and actually even Europe) in one single image.

#### Technical corrections:

The first sentence of the abstract is quite long. Consider fragmenting and re-wording to improve impact.

### Thank you, the sentence has been changed (Line 10):

Canopy nitrogen (N) concentration and content are linked to several vegetation processes at leaf and canopy levels. Therefore, canopy N concentration is a state variable in global vegetation models with coupled carbon (C) and N cycles.

Line 12 and throughout – Data "is" should be changed to data "are" since data are Plural This has been changed.

Line 13 etc. – The abstract should include some justification as to why the work is important. This could be more clearly explained in the abstract as opposed to simply saying x did this and we are doing that. The key question is why?

We added a sentence to specify the opportunity to use vegetation indices to detect canopy N data at larger scale (Line 14):

*Vegetation indices could be a valuable tool to detect canopy N concentration and canopy N content at larger scale.* 

Line 32: Delete "," after processes This has been changed.

Line 36: Insert "," after (N g m-2) This has been changed.

Line 49: Delete "Currently". This has been changed.

Line 49: Insert "," either side of from and sensors This has been changed.

Line 48 – 52: This is a very long sentence. Consider fragmenting.

This has been changed (Line 49 - 52):

Imaging spectrometry has proven efficient in improving N sensing capabilities at the local scale. Imaging spectrometry images are acquired from either airborne of spaceborne sensors and are analysed with different methods, including partial least squares regression (PLS), continuum removal, spectral unmixing or vegetation indices.

Line 53: No need for a new paragraph. This has been changed.

# Line 71: "were aimed" is an odd choice of words. Consider re-wording

This has been changed:

most studies were carried out in agricultural crops using MTCI values computed from in situ hyperspectral reflectance data

# Line 74: Do the authors mean "a few studies" or "few studies"? It's not clear as no references are referred to.

We mean here "few studies". A reference has been added (Line 76 - 78):

Remote detection of foliage N status has been extensively studied at the leaf scale (Hansen and Schjoerring, 2003; Ferwerda et al., 2005; Li et al., 2014) and few studies have investigated the processes underlying the relationships between vegetation indices and foliar N (Pacheco-Labrador et al., 2014).

Line 83: "stated that the NIR – canopy N relationship was not necessarily spurious as plant traits have been known to covary along the leaf economic spectrum" This statement needs further explanation. What is meant by the leaf economic spectrum?

### We have modified this sentence to make it clearer (Line 89 - 93):

Other authors, although agreeing that canopy structure was a confounding factor to account for, stated that the NIR – canopy N relationship was not necessarily spurious and stemmed from an association between canopy N and structural traits (Ollinger et al., 2013; Townsend et al., 2013). Canopy traits are interrelated (Wright et al., 2004) and have been known to covary due to evolutionary convergence, as stated by Ollinger (2011).

We also have expended the previous paragraph to include more detailed explanations on the complexity of the reflectance signal (Line 82 - 85):

At canopy level, however, spectral reflectance is a complex function of vegetation cover, plant activity, water content, illumination angle, viewing angle and atmospheric composition (Kumar et al., 2006) and it is not straightforward to disentangle the influence of nitrogen from the other contributions in the spectra. It is thus not clear how the relationships observed at the leaf level translate at the canopy level. The mechanisms possibly modifying the remote detection of foliage N status at the canopy scale are still not clearly understood (Ollinger, 2011).

Line 89: "MTCI time series could be applied to estimate canopy N at a larger scale" Be careful with the terms scale here. Do you mean over a larger spatial extent?

This has been changed (Line 98 - 99):

Due to its almost global coverage, MTCI time series could be applied to estimate canopy N over a larger spatial extent.

Line 106: Suggests that there are 1075 forest plots but line 123 suggest that there are 2300 and in line 2017 there are 846 plots. Were some removed from the sample?

Thank you for noticing this mistake. On Line 116, 846 plots should have been written in place of 1075. This has been changed. The 2300 plots (Line 137) refers to the original number of plots included in the forest inventory before applying the selection criteria explained in the Material and Methods section (Line 251 - 253).

### Line 110: What are the re-sampled resolutions and what is the justification for this?

Thank you for your comment. The resampled resolution are now clearly indicated in the text (Line 119 - 121):

Next, both data sets are resampled to the same, lower, spatial resolutions, i.e. 5 km, 10 k, 15 km and 20 km, in order to overcome the initial spatial discrepancy between MTCI spatial resolution (1 km) and the size of the forest plots (6 m).

Line 117: duplicate word "create": This has been changed.

Line 150: "Several (up to two times) " does not make sense. Several suggests three or more. Consider re-wording.

Thank you for your comment. This has been changed (Line 146 - 148):

A proportion of 96% of the plots included in this analysis were monospecific (Sardans et al., 2011). 4% of the plots (n = 30) had two codominant species. For these plots, two leaf samples were collected, one for each of the codominant species found on the plots.

Line 200: MTCI was not re-sampled as the product was already a 1 km product.

We agree with you that the initial spatial resolution of the MTCI product is 1 km. In the manuscript this is called the "higher spatial resolution". However, in our study we first analyse the relationship between MTCI and canopy N data after resampling both datasets to a lower spatial resolution (section 2.3.2 Relationship between MTCI and canopy N data at lower spatial resolution", Line 231). This was done to overcome the initial resolution discrepancy between the two datasets. To make this more clear, the resampled spatial resolution was added (Line 232 - 237):

### 2.3.2 Relationship between MTCI and canopy N data at lower spatial resolution

In a first step, the relationships between MTCI and canopy N data values were investigated after resampling both datasets to the same, lower, spatial resolution. The resampled spatial resolutions were 5 km, 10 km, 15 km, and 20 km. This was done because of the initial difference in support size between MTCI spatial resolution and the forest plots size (i.e. 1 km and 6 m, respectively). This enabled us to investigate the relationships between MTCI and canopy N data independently of differences in initial support size.

Line 303-204: "This enabled us to investigate the relationships between MTCI and canopy N data independently of differences in initial support size." I don't entirely agree. Just because they now match on a spatial grid does not mean that the difference in sampling support size no longer matters. The crucial point is how well do the 6 m forest data represent the 1 km grid scale? Anything can be re-sampled. Whether it makes sense to do so is a different question.

Thank you for your comment. We chose to include the resampling analysis in our study due to the initial spatial discrepancy between the two datasets used, i.e. the ground measurements (6 m plots) and the MTCI pixel (1 km). By resampling both dataset to a lower and equal spatial resolution, i.e. 5 km, 10 km, 15 km, and 20 km, we wanted to study the relationship between the two variables when the spatial discrepancy was accounted for. The statistical basis of our approach is that we bring both sources of information (field sampling and MTCI values) to the same support size (representative area). By averaging out point samples (plot observations) within this support size, we calculate the expectation (mean) of the canopy N at that support size. By resampling the MTCI values to that same support size, the obtained result consist of an expectation

(mean) of the MTCI at that support size. We then regressed the expected canopy N values (at the new support size) against the expected MTCI values (at the new support size). As you mention it, this process is dependent on the spatial representability of the plots within the support size. This has been addressed on the page 3 and 4 of this response. We looked at the number of plots per resampled pixel, the number of different species and PFT per resampled pixel as well as the number of different sampling years per resampled pixel. The number of plots per resampled pixel may be low, but in our opinion, is still sufficient to calculate the value representative for the resampled pixel. In our opinion, this is the best we can do to make the data sets the same regarding support.

Section 2.3.3. It seems a bit odd to investigate relationships at a lower resolution before you investigate it at the original spatial resolution.

Thank you for your comment. We choose to study the relationship at the lower spatial resolution before because we wanted to explore the relationship at higher spatial resolution in more details, i.e. by also PFT and species into account. At lower spatial resolution, this information about PFT and species is lost due to the resampling process.

Line 215: Refer to section numbers as opposed to "explained above"

The section number has been added.

Lines 219 and 220: delete the word "then" This has been changed.

Line 223: "The spatial analyses were done with the PCRaster software" It is not clear what spatial analyses were "done". Consider re-wording.

This has been added (Line 263 - 264):

Resampling both datasets as well as linking the plots to the MTCI pixels was done with the PCRaster software

Figure 3: I am not sure what the purpose of this figure is since some of the variables being correlated are actually included in the calculation of others e.g. biomass and N concentration are both used to calculate N content – they are bound to be correlated. Hence line 238 is not really a finding.

This figure was included in the manuscript to summarize the information about the forest plots dataset. We also wished to be explicit about the correlation between the variables included in the analysis. Line 238 was not meant to be understood as a new finding but rather a statement about the correlation existing between the canopy N content and the biomass. This figure also shows the skewness of the variables. The original sentence was replaced by (Line 277 - 279):

The correlation between each pair of variables was significant and the correlation between canopy  $N_{[area]}$  and foliar biomass was strongest (r = 0.88). This result was expected as the foliar biomass was included in the  $N_{[area]}$  calculation.

Line 282: I don't understand what this sentence means I'm afraid "This shows that, when the influence of the discrepancy between the original datasets was taken into account, MTCI and canopy N data were linked" what discrepancies were observed?

This sentence referred to the spatial discrepancy between the spatial resolution of the MTCI (1 km) and the forest plots (6m). The sentence has been rephrased (Line 328–330):

This showed that, when the influence of the spatial discrepancy between the original datasets, i.e. 6 m and 1 km, was taken into account, MTCI and canopy N data were linked and that the MTCI-canopy N relationship was not strongly affected by the resampled spatial resolution.

Line 294 "there is no general agreement about MTCI ability for canopy N[%] detection across vegetation and sensor types" Can the authors bring any insights as to why this may be the case? What are the issues?

Thank you. The issue is that across different studies that investigate remote sensing of canopy N at larger scale, i.e. larger than with a handheld spectrometer, the prediction accuracy of the result is highly variable. When using RapidEye at 5m resolution the prediction is similar to what we obtain, while the results obtained by Wang, 2016, even though at very high spatial resolution (3 m) were not significant. As one would maybe expect that the coarse spatial resolution might be a big obstacle to sense canopy N, our results showed that even though the spatial resolution was comparatively low (min 1km) we still get significant results. This thus adds to the discussion about canopy N remote sensing.

## The paragraph has been edited to stress these distinctions (Line 333 - 344):

The overall relationship between MTCI and canopy N[%] at 1 km spatial resolution for all the forest plots (n=846) was significant and the r2 value was equal to 0.32 (Fig. 5). This result showed that canopy N[%] could be related to MTCI in Mediterranean forests. The performance of the MTCI vegetation index to detect canopy N[%] in Mediterranean vegetation was similar to the results obtained from previous studies using spaceborne MTCI at higher spatial resolution. For example, using MTCI computed from the spaceborne RapidEye sensor at 5 m spatial resolution, it was possible to detect canopy N[%] in grassland savannah and sub-tropical forest with similar coefficients of determination, r2=0.35 and r2=0.52, respectively (Ramoelo et al., 2012; Cho et al., 2013). However, while there is a consensus regarding MTCI ability for in situ leaf or canopy N[%] detection in a variety of crops using handheld spectrometers (Tian et al., 2011; Li et al., 2014), there is no general agreement about MTCI ability for canopy N[%] detection across vegetation and sensor types at larger scales. For example, MTCI computed from airborne data at 3 m spatial resolution could not be related to canopy N[%] from a mixed temperate forest (Wang et al., 2016). In this context our finding brings new insight into MTCI N[%] sensing capabilities at a much coarser spatial resolution (1 km) compared to what has been done before.

Line 315-316 Consider re-wording. Also note that there were only 15 plots of Fagus sylvatica! Can you make such a conclusion based on relatively few samples?

Thank you. The sentence has been changed (Line 363 - 364):

Moreover, when studied separately, the results observed for Fagus sylvatica plots (n = 15) were consistent with the stronger relationship observed for DBF plots.

We agree that compared to the general size of our dataset, 15 Beech plots is relatively small subset but it provides a first indication. Furthermore, many studies into canopy N detection include very few samples in total. For example, in a mixed temperate forest, Wang et al. (2016) included 26 plots (30 x 30 m) in total. In 2008, Huber et al. studied the remote sensing of canopy in a temperate forest using 28 plots (50 x 50 m) in total. In an arid shrubland, Mitchell et al. (2012) studied 35 plots (7 x 7m). These examples concern remote sensing of canopy N in general, i.e. they do not necessarily include MTCI, nor vegetation indices and use remote sensing sensors with high spatial resolution. Nonetheless, this can still give you an impression that 15 plots is not so uncommon.

Line 348 "Other authors, although agreeing that canopy structural properties needed to be accounted for, suggested that a direct biochemical link between canopy N and reflectance data was not necessary to detect canopy N with reflectance data (Ollinger et al., 2013;Townsend et al., 2013)." What did the authors suggest was necessary?

Thank you. Ollinger et al. (2008) used overall reflectance in the NIR and found a correlation with canopy N in boreal forest. Knyazikhin et al. (2013) argued that this relationship was spurious and resulted solely from differences in canopy structures linked to differences in PFT. Ollinger et al. (2013) and Townsend et al. (2013) argued that the observed relationship was not the result of a direct biochemical mechanism between nitrogen and incoming radiation but rather of an indirect link between

nitrogen and plant structure, which would result from adaptive processes. We have modified the paragraph to add this information to the revised manuscript (Line 396 – 401):

Other authors, suggested that the role of canopy structure as confounding factor can be explained by an indirect association between canopy N and canopy structure resulting from convergent adaptive processes (Ollinger et al., 2013; Townsend et al., 2013).

Section 4.4 doesn't really come to any conclusions or suggest reasons for the PFT differences and so it is somewhat superfluous as it stands. Better to integrate this in a wider discussion or include some more detailed interpretation of the data.

Thank you for your suggestion. We have integrated the differences induced by the PFT in a wider discussion about the possible confounding factors that might influence the relationship between MTCI and canopy N. These confounding factors include biomass, canopy structure and climatic variables (Line 391 - 411):

# 4.4 Possible confounding factors of the MTCI canopy N relationship

The relationships between MTCI and both canopy N[%] and canopy N[area] were influenced by the PFT of the plots. The relationship between MTCI and canopy N[%] was stronger for DBF and mixed plots compared to EBF and ENF plots while the opposite was true for the MTCI-canopy N[area] relationship. In the ongoing discussion about the mechanisms underlying the remote detection of canopy N, some authors argued that the difference in structural properties between different PFTs was a confounding factor of the observed relationship between canopy N and remote sensing data, rendering it spurious (Knyazikhin et al., 2013). Other authors, suggested that the role of canopy structure as confounding factor can be explained by an indirect association between canopy N and canopy structure resulting from convergent adaptive processes (Ollinger et al., 2013; Townsend et al., 2013). In this context, our analysis showed that the PFTs of the plots had an influence on the MTCI canopy N relationship in a specific type of ecosystem, namely Mediterranean forests. Other confounding factors associated with N availability that might affect the observed relationship possibly include biomass, biomass allocation, leaf area index (LAI), water availability, soil type, etc. The data from the forest inventory used in this analysis, i.e. the Catalonian National Forest Inventory, were extensively studied, showing that water availability was the most limiting factor in this region. Water availability was positively correlated with both the N[area] and N[%] in leaves, as well as with foliar and total above-ground biomass through MAP (Sardans, 2011, Sardans, 2013). The MAP also influenced the PFT distribution as DBF plots were located in wetter areas than EBF plots, which were found in wetter sites than ENF plots. Regarding the influence of PFT on the foliar biomass, DBF plots had on average 45% less foliar biomass than EBF or ENF plots (Sardans, 2013). This shows that canopy N[%] and canopy N[area] were interrelated to biomass, PFT and MAP.

# Lines 359-362. I do not follow this point here. What treatments were required and what "might reveal laborious" Consider re-wording.

Thank you. This sentence refers to the different treatments applied to images obtained with imaging spectroscopy at high spatial resolution with airborne or spaceborne sensors. These images need to be corrected for the influence of the atmosphere and clouds (atmospheric correction). In addition, depending on the initial sensor swath width as well as the size of the region to investigate, the images may need to be mosaicked into an image covering a larger area than the initial image acquired by the sensor. Depending on the time period for the ground measurements, the remote sensing images may also need to be temporally averaged.

This has been added to the manuscript (Line 418 - 423):

However, due to the different treatments required as well as the limited swath width associated with the high spatial resolution (from 3 m to 30 m for Hyspex airborne and Hyperion spaceborne sensors, respectively (Wang et al., 2016; Smith et al., 2003), applying imaging spectrometry at a broader scale might reveal laborious. Depending on the sensors as well as on the extent of the study area, this might involve correcting the acquired images for atmospheric influences and cloud cover as well as combining several images into a larger scale image.

### References:

Bartlett, M. K., Ollinger, S. V., Hollinger, D. Y., Wicklein, H. F., and Richardson, A. D.: Canopy-scale relationships between foliar nitrogen and albedo are not observed in leaf reflectance and transmittance within temperate deciduous tree species, Botany, 89, 491-497, 2011.

Cho, M. A., Ramoelo, A., Debba, P., Mutanga, O., Mathieu, R., van Deventer, H., and Ndlovu, N.: Assessing the effects of subtropical forest fragmentation on leaf nitrogen distribution using remote sensing data, Landscape Ecology, 28, 1479-1491, doi:10.1007/s10980-013-9908-7, 2013.

Huber, S., Kneubühler, M., Psomas, A., Itten, K., and Zimmermann, N. E.: Estimating foliar biochemistry from hyperspectral data in mixed forest canopy, Forest Ecology and Management, 256, 491-501, doi:10.1016/j.foreco.2008.05.011, 2008.

Huber, S., Koetz, B., Psomas, A., Kneubuehler, M., Schopfer, J. T., Itten, K. I., and Zimmermann, N.: Impact of multiangular information on empirical models to estimate canopy nitrogen concentration in mixed forest, Journal of Applied Remote Sensing, 4, 2010.

Knyazikhin, Y., Schull, M. A., Stenberg, P., Mõttus, M., Rautiainen, M., Yang, Y., Marshak, A., Latorre Carmona, P., Kaufmann, R. K., Lewis, P., Disney, M. I., Vanderbilt, V., Davis, A. B., Baret, F., Jacquemoud, S., Lyapustin, A., and Myneni, R. B.: Hyperspectral remote sensing of foliar nitrogen content, Proceedings of the National Academy of Sciences, 110, E185–E192, 10.1073/pnas.1210196109, 2013.

Mitchell, J. J., Glenn, N. F., Sankey, T. T., Derryberry, D. R., and Germino, M. J.: Remote sensing of sagebrush canopy nitrogen, Remote Sensing of Environment, 124, 217-223, 2012.

Ollinger, S. V., Richardson, A. D., Martin, M. E., Hollinger, D. Y., Frolking, S. E., Reich, P. B., Plourde, L. C., Katul, G. G., Munger, J. W., Oren, R., Smith, M. L., Paw U, K. T., Bolsta, P. V., Cook, B. D., Day, M. C., Martin, T. A., Monson, R. K., and Schmid, H. P.: Canopy nitrogen, carbon assimilation, and albedo in temperate and boreal forests: Functional relations and potential climate feedbacks, Proceedings of the National Academy of Sciences of the United States of America, 105, 19336-19341, doi:10.1073/pnas.0810021105., 2008.

Ollinger, S. V., Reich, P. B., Frolking, S., Lepine, L. C., Hollinger, D. Y., and Richardson, A. D.: Nitrogen cycling, forest canopy reflectance, and emergent properties of ecosystems, Proceedings of the National Academy of Sciences, 110, E2437, 10.1073/pnas.1304176110, 2013.

Pacheco-Labrador, J., González-Cascón, R., Pilar Martín, M., and Riaño, D.: Understanding the optical responses of leaf nitrogen in mediterranean holm oak (Quercus ilex) using field spectroscopy, International Journal of Applied Earth Observation and Geoinformation, 26, 105-118, doi:10.1016/j.jag.2013.05.013, 2014.

Schlemmer, M., Gitelson, A., Schepers, J., Ferguson, R., Peng, Y., Shanahan, J., and Rundquist, D.: 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, 2013.

Townsend, P. A., Serbin, S. P., Kruger, E. L., and Gamon, J. A.: Disentangling the contribution of biological and physical properties of leaves and canopies in imaging spectroscopy data, Proceedings of the National Academy of Sciences of the United States of America, 110, 10.1073/pnas.1300952110, 2013.

Vilà-Cabrera, A., Coll, L., Martínez-Vilalta, J., and Retana, J.: Forest management for adaptation to climate change in the Mediterranean basin: A synthesis of evidence, Forest Ecology and Management, 407, 16-22, 10.1016/j.foreco.2017.10.021, 2018.

Wang, Z., Wang, T., Darvishzadeh, R., Skidmore, A. K., Jones, S., Suarez, L., Woodgate, W., Heiden, U., Heurich, M., and Hearne, J.: Vegetation indices for mapping canopy foliar nitrogen in a mixed temperate forest, Remote Sensing, 8, doi:10.3390/rs8060491, 2016.

# Regional detection of canopy nitrogen in Mediterranean forests using the spaceborne MERIS Terrestrial Chlorophyll Index

Yasmina Loozen<sup>1</sup>, Karin T. Rebel<sup>1</sup>, Derek Karssenberg<sup>2</sup>, Martin J. Wassen<sup>1</sup>, Jordi Sardans<sup>3,4</sup> and Josep Peñuelas<sup>3,4</sup>, Steven M. De Jong<sup>2</sup>

5

10

15

20

25

30

35

40

Correspondence to: Yasmina Loozen (y.m.a.loozen@uu.nl)

Abstract. Canopy nitrogen (N) concentration and content are linked to several vegetation processes at leaf and canopy levels. and Therefore, canopy N concentration is a state variable in global vegetation models with coupled carbon (C) and N cycles. While there is ample C data available to constrain the models, widespread N data issue lacking. Remote sensing and vegetation indices have been used to detect canopy N concentration and canopy N content at the local scale in grasslands and forests. Vegetation indices could be a valuable tool to detect canopy N concentration and canopy N content at larger scale. In this paper we conducted a regional case-study analysis investigating to investigate the relationship between the Medium Resolution Imaging Spectrometer (MERIS) Terrestrial Chlorophyll Index (MTCI) time series from ESA ENVISAT at 1 km spatial resolution and both canopy N concentration (%N) and canopy N content (g m<sup>-2</sup>) from a Mediterranean forests inventory in the region of Catalonia, NE of Spain. The relationships between the datasets were studied after resampling both datasets to lower spatial resolutions (20 km, 15 km, 10 km and 5 km) and at the initial higher spatial resolution of 1 km. The results at the higher spatial resolution yielded significant relationships between MTCI and both canopy N concentration and content,  $r^2 = 0.32$  and  $r^2 = 0.17$ , respectively. We also investigated these relationships per plant functional type. While the relationship between MTCI and canopy N concentration was strongest for deciduous broadleaf and mixed plots ( $r^2 = 0.25$  and  $r^2 = 0.47$ , respectively), the relationship between MTCI and canopy N content was strongest for evergreen needleleaf trees ( $r^2 = 0.20$ ). At the species level, canopy N concentration was strongly related to MTCI for European Beech plots ( $r^2 = 0.71$ ). These results present a new perspective on the application of MTCI time series for canopy N detection, ultimately leading towards the generation of canopy N maps that can be used to constrain global vegetation models.

**Keywords**: vegetation index, MERIS, foliar nitrogen concentration, foliar nitrogen content, plant functional types, Mediterranean forest, remote sensing

## 1 Introduction

Canopy nitrogen (N) concentration is an essential state variable in regional (Ollinger and Smith, 2005) and global vegetation models including both the carbon (C) and the N cycles (Zaehle and Friend, 2010;Smith et al., 2014). This variable has been linked to several vegetation traits and processes; at the leaf and canopy levels. At the leaf level, leaf N concentration, which represents the leaf N status expressed as a percentage of leaf dry matterass (%N, N g 100g<sup>-1</sup> DM), has been related to photosynthetic capacity (Evans, 1989;Reich et al., 1995;Reich et al., 1997;Reich et al., 1999;Wright et al., 2004), specific leaf area, leaf life span (Reich et al., 1999;Wright et al., 2004) and light use efficiency (Kergoat et al., 2008). Leaf N concentration expressed on a leaf area basis, also called leaf N content (N g m<sup>-2</sup>), has also been linked with chlorophyll content, Rubisco content (Evans, 1989) and

<sup>&</sup>lt;sup>1</sup>Copernicus Institute of sustainable development, Faculty of Geosciences, Utrecht University, Utrecht, The Netherlands

<sup>&</sup>lt;sup>2</sup>Physiscal geography, Faculty of Geosciences, Utrecht University, Utrecht, The Netherlands

<sup>&</sup>lt;sup>3</sup>CSIC, Global Ecology Unit CREAF-CSIC-UAB, 08913 Bellaterra, Catalonia, Spain

<sup>&</sup>lt;sup>4</sup>CREAF, 08913 Cerdanyola del Vallès, Catalonia, Spain

photosynthetic capacity (Evans, 1989;Reich et al., 1995). At stand scale, canopy nitrogen concentration, which represents the leaf N concentration averaged over the stand canopy, has also been found to correlate with above ground Net Primary Productivity (NPP) (Reich, 2012), while canopy N content has been linked with the canopy light use efficiency (Green et al., 2003).

Given their links to many vegetation processes, leaf and canopy N variables could be used to constrain N cycle modules in global vegetation models. At the global scale, ample data is available to constrain models for the C cycle; however, data to constrain the N cycle are limited. Currently, canopy N data arcis not widely available and canopy N sampling campaigns are time-consuming and thus expensive tasks. Moreover, upscaling from local sampling campaign measurements represents an additional limitation. In this perspective, local, regional or even global remotely sensed canopy N estimates will be a valuable addition, enabling us to collect information in a less time intensive and expensive manner than traditional on-field sampling campaigns. Such near global canopy N estimates will be beneficial as input in global vegetation models or to calibrate and validate these models.

DCurrently, different remote sensing techniques have been applied to detect canopy N in terrestrial vegetation. Imaging spectrometry has proven efficient in improving N sensing capabilities at the local scale. Imaging spectrometry images are acquired from either airborne orf spaceborne sensors and are analysed with different methods, including partial least squares regression (PLS), continuum removal, spectral unmixing or vegetation indices. Imaging spectrometry from either airborne or spaceborne sensors coupled with different analysis methods, including partial least squares regression (PLS), continuum removal, spectral unmixing or vegetation indices, has proven efficient in improving N sensing capabilities at the local scale (Smith et al., 2003;Ollinger et al., 2008;Huber et al., 2008;Martin et al., 2008;Schlerf et al., 2010;Wang et al., 2016). Among other techniques, ratios or normalized differences of reflectance bands in the Red and Near Infrared (NIR) regions of the spectrum, the so called vegetation indices (VI) (Glenn et al., 2008), are one of the most straightforward methods for canopy N detection. Combined with in situ hyperspectral devices, vegetation indices have been extensively used for leaf or canopy N detection in agricultural systems (Peñuelas et al., 1994;Filella et al., 1995;Hansen and Schjoerring, 2003;Tian et al., 2011;Schlemmer et al., 2013;Li et al., 2014). Vegetation indices have also been applied to airborne or spaceborne acquired imagery in natural environments (Ramoelo et al., 2012;Wang et al., 2016).

A particular vegetation index, the MERIS Terrestrial Chlorophyll Index (MTCI) has been proposed for detecting canopy N (Clevers and Gitelson, 2013). MTCI was originally computed from three reflectance bands from the Medium Resolution Imaging Spectrometer (MERIS) aboard the European Space Agency (ESA) ENVISAT satellite at a spatial resolution of 1 km. However, it can also be obtained from other sensors' reflectance data and a similar product will be available from the ESA Sentinel-2 satellite mission (Drusch et al., 2012). It was first developed to estimate chlorophyll content (Dash and Curran, 2004, 2007). Since then, other applications of this index have been described, among which the possibility to estimate Gross Primary Productivity (GPP) from natural (Harris and Dash, 2010, 2011;Boyd et al., 2012) and cultivated lands (Peng and Gitelson, 2011). Furthermore, MTCI has been used to discriminate between C3 and C4 grasses (Foody and Dash, 2007) and to monitor vegetation phenology at the sub-regional (Boyd et al., 2011) and continental scales (Rodriguez-Galiano et al., 2015;Crabbe et al., 2016). Regarding canopy N detection, most studies were carried outapplications were aimed at in agricultural crops using MTCI values computed from in situ hyperspectral reflectance data (Tian et al., 2011;Clevers and Gitelson, 2013;Li et al., 2014). A few were directed towards sensing N concentration in natural environments using airborne data, e.g. in temperate forests (Wang et al., 2016), or spaceborne data, for example in grasslands (Ramoelo et al., 2012;Ullah et al., 2012) or sub-tropical forests (Cho et al., 2013).

Remote detection of foliage N status has been extensively studied at the leaf scale (Hansen and Schjoerring, 2003; Ferwerda et al., 2005; Pacheco Labrador et al., 2014; Li et al., 2014) and a few studies have investigated the processes underlying the relationships between vegetation indices and foliarcanopy N (Pacheco-Labrador et al., 2014). Detection of foliage N status with vegetation indices is attributed to the strong link between foliar nitrogen and chlorophyll content (Schlemmer et al., 2013) and is often based on the NIR and red-edge region of the spectrum, hence similar to the ones used for chlorophyll detection (Filella and Penuelas, 1994;Dash and Curran, 2004;Clevers and Gitelson, 2013). It is not clear however how this translates at the canopy level At canopy level, however, spectral reflectance is a complex function of vegetation cover, plant activity, water content, illumination angle, viewing angle and atmospheric composition (Kumar et al., 2006) and it is not straightforward to disentangle the influence of nitrogen from the other contributions in the spectra. It is thus not clear how the relationships observed at the leaf level translate at the canopy level.ast The mechanisms possibly modifying the remote detection of foliage N status at the canopy scale are still not clearly understood (Ollinger, 2011). High correlation between canopy N and both NIR reflectance and albedo has been reported in boreal forests (Ollinger et al., 2008). However, the mechanism behind these findings is still controversial. Knyazikhin et al. (2013) argued that the observed correlation solely resulted from canopy structural differences between broad and needleleaf forests and was thus spurious. Other authors, although agreeing that canopy structure was a confounding factor to account for, stated that the NIR - canopy N relationship was not necessarily spurious Ollinger et al., 2013; Townsend et al., 2013) and stemmed from an association between canopy N and structural traits (Ollinger et al., 2013; Townsend et al., 2013). as Canopyplant traits are interrelated (Wright et al., 2004) and have been known to covary due to evolutionary convergence, as stated by Ollinger (2011). along the leaf economic spectrum (Wright et al., 2004; Ollinger et al., 2013; Townsend et al., 2013).

85

90

95

100

105

110

115

120

In this context, there are several knowledge gaps that we would like to address in this paper. First, although 1 km spatial resolution spaceborne MTCI time series are available from the ESA, MTCI has mainly been employed to detect canopy N in agricultural applications with in situ devices and rarely in a broader range of natural ecosystems and scales using spaceborne data. Due to its almost global coverage, MTCI time series could be applied to estimate canopy N over a larger spatial extentat a larger seale. Moreover, Mediterranean forests have specific functional characteristic due to their great forest ecosystems diversity, influenced by contrasting climatic and topographic conditions, and their high tree species richness (Vilà-Cabrera et al., 2018). However, to our knowledge, limited research has been conducted to sense canopy N in Mediterranean ecosystems (Serrano et al., 2002) and even more so in Mediterranean forests. In addition, although in a temperate forest the reflectance spectrum of individual plant functional types (PFT) has been shown to be different (Wang et al., 2016), the relationship between MTCI and canopy N has seldom been studied and compared between PFTs. Moreover, investigating the influence of PFTs on this relationship might give further insight into the influence of structural effects in canopy N detection. Finally, the difference between sensing canopy N concentration ( $N_{[\%]}$ , %N) and canopy N content ( $N_{[area]}$ , g m<sup>-2</sup>) has rarely been investigated. The relationship between MTCI and both of these variables has been studied separately (Clevers and Gitelson, 2013; Wang et al., 2016), but very few analyses (Mirik et al., 2005; Ullah et al., 2012) have compared the ability to detect canopy N concentration and canopy N content simultaneously, especially in forest ecosystems. The objective of our study is thus to investigate the relationship between the spaceborne MTCI remote sensing product and canopy N in Mediterranean forests at the regional scale. More specifically, the relationships between MTCI and both canopy N concentration and canopy N content are investigated and compared. We then also examine these relationships per PFT and at the species level.

Detection of canopy N is often limited to local scale studies due to the spatial restrictions associated with N data acquisition in the field and treatment of high spatial resolution remote sensing imagery with limited spatial coverage (Lepine et al., 2016). Our case-study exploits the broadly and readily available MTCI time series at 1 km spatial resolution from the ESA ENVISAT mission and combines it with canopy N data, both concentration and content, from 8461075 forest plots measured between 1988 and 2001 by the Catalonian National Forest Inventory (Gracia et al., 2004). First, we develop a methodology to overcome the time discrepancy between our two sets of data. Next, both data sets are resampled to the same, lower, spatial resolutions, i.e. 5 km, 10 k, 15 km and 20 km, in order to overcome the initial spatialscale discrepancy between MTCI spatial resolution (1 km) and the size of the forest plots (6 m). Subsequently, we analyse the relationship between MTCI and both canopy N concentration and canopy N content variables, both at the resampled and initial spatial resolutions. These relationships at the initial spatial resolution are then stratified according to the PFT of the plots PFT. The results are presented and discussed. Finally, we address the implications for future research and draw a conclusion.

### 2 Material and methods

# 2.1 Study area

125

130

135

140

145

150

155

160

Our study area corresponds to the region of Catalonia (Fig. 1) which is located in north eastern Spain and has a spatial extent of 32,114 km² (Sardans et al., 2011). While the region is characterised by a Mediterranean climate, the presence of the Pyrenees to the northwest and the Mediterranean Sea to the east creates create contrasting climate conditions with an altitudinal gradient from north to south and a continental gradient from west to east. Following this pattern, the mean annual temperature varies from 1 °C in the north to 17 °C in the south (Sardans et al., 2011). While mean annual precipitation (MAP) is 1400 mm in the Pyrenees, in the south, the MAP is lower than 350 mm (Sardans et al., 2011), leading to seasonal drought (Lana and Burgueño, 1998) and fires (González and Pukkala, 2007), impacting the vegetation (Liu et al., 2015).

### 2.2 Data collection

# 2.2.1 Canopy N data

The canopy N data used in this research was collected by the Ecological and Forestry Applications Research Centre (CREAF), Universitat Autònoma de Barcelona. The data included 2300 closed canopy forest plots sampled between 1988 and 2001 by the Catalonian National Forest Inventory (Gracia et al., 2004).

The forest plots (Fig. 1) had a minimum diameter of 6 m, which varied depending on the tree density in order to include between 15 and 25 trees with a diameter at breast height (DBH) of at least 5 cm. The DBH was recorded for all the trees present on the plot with a DBH of minimum 5 cm. The plots were investigated for canopy N concentration (N<sub>[%]</sub>, %N) defined as g of N per 100 g of dry leaf dry matter. Each leaf sample was constituted by the leaves of at least three different trees of the dominant tree species in the canopy. The leaf samplesves were collected from the upper central part of the crown using extensible loppers. All foliar cohorts present in the canopy were included in the leaf sample. Each leaf sample was constituted by the leaves of at least three different trees of the dominant tree species in the canopy. The species dominance was determined by the tallest individual. A proportion of 96% of the plots included in this analysis were monospecific (Sardans et al., 2011). 4% of the plots (n = 30) had two codominant species. For these plots, two leaf samples were collected, one for each of the codominant species found on the plots.

The <u>leaf</u> samples were dried and then ground using a Braun Mikrodismembrator-U (B. Braun Biotech International, Melsungen, Germany). They were analysed for foliar N concentration using the combustion technique coupled to

gas chromatography using a Thermo Electron Gas Chromatograph (model NA 2100, CE Instruments-Thermo Electron, Milan, Italy) (Gracia et al., 2004). To scale from leaf to canopy level, we used the leaf nitrogen concentration averaged over three individuals as the plot level value (Schlerf et al., 2010). We did not weight the average by species abundance (Smith and Martin, 2001) as only 4% of the plots had two different species.

Along with the canopy  $N_{[\%]}$  data, we used foliar biomass data (g m<sup>-2</sup>) acquired during the same forest inventory (n = 2286). The foliar biomass data were obtained for each plot from allometric equations relating the diameter at of the branches to theat breast height to the leaves dry weight. The allometric equations were species specific (Gracia et al. (2004), Fig. A1). (Sardans et al. (2015), Table A 2). These foliar biomass data were used to calculate canopy N content ( $N_{[area]}$ , g of N per m<sup>-2</sup> of ground) for each plot following Eq. (1):

canopy 
$$N_{[area]} = \frac{\text{canopy } N_{[\%]} * fbiom}{100},$$
 (1)

where *canopy*  $N_{[area]}$  is the canopy N content (N g per square meter of ground area, g m<sup>-2</sup>), *canopy*  $N_{[\%]}$  is the canopy N concentration (%N) and *fbiom* is the foliar biomass (<u>dry matter</u>N g per square meter of ground area, g m<sup>-2</sup>).

For the plots with two codominant species, the concentration measurements. After were done separately separate

measurements, the obtained foliar N concentration values and biomass values—were then averaged in order to obtain a single canopy N<sub>[%]</sub> and canopy<sub>[area]</sub> value for each plot with twoseveral codominant species. Among these 30 plots with codominant species, 16 plots had codominant species from different PFT. Their PFT is thus labelled as mixed while the plots with several codominant species from the same PFT are labelled according to their PFTs. Catalonian forests include both deciduous and evergreen broadleaf as well as evergreen needleleaf tree species. These three PFTs are referred to as Deciduous Broadleaf Forest (DBF), Evergreen Broadleaf Forest (EBF) and Evergreen Needleleaf Forest (ENF), respectively. The main tree species are *Pinus halepensis* Mill., *Pinus sylvestris* L., *Quercus ilex* L., *Pinus uncinata* Ramond ex DC., *Pinus nigra* J.F. Arnold, *Quercus suber* L., *Quercus cerrioides* Willk. & Costa., *Quercus petraea* Liebl. and *Fagus sylvatica* L. These species accounted for 92% of the sampled forest plots. The 15 tree species included in this analysis are listed in Table 1. Plots with a rare dominant tree species, i.e. species that were detected in only one single plot, were excluded from the analysis. This applied to plots with these dominant species: *Abies alba* Mill., *Fraxinus augustifolia* Vahl, *Fraxinus excelsior* L., *Pinus radiata* D. Don, *Populus nigra* L., *Populus tremula* L., *Quercus robur* L.

A proportion of 96% of the plots included in this analysis had a single dominant tree species. The plots with codominant species (n = 30) were sampled several (up to two) times, each time for each of the codominant tree species found on the plots. After separate measurements, the obtained foliar N concentration values were averaged in order to obtain a single canopy N<sub>[%]</sub> value for each plot with several codominant species. Among these 30 plots with codominant species, 16 plots had codominant species from different PFT. Their PFT is thus labelled as mixed while the plots with several codominant species from the same PFT are labelled according to their PFTs.

### 2.2.2 MTCI product

170

175

180

185

190

195

200

The MERIS Terrestrial Chlorophyll Index (MTCI) was first developed to estimate chlorophyll content in canopies. MTCI is has been described to be sensitive to high chlorophyll content while presenting low sensitivity to soil brightness (Curran and Dash, 2005). Its calculation, presented in Eq. (2), is based on three reflectance bands, located around the red edge point (REP) (Dash and Curran, 2004):

$$MTCI = \frac{R_{band10} - R_{band9}}{R_{band9} - R_{band8}} = \frac{R_{753.75} - R_{708.75}}{R_{708.75} - R_{681.25}}$$
(2)

where  $R_{band8}$ ,  $R_{band9}$  and  $R_{band10}$  represent the 8<sup>th</sup>, 9<sup>th</sup> and 10<sup>th</sup> bands of MERIS, respectively. Following MERIS standard bands settings, the centres of the bands were located at 681.25 nm, 708.75 nm and 753.75 nm on the electromagnetic spectrum.

While the ESA ENVISAT satellite mission producing MERIS data came to an end in 2012, MERIS products and MTCI in particular are still relevant because the new ESA Sentinel-2 satellite mission has improved band settings compared to those of MERIS and increased the spatial resolution to 20 m (Drusch et al., 2012). The Sentinel-2 mission will also release a chlorophyll product that will continue the time series already available for MTCI. In this study, we put emphasis on ENVISAT-MERIS as our field data are closer to the MERIS acquisition period.

MTCI has been described to be sensitive to high chlorophyll content while presenting low sensitivity to soil brightness. MTCI 10 year time series are available almost globally and are already corrected for atmospheric influences and cloud cover (Curran and Dash, 2005).

MTCI level 3 imagery was obtained from the NERC Earth Observation Data Centre (NEODC, 2015) for the region of Catalonia between 2002 and 2012. The original data were provided by the European Space Agency and then processed by Airbus Defence and Space. The original MERIS reflectance images—data, following ENVISAT specifications, have a revisit time of three days and a spatial resolution of 300 m.—and the processed imagery is available as an either weekly or monthly averaged product. The spatial resolution of the processed data is approximately 1 km. Compared to the original reflectance images, the MTCI processed imagery has been corrected for atmospheric influences and cloud cover (Curran and Dash, 2005) and is available as an either weekly or monthly averaged product. The spatial resolution of the processed data is approximately 1 km. As there is no temporally averaged product available at full resolution, we chose to carry out this analysis with the MTCI monthly averaged processed imagery. This was done to decrease the uncertainty resulting from the use of single daily reflectance values. The spatial resolution of the processed data is approximately 1 km. An MTCI time series of 10 years is available almost globally. One MTCI monthly averaged imagery product covering the entire study area was obtained for every month between June 2002 and March 2012, except for October 2003, when no valid product was available.

## 2.3 Data handling

205

210

215

220

225

230

235

240

# 2.3.1 Methodology to link canopy N data to MTCI values

There is a discrepancy between the timing of the ground truth sampling and the satellite image acquisition period. While the plot sampling campaigns were carried out between 1988 and 2001, the ENVISAT satellite mission was launched in 2002 and ended in 2012. To overcome the discrepancy, MTCI images were averaged by month over the 10 years of the satellite mission period. This process yielded twelve MTCI averaged images, one for each month. The averaged MTCI images were then linked to the forest plots based on the forest plot coordinates and sampling month, as the exact sampling date was known for each plot. The period between the 1st of June and the 31st of October was determined to be the growing season after a pre-analysis, where we studied yearly temporal variation of MTCI in several locations and forest types in Catalonia.—This extended period was chosen to encompass the different vegetation phenology types corresponding to the contrasted climate conditions in this region. The forest plots sampled outside of the growing season were excluded from the analysis. The inter-annual variation of canopy N<sub>[96]</sub> data was analysed for each month included in the analysis to ensure that the ground data could be related with MTCI data (Figure A 2). The Globcover 2009 land cover map was used to exclude forest plots located on unsuitable

land surface. The Globcover map was created by ESA using MERIS reflectance data from 2009 (Bontemps et al., 2011). It was downloaded from the ESA data user elements website (ESA, 2010). This map comprises 22 land cover classes and has a spatial resolution of 300 m. Using this map, we excluded forest plots that had undergone a land cover change since the sampling period and did not have a natural vegetation cover any more at the time of remote sensing image acquisition. To do so, the landcover map was first resampled to a spatial resolution of 1 km to be in accordance with MTCI spatial resolution. Then, the plots located on land area classified as either rainfed cropland, mosaic between croplands and natural vegetation, sparse vegetation or artificial surfaces were excluded from the analysis.

# 2.3.2 Relationship between MTCI and canopy N data at lower spatial resolution

In a first step, the relationships between MTCI and canopy N data values were investigated after resampling both datasets to the same, lower, spatial resolution. The resampled spatial resolutions were 5 km, 10 km, 15 km, and 20 km. This was done because of the initial difference in support size between MTCI spatial resolution and the forest plots size (i.e. 1 km and 6 m, respectively). This enabled us to investigate the relationships between MTCI and canopy N data independently of differences in initial support size.

The monthly averaged MTCI images obtained previously (section 2.3.1) were resampled successively to 5 km, 10 km, 15 km, and 20 km. Beforehand, The Globcover 2009 land cover map was used to exclude from the resampling computation the MTCI pixels located on land surface without natural vegetation cover. As for the forest plots, MTCI pixels whose land cover class corresponded to rainfed cropland, mosaic between croplands and natural vegetation, sparse vegetation or artificial surfaces were excluded from the upscaling analysis. The monthly averaged MTCI images obtained previously were resampled successively to 5 km, 10 km, 15 km, and 20 km. Forest plots data were then averaged per month over the newly obtained pixel. -The relationship between the resampled MTCI values and canopy N data was analysed using linear regression.

# 2.3.3 Relationship between MTCI and canopy N data at initial higher spatial resolution

In a second step, the relationships between MTCI and canopy N data, both canopy  $N_{[\%]}$  and canopy  $N_{[area]}$ , were examined at the original spatial resolution of 1 km. This allowed us to investigate the influence of PFT and species on the relationships as this information was lost in the resampling process. The relationships between MTCI and canopy N at 1 km-spatial resolution were analysed with linear regression for the whole dataset, for each PFT separately as well as for individual species.

# 2.3.4 Statistical analysis

245

250

255

260

265

270

275

280

After applying the selection criteria <u>as explained in the section 2.3.1-above</u>, i.e. plots measured between June 1<sup>st</sup> and October 31<sup>st</sup>, exclusion of plots with infrequent species and selection based on Globcover 2009, 846 forest plots were available for analysis, including 841 plots with foliar biomass and canopy N content information. Descriptive statistics of canopy N<sub>[%]</sub>, foliar biomass and canopy N<sub>[area]</sub> were then produced for each of the tree species and PFT included in the analysis. The linear regressions between MTCI and canopy N were then performed for both resampled and non-resampled datasets. Preliminary analysis showed that using a natural logarithm transformation (log) of the canopy N variables was necessary to fulfil linear regression model assumptions, namely normality and homogeneity of variance of the residuals. The minimum number of data points needed to carry out the linear regression analysis was fixed at 10. We calculated the Relative Root Mean Square Error of cross-validation

(RRMSEcv, %) using the leave-one-out cross validation method (Clevers and Gitelson, 2013). Its calculation is presented in Eq. (3) following (Yao et al., 2010):

$$RRMSEcv = \sqrt{\frac{1}{n} \times \sum_{i=1}^{n} (P_i - O_i)^2} \times \frac{100}{\overline{O}_i}$$
 (3)

where  $P_i$  represents the predicted value,  $O_i$ , the observed value,  $\bar{O}_i$  the mean of all observed value and n the total number of measurement. Resampling both datasets as well as linking the plots to the MTCI pixels The spatial analyses wasere done with the PCRaster software (Karssenberg et al., 2010). The statistical analyses were performed in the R environment (R Development Core Team, 2014) and the ggplot2 package was used for the graphics (Wickham, 2009).

## 3 Results

285

290

295

300

305

310

315

# 3.1 Descriptive statistics

Descriptive statistical analysis of canopy  $N_{[s]}$ , canopy  $N_{[area]}$  and foliar biomass were performed for each tree species included in the dataset (Table 1). The four most abundant species (Pinus halepensis, Pinus sylvestris, Quercus ilex and Pinus uncinata) dominated 667 plots i.e. almost 80% of the plots. The cumulated abundance percentages of ENF, EBF and DBF species were equal to 66 %, 22 % and 9 %, respectively. From this data, it is clear that the forests plots were mainly dominated by ENF species. On average, Pinus uncinata plots had the highest biomass values while Quercus suber plots showed the lowest mean value for this variable. Descriptive statistics were also analysed by PFT. The mean canopy  $N_{[\%]}$  was lowest for ENF species, 0.97 %N, and highest for DBF trees, 2.17 %N (Fig. 2a). Canopy N<sub>[%]</sub> value ranges were equal to 1.91 %N, 2.06 %N, 1.68 %N and 1.42 %N for DBF, EBF, ENF and mixed plots, respectively. The canopy  $N_{\text{[area]}}$  statistics were analysed by PFT as well (Fig. 2b) and the averaged canopy  $N_{\text{Jareal}}$  values ranged from 1.82 g m<sup>-2</sup> to 4.61 g m<sup>-2</sup>. A Pearson correlation matrix (Fig. 3) was computed between the variables for the whole dataset and while the. The correlation between each pair of variables was significant, and the relationship correlation between canopy  $N_{\text{[area]}}$  and foliar biomass was strongest (r = 0.88). This result was expected as the foliar biomass was included in the  $N_{\text{farea}}$  calculation. This matrix also shows distribution histograms of the three variables. As canopy  $N_{[86]}$  and canopy  $N_{[area]}$  distributions are positively skewed, a logarithmic transformation was applied to these variables to fulfil linear model assumptions. Correlation matrices for each DBF, EBF and ENF plots are presented in the Appendix (Fig. A 2-4).

# 3.2 Relationship between MTCI and canopy N data at lower spatial resolution

The relationships between MTCI and both canopy  $N_{[\%]}$  and canopy  $N_{[area]}$  were studied after resampling both datasets to the same, lower, spatial resolution. This was done to investigate the relationship between MTCI and canopy N data independently of differences in support size. The results showed that the relationships between MTCI and either canopy  $N_{[\%]}$  or canopy  $N_{[area]}$  were all highly significant (p<0.000). Moreover, the relationship between MTCI and canopy  $N_{[\%]}$  was always stronger than the relationship for MTCI and canopy  $N_{[area]}$  for each resampling factor. Moreover, the relationships between MTCI and either canopy  $N_{[\%]}$  or canopy  $N_{[area]}$  were all highly significant (p<0.000). The  $r^2$  values of the relationship between MTCI and canopy  $N_{[\%]}$  were equal to 0.33, 0.37, 0.354 and 0.423 for 5 km, 10 km, 15 km and 20 km resampled spatial resolution, respectively. The  $r^2$  values of the relationship between MTCI and canopy  $N_{[area]}$  were equal to 0.2047, 0.20, 0.20 and 0.187 at 5 km, 10 km, 15 km and 20 km spatial resolution. The relationship between MTCI and canopy  $N_{[\%]}$  at 20 km spatial resolution is shown in Figure 5. Table 2 shows the number of plots per pixel for different pixel sizes (km). As expected, the number of

plots per pixel increased with the pixel size, with a mean of 4.1 plots at 20 km spatial resolution. The descriptive statistics of the number of different PFT, species and sampling years per pixel spatial resolution are provided in the Appendix (Table A 2 – A 4).

# 3.3 Relationship between MTCI and canopy N data at higher spatial resolution

# 3.3.1 Relationship between MTCI and canopy N concentration

325

The relationships between MTCI and canopy N data were studied after resampling both datasets to at the same, lower, initial higher spatial resolution (1 km). The results showed that the linear regression between MTCI and canopy  $N_{[\%]}$  for the whole dataset (n = 846) was highly significant (p<0.000) and had an  $r^2$  value of 0.32 and a RRMSEcv value of 18.7 % (

Table 723, Fig. 5a). The relationship between MTCI and Canopy  $N_{[\%]}$  was also investigated for each PFT individually (Fig. 5b-e). For DBF plots, the relationship between MTCI and canopy  $N_{[\%]}$  had an  $r^2$  value of 0.25 (n = 80) and was significant. However, although significant, the  $r^2$  of the relationship between MTCI and canopy  $N_{[\%]}$  for EBF and ENF plots were lower and equal to 0.03 (n = 186) and 0.10 (n = 564), respectively.

The relationship between MTCI and canopy  $N_{[\%]}$  was also significant for one individual species, *Fagus sylvatica*. The proportion of explained variance for this species was equal to 0.71 (n = 15). This result, although obtained on a restricted number of plots, showed that the significant relationships between MTCI and canopy  $N_{[\%]}$  not only existed when all DBF plots were included but also held for one individual DBF species.

# 3.3.2 Relationship between MTCI and canopy N content

Significant relationships between MTCI and canopy  $N_{[area]}$  were found for the whole dataset as for EBF and ENF plots (Table 3). The scatterplots between MTCI and canopy  $N_{[area]}$  are presented in Figure 6. The proportion of explained variance was higher for ENF plots compared to the other PFTs and compared to the overall relationship across all plots. The relationship between MTCI and canopy  $N_{[area]}$  was also investigated for 10 individual species and one of them showed significant relationships: *Ouercus ilex* ( $r^2 = 0.10$ , r = 160).

### 4 Discussion

330

335

340

345

Our aim was to explore the relationship between the MTCI vegetation index and both canopy  $N_{[\%]}$  and canopy  $N_{[area]}$  in Mediterranean forests at the regional scale in Catalonia, north eastern of Spain. This was done by using the ESA spaceborne MTCI remote sensing product and canopy N data from a forest inventory. The relationship was first investigated using MTCI and canopy N data resampled to the same, lower, spatial resolution. The relationship was then investigated across all plots and by PFT at MTCI initial spatial resolution of 1 km.

### 4.1 Relationship between MTCI and canopy N data at lower spatial resolution

This pre-analysis was undertaken to study the MTCI-canopy N relationships independently of the discrepancy between MTCI original spatial resolution (1 km) and the size of the forest plots (diameter of 6 m). By resampling both datasets to a lower spatial resolution, i.e. 5 km, 10 km, 15 km and 20 km, the obtained values were less impacted by small-scale variations because they were obtained by averaging several values over a larger area. The results showed that the relationship between MTCI and canopy N data was significant and consistent across all spatial resolutions investigated: 5 km, 10 km, 15 km and 20 km. This showeds that, when the influence of the spatial discrepancy between the original datasets, i.e. 6 m and 1 km, was taken into account, MTCI and canopy N data were linked and that the MTCI-canopy N relationship was not strongly affected by the resampled spatial resolution.

#### 4.2 Relationship between MTCI and canopy N data at higher spatial resolution

### 4.2.1 Canopy N concentration detection

The overall relationship between MTCI and canopy  $N_{[\%]}$  at 1 km spatial resolution for all the forest plots (n = 846) was significant and the  $r^2$  value was equal to 0.32 (Table 3, Fig. 5). This result showed that canopy  $N_{[\%]}$  could be related to MTCI in Mediterranean forests. The performance of the MTCI vegetation index to detect canopy  $N_{[\%]}$  in Mediterranean vegetation was similar to the results obtained from previous studies using spaceborne MTCI at higher spatial resolution. For example, using MTCI computed from the spaceborne RapidEye sensor at 5 m spatial resolution, it was possible to detect canopy  $N_{[\%]}$  in grassland savannah and sub-tropical forest with similar coefficients of determination,  $r^2 = 0.35$  and  $r^2 = 0.52$ , respectively (Ramoelo et al., 2012;Cho et al., 2013). More generallyHowever, while there is a consensus regarding MTCI ability for in situ leaf or canopy  $N_{[\%]}$  detection in a variety of crops using handheld spectrometers (Tian et al., 2011;Li et al., 2014), there is no general agreement about MTCI ability for canopy  $N_{[\%]}$  detection across vegetation and sensor types at larger scales. For example, MTCI computed from airborne data at 3 m spatial resolution could not be related to canopy  $N_{[\%]}$  from a mixed temperate forest (Wang et al., 2016). In this context our finding brings new insight into MTCI  $N_{[\%]}$  sensing capabilities at a much coarser spatial resolution (1 km) compared to what has been done before.

Investigating the influence of the PFTS on the overall relationship highlighted the difference between DBF, EBF and ENF types of vegetation regarding canopy  $N_{[\%]}$  detection by spaceborne MTCI. The relationships between MTCI and canopy  $N_{[\%]}$  were significant for all the PFT taken separately (p-value<0.05). However, a higher proportion of variance was explained for DBF and mixed plots ( $r^2 = 0.25$  and  $r^2 = 0.47$  for DBF and mixed plots, respectively) compared to the other plant functional types ( $r^2 = 0.10$  and  $r^2 = 0.03$  for ENF and EBF trees, respectively) and the relationship between MTCI and canopy  $N_{[\%]}$  was especially weaker for EBF plots. Therefore, the relationship observed for all the forest plots may be mainly driven by DBF and mixed plots. This result is different from what was observed by Ollinger et al. (2008) in boreal forests, where canopy  $N_{[\%]}$  was related to NIR reflectance for both broadleaf and needleleaf plots taken separately. Moreover, the results obtained for ENF tree species are surprising as previous studies investigating the relationship between foliar  $N_{[\%]}$  and in situ measured spectra reported higher  $r^2$  values,  $r^2 = 0.59$  and  $r^2 = 0.81$  in spruce and pine forest, respectively (Stein et al., 2014;Schlerf et al., 2010). The differences in scale and methodology might explain the divergent results compared to previous findings. Indeed, in our study, the analysis is carried out at a much coarser spatial resolution using spaceborne data compared to the fine spatial scale obtained with in situ devices. Moreover, most of these studies were carried out in temperate forests and studies investigating canopy  $N_{[\%]}$  detection in Mediterranean regions are scarce. When investigating the relationship between canopy  $N_{[\%]}$  and MTCI at the species level, we also found that it was significant for Fagus sylvatica plots ( $r^2 = 0.71$ ).

In the literature, the relationship between MTCI and canopy  $N_{[\%]}$  is often not stratified by PFT or species (Sullivan et al., 2013; Wang et al., 2016). In this study, we showed that investigating this relationship for each PFT taken separately yielded additional insight. Indeed, to our knowledge the difference in explained variance between DBF and other PFTs in MTCI and canopy  $N_{[\%]}$  relationship has not been observed before. Moreover, the results observed for *Fagus sylvatica* plots (n = 15) were

consistent with the stronger relationship observed for DBF plots. Moreover, we could show that the stronger relationship observed for DBF plots was consistent for Fagus sylvatica plots taken separately.

## 4.2.2 Canopy N content detection

395

400

405

410

415

420

The relationship between MTCI and canopy  $N_{[area]}$ , which was obtained by combining canopy N concentration values with biomass data, was significant across all plots (n = 841) (Table 43, Fig. 6). Although the  $r^2$  value was lower for the relationship between MTCI and canopy  $N_{[area]}$  ( $r^2 = 0.17$ ) than for the relationship between MTCI and canopy  $N_{[m]}$  ( $r^2 = 0.32$ ), it is interesting to note that canopy  $N_{[area]}$  can be related to spaceborne MTCI as remotely sensed detection of canopy  $N_{[area]}$  is rarely investigated in forest environments (Mirik et al., 2005). In comparison, previous studies conducted in grasslands reported higher prediction accuracy e.g. by using spaceborne MTCI at 300 m spatial resolution or a simple ratio-type vegetation index computed from airborne imagery at 1 m spatial resolution, canopy  $N_{[area]}$  was detected with  $r^2$  values equal to 0.29 and 0.66, respectively (Mirik et al., 2005; Ullah et al., 2012).

The relationship between MTCI and canopy  $N_{[area]}$  was only significant for ENF and EBF plots (Fig. 6b-e), with a higher proportion of explained variance for ENF plots ( $r^2 = 0.20$ ). However, when this relationship was investigated at the species scale, significant results were found for *Quercus ilex* (EBF) plots. This is accordance with a previous study examining the detection of canopy  $N_{[area]}$  in *Quercus ilex* trees by MTCI computed from in situ spectra ( $r^2 = 0.43$ ) (Pacheco-Labrador et al., 2014).

## 4.3 Comparing results obtained for canopy N concentration and canopy N content detection

This analysis highlighted the difference between canopy N expressed as a percentage of leaf dry matter (canopy  $N_{[\%]}$ ) and on an area basis (canopy  $N_{[area]}$ ) regarding detection by spaceborne MTCI for the different PFTs. Canopy  $N_{[\%]}$  of DBF and mixed plots showed higher correlation with MTCI compared to EBF and ENF plots while the relationship between canopy  $N_{[area]}$  of ENF plots with MTCI was stronger than for any other PFTs. These differences between canopy  $N_{[\%]}$  and canopy  $N_{[area]}$  detection by remote sensing can be related to previous findings showing that canopy  $N_{[area]}$  but not canopy  $N_{[\%]}$  could be detected by MTCI in grassland (Ullah et al., 2012) and by a simple ratio index in heterogeneous rangelands (Mirik et al., 2005) at various spatial scales, 300 m and 1 m, respectively. In the literature, canopy  $N_{[\%]}$  is more often used to detect N state of foliage in forest while canopy  $N_{[area]}$  is regularly employed in grasslands but also in crops (Clevers and Gitelson, 2013;Schlemmer et al., 2013). Our results showed that, for ENF plots, when biomass was accounted for, as in canopy  $N_{[area]}$ , the relationship between MTCI and canopy  $N_{[area]}$  was stronger compared to canopy  $N_{[\%]}$ .

## 4.4 Influence of the plant functional type on Possible confounding factors of the MTCI canopy N relationship

The relationships between MTCI and both canopy  $N_{[\%]}$  and canopy  $N_{[area]}$  were influenced by the PFT of the plots. The relationship between MTCI and canopy  $N_{[\%]}$  was stronger for DBF and mixed plots compared to EBF and ENF plots while the opposite was true for the MTCI-canopy  $N_{[area]}$  relationship. In the ongoing discussion about the mechanisms underlying the

remote detection of canopy N, some authors argued that the differences in structural properties in canopy structure between different PFTs was were causing a confounding factor of the observed relationship between canopy N and remote sensing data, rendering it spurious (Knyazikhin et al., 2013). Other authors, although agreeing that canopy structural properties needed to be accounted for, suggested that the role of canopy structure as confounding factor can be explained by direct biochemical an indirect association between canopy N and canopy structure resulting from convergent adaptive processes link between canopy N and reflectance data was not necessary to detect canopy N with reflectance data (Ollinger et al., 2013; Townsend et al., 2013). In this context, our analysis showed that the PFTs of the plots had an influence on the MTCI canopy N relationship in a specific type of ecosystem, namely Mediterranean forests. Further analysis is needed to disentangle the influence of structural properties on canopy N detection. Other confounding factors associated with N availability that might affect the observed relationship possibly include biomass, biomass allocation, leaf area index (LAI), water availability, soil type, etc. The data from the forest inventory used in this analysis, i.e. the Catalonian National Forest Inventory, were extensively studied, showing that water availability was the most limiting factor in this region. Water availability was positively correlated with both the  $N_{\text{fareal}}$  and  $N_{\text{fish}}$ in leaves, as well as with foliar and total above-ground biomass through MAP (Sardans et al., 2011:Sardans and Peñuelas, 2013). The MAP also influenced the PFT distribution as DBF plots were located in wetter areas than EBF plots, which were found in wetter sites than ENF plots. Regarding the influence of PFT on the foliar biomass, DBF plots had on average 45% less foliar biomass than EBF or ENF plots (Sardans and Peñuelas, 2013). This shows that canopy N<sub>[%]</sub> and canopy N<sub>[area]</sub> were interrelated to biomass, PFT and MAP.

## 4.5 Perspectives for future for larger scale applications

425

430

435

440

445

450

The methodology applied in this paper is different from the usual methodology implemented to detect canopy N concentration in forests. Remote sensing of N in forest canopies by hyperspectral sensors is often coupled with intensive forest sampling measurements. This method has been effective at detecting canopy N concentration locally in a vast range of environments (Serrano et al., 2002;Smith et al., 2002;Townsend et al., 2003;Ollinger et al., 2008;Wang et al., 2016). Applying this technique at larger scales has already been explored. For example, Martin *et al.*, (2008) compiled 137 field plots data from previous studies in various forest types and investigated the possibility to find a common detection algorithm. However, due to the different treatments required as well as the <u>limited swath width associated with the</u> high spatial resolution (from 3 m to 30 m for Hyspex airborne and Hyperion spaceborne sensors, respectively, Wang et al., 2016;Smith et al., 2003), applying imaging spectrometryseopy at a broader scale might reveal laborious. Depending on the sensors as well as on the extent of the study area, this might involve correcting the acquired images for atmospheric influences and cloud cover as well as combining several images into a larger scale image. A recent study in northern temperate forests explored the effect of spatial resolution on canopy N<sub>[%]</sub> estimation. The results showed that, although the prediction accuracy was reduced compared to what was achieved using PLS regression at higher spatial resolution, it was still possible to estimate canopy N<sub>[%]</sub> with r<sup>2</sup> between 0.34 and 0.81 using various vegetation indices computed from MODIS reflectance data at 500 m spatial resolution (Lepine et al., 2016). In this context, the methodology applied in this article could be a valuable alternative to explore canopy N detection at larger scale.

Using published data from an extensive field plot inventory, we were able to relate both canopy  $N_{[m]}$  and canopy  $N_{[area]}$  to MTCI at different spatial resolutions. Although the relationships found were modest, our study contributes to the ongoing discussion about how to remotely sense canopy N over larger area. As MTCI time series are readily and almost globally available, it could eventually be possible to assess<del>apply</del> our approach at a broader scale in different types of biomes. The results obtained for DBF species and Fagus sylvatica in particular suggest that this method may be efficient at estimating canopy N in temperate forests. If the strength of the relationship between MTCI and canopy N can further be improved, this could lead to canopy N monitoring possibilities at regional scale. In this context, the new sensors OLCI, onboard Sentinel 3 satellite, and especially MSI, onboard Sentinel 2 satellite might be promising due to their higher spatial resolution, from 10 to 60 m for Sentinel 2. They have bands well positioned to compute the MTCI vegetation index. Although the OLCI Terrestrial Chlorophyll Index (OTCI), the successor of the MTCI for the OLCI sensor, is already included in the OLCI level 2b reflectance image, no level 3 product (mosaicked over larger areas and temporally averaged hence similar to the MTCI time series used in this analysis) is available yet. In addition to more detailed remote sensing data, adding additional ground based canopy N observations to the regression models could better constrain these models as well. It would in particular be promising to use canopy N data over larger scale areas and for more diverse and globally distributed vegetation types. Obtaining reliable canopy N estimates over larger scale and for diverse and globally distributed type of vegetation would be necessary to be able to use such estimates to improve and calibrate global vegetation models.

#### 5 Conclusion

455

460

465

470

475

480

485

In this study, we investigated the relationship between spaceborne MTCI from ENVISAT and both canopy  $N_{[\$\epsilon]}$  and canopy  $N_{[area]}$  at regional scale in Mediterranean forests. We found significant results across all plots both when the original data were resampled to 5 km, 10 km, 15 km and 20 km and for the original spatial resolution of 1 km. The relationship between MTCI and canopy N data was also significant for some individual PFTs and species. The  $r^2$  values were 0.32 and 0.17 for the overall relationships between MTCI and either canopy  $N_{[\$\epsilon]}$  or canopy  $N_{[area]}$ , respectively. We highlighted the differences between PFTs and both canopy  $N_{[\$\epsilon]}$  and canopy  $N_{[area]}$ : the relationship between MTCI and canopy  $N_{[\$\epsilon]}$  was stronger for DBF and mixed plots while canopy  $N_{[area]}$  was more linked to MTCI for ENF plots. Such differences in relationships between MTCI and either canopy  $N_{[\$\epsilon]}$  or canopy  $N_{[area]}$  were already observed in grasslands ecosystem. Our results showed that MTCI could be related to canopy N for some individual PFTs, indicating an influence of the PFTs on the MTCI-canopy N relationship. The methodology developed in this study could be investigated at larger scales in different types of ecosystem. While this could already be undertaken using the ENVISAT MTCI 10 years time series as it is almost globally available, ESA new Sentinel-2 satellite launched on 23 June 2015 yields reflectance data at improved spatial and temporal resolution than ENVISAT-MERIS. Canopy N estimates collected through larger scales applications could be exploited in vegetation modelling studies including both the C and N cycles.

#### 6 Data availability

The canopy data used in this study can be obtained from the TRY Plant Trait Database (https://www.try-db.org/TryWeb/Home.php, dataset 91) or by directly contacting the authors.

#### 7 Appendix A

This appendix presents the inter-annual variation of canopy N<sub>[%]</sub> (Fig. A 1), the Allometric relationship between the branch diameter (mm) and the foliar dry mass (g) from which the biomass data was obtained (Fig. A1) and the correlation matrices for DBF (Fig. A2), EBF (Fig. A3) and ENF plots (Fig. A3) as well as the tables representing the allometric relationships between foliar biomass and diameter at breast height (DBH, Table A 1), the number of PFT (Table A 2), the number of species (Tables A 3) and the number of sampling years (Table A 4) per resampled pixel, by pixel spatial resolution-

#### 8 Competing interest

495

500

505

The authors declare that they have no conflict of interest.

#### 9 Acknowledgements

This research was funded by The Netherlands Organisation for Scientific Research (NWO) under the project number NWO ALW-GO-AO/14-12. J. Sardans and J. Peñuelas were funded by the European Research Council Synergy grant SyG-2013-610028 IMBALANCE-P, the Spanish Government projects CGL2013-48074-P and the Catalan Government project SGR 2014-274. We would like to acknowledge Scott Ollinger and Lucie Lepine for their valuable comments and discussions on our research project as well as Ton Markus for his help with the figures presented in this paper.

#### 10 References

510

520

- Boyd, D. S., Almond, S., Dash, J., Curran, P. J., and Hill, R. A.: Phenology of vegetation in southern england from envisat meris terrestrial chlorophyll index (MTCI) data, International Journal of Remote Sensing, 32, 8421-8447, doi:10.1080/01431161.2010.542194. 2011.
- Boyd, D. S., Almond, S., Dash, J., Curran, P. J., Hill, R. A., and Foody, G. M.: Evaluation of envisat MERIS terrestrial chlorophyll index-based models for the estimation of terrestrial gross primary productivity, IEEE Geoscience and Remote Sensing Letters, 9, 457-461, 2012.
- Cho, M. A., Ramoelo, A., Debba, P., Mutanga, O., Mathieu, R., van Deventer, H., and Ndlovu, N.: Assessing the effects of subtropical forest fragmentation on leaf nitrogen distribution using remote sensing data, Landscape Ecology, 28, 1479-1491, doi:10.1007/s10980-013-9908-7, 2013.
  - Clevers, J. G. P. W., and Gitelson, A. A.: Remote estimation of crop and grass chlorophyll and nitrogen content using rededge 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, 2013.
  - Crabbe, R. A., Dash, J., Rodriguez-Galiano, V. F., Janous, D., Pavelka, M., and Marek, M. V.: Extreme warm temperatures alter forest phenology and productivity in Europe, Science of The Total Environment, 563–564, 486-495, http://dx.doi.org/10.1016/j.scitotenv.2016.04.124, 2016.
    - Curran, P. J., and Dash, J.: Algorithm Theoretical basis document ATBD 2.22 Chlorophyll Index, University of Southampton, Southampton, 41 pp., available at: <a href="https://earth.esa.int/documents/700255/2042855/MERIS\_ATBD\_2042855/MERIS\_ATBD\_2042852.2042855/MERIS\_ATBD\_2042852.2042852.2042851.2042852++2042005.pdf">https://earth.esa.int/documents/700255/2042855/MERIS\_ATBD\_2042852.2042822\_v2042851.2042852++2042005.pdf</a>, last access 11 November 16, 2005.
    - Dash, J., and Curran, P. J.: The MERIS terrestrial chlorophyll index, International Journal of Remote Sensing, 25, 5403-5413, doi:10.1080/0143116042000274015, 2004.
    - Dash, J., and Curran, P. J.: Evaluation of the MERIS terrestrial chlorophyll index (MTCI), Advances in Space Research, 39, 100-104, doi:10.1016/j.asr.2006.02.034, 2007.
    - Drusch, M., Del Bello, U., Carlier, S., Colin, O., Fernandez, V., Gascon, F., Hoersch, B., Isola, C., Laberinti, P., Martimort, P., Meygret, A., Spoto, F., Sy, O., Marchese, F., and Bargellini, P.: Sentinel-2: ESA's Optical High-Resolution Mission for GMES Operational Services, Remote Sensing of Environment, 120, 25-36, doi:10.1016/j.rse.2011.11.026, 2012.
- Evans, J. R.: Photosynthesis and nitrogen relationships in leaves of C3 plants, Oecologia, 78, 9-19, doi:10.1007/BF00377192, 1989.
  - Ferwerda, J. G., Skidmore, A. K., and Mutanga, O.: Nitrogen detection with hyperspectral normalized ratio indices across multiple plant species, International Journal of Remote Sensing, 26, 4083-4095, 10.1080/01431160500181044, 2005.
  - Filella, I., and Penuelas, J.: The red edge position and shape as indicators of plant chlorophyll content, biomass and hydric status, International Journal of Remote Sensing, 15, 1459-1470, http://dx.doi.org/10.1080/01431169408954177, 1994.

- Filella, I., Serrano, L., Serra, J., and Penuelas, J.: Evaluating wheat nitrogen status with canopy reflectance indices and discriminant analysis, Crop Science, 35, 1400-1405, doi:10.2135/cropsci1995.0011183X003500050023x, 1995.
  - Foody, G. M., and Dash, J.: Discriminating and mapping the C3 and C4 composition of grasslands in the northern Great Plains, USA, Ecological Informatics, 2, 89-93, doi:10.1016/j.ecoinf.2007.03.009, 2007.
- Glenn, E. P., Huete, A. R., Nagler, P. L., and Nelson, S. G.: Relationship between remotely-sensed vegetation indices, canopy attributes and plant physiological processes: What vegetation indices can and cannot tell us about the landscape, Sensors, 8, 2136-2160, doi:10.3390/s8042136, 2008.
  - González, J. R., and Pukkala, T.: Characterization of forest fires in Catalonia (north-east Spain), European Journal of Forest Research, 126, 421-429, doi:10.1007/s10342-006-0164-0, 2007.
  - Gracia, C., Ibàñez, J. J., Burriel, J. A., Mata, T., and Vayreda, J.: Inventari Ecològic i Forestal de Catalunya. Mètodes, CREAF, Bellaterra, available at: http://www.creaf.uab.es/iefc/pub/Metodes/index.htm, last access 11 November 2016, 2004.

555

560

565

- Green, D. S., Erickson, J. E., and Kruger, E. L.: Foliar morphology and canopy nitrogen as predictors of light-use efficiency in terrestrial vegetation, Agricultural and Forest Meteorology, 115, 163-171, http://dx.doi.org/10.1016/S0168-1923(02)00210-1, 2003.
- Hansen, P. M., and Schjoerring, J. K.: Reflectance measurement of canopy biomass and nitrogen status in wheat crops using normalized difference vegetation indices and partial least squares regression, Remote Sensing of Environment, 86, 542-553, doi:10.1016/S0034-4257(03)00131-7, 2003.
- Harris, A., and Dash, J.: The potential of the MERIS Terrestrial Chlorophyll Index for carbon flux estimation, Remote Sensing of Environment, 114, 1856-1862, doi:10.1016/j.rse.2010.03.010, 2010.
- Harris, A., and Dash, J.: A new approach for estimating northern peatland gross primary productivity using a satellite-sensor-derived chlorophyll index, Journal of Geophysical Research: Biogeosciences, 116, doi:10.1029/2011JG001662, 2011.
- Huber, S., Kneubühler, M., Psomas, A., Itten, K., and Zimmermann, N. E.: Estimating foliar biochemistry from hyperspectral data in mixed forest canopy, Forest Ecology and Management, 256, 491-501, doi:10.1016/j.foreco.2008.05.011, 2008.
- Kergoat, L., Lafont, S., Arneth, A., Le Dantec, V., and Saugier, B.: Nitrogen controls plant canopy light-use efficiency in temperate and boreal ecosystems, Journal of Geophysical Research: Biogeosciences, 113, doi:10.1029/2007JG000676, 2008.
- Knyazikhin, Y., Schull, M. A., Stenberg, P., Mõttus, M., Rautiainen, M., Yang, Y., Marshak, A., Latorre Carmona, P., Kaufmann, R. K., Lewis, P., Disney, M. I., Vanderbilt, V., Davis, A. B., Baret, F., Jacquemoud, S., Lyapustin, A., and Myneni, R. B.: Hyperspectral remote sensing of foliar nitrogen content, Proceedings of the National Academy of Sciences, 110, E185–E192, 10.1073/pnas.1210196109, 2013.
- Kumar, L., Schmidt, K., Dury, S., and Skidmore, A.: Imaging Spectrometry and Vegetation Science, in: Imaging Spectrometry: Basic Principles and Prospective Applications, edited by: Meer, F. D. v. d., and Jong, S. M. D., Springer Netherlands, Dordrecht, 111-155, 2006.

- Lana, X., and Burgueño, A.: Spatial and temporal characterization of annual extreme droughts in Catalonia (Northeast Spain), International Journal of Climatology, 18, 93-110, 1998.
- Lepine, L. C., Ollinger, S. V., Ouimette, A. P., and Martin, M. E.: Examining spectral reflectance features related to foliar nitrogen in forests: Implications for broad-scale nitrogen mapping, Remote Sensing of Environment, 173, 174-186, doi:10.1016/j.rse.2015.11.028, 2016.

585

590

595

600

- Li, F., Miao, Y., Feng, G., Yuan, F., Yue, S., Gao, X., Liu, Y., Liu, B., Ustin, S. L., and Chen, X.: Improving estimation of summer maize nitrogen status with red edge-based spectral vegetation indices, Field Crops Research, 157, 111-123, doi:10.1016/j.fcr.2013.12.018, 2014.
- Liu, D., Ogaya, R., Barbeta, A., Yang, X., and Peñuelas, J.: Contrasting impacts of continuous moderate drought and episodic severe droughts on the aboveground-biomass increment and litterfall of three coexisting Mediterranean woody species, Global Change Biology, 21, 4196-4209, doi:10.1111/gcb.13029, 2015.
- Martin, M. E., Plourde, L. C., Ollinger, S. V., Smith, M. L., and McNeil, B. E.: A generalizable method for remote sensing of canopy nitrogen across a wide range of forest ecosystems, Remote Sensing of Environment, 112, 3511-3519, doi:10.1016/j.rse.2008.04.008, 2008.
- Mirik, M., Norland, J. E., Crabtree, R. L., and Biondini, M. E.: Hyperspectral one-meter-resolution remote sensing in Yellowstone National Park, Wyoming: I. Forage nutritional values, Rangeland Ecology and Management, 58, 452-458, doi:10.2111/04-17.1, 2005.
  - NEODC NERC Earth Observation Data Centre: http://neodc.nerc.ac.uk/, access: 06/02/2015, 2015.
- Ollinger, S. V., and Smith, M. L.: Net primary production and canopy nitrogen in a temperate forest landscape: An analysis using imaging spectroscopy, modeling and field data, Ecosystems, 8, 760-778, doi:10.1007/s10021-005-0079-5, 2005.
- Ollinger, S. V., Richardson, A. D., Martin, M. E., Hollinger, D. Y., Frolking, S. E., Reich, P. B., Plourde, L. C., Katul, G. G., Munger, J. W., Oren, R., Smith, M. L., Paw U, K. T., Bolsta, P. V., Cook, B. D., Day, M. C., Martin, T. A., Monson, R. K., and Schmid, H. P.: Canopy nitrogen, carbon assimilation, and albedo in temperate and boreal forests: Functional relations and potential climate feedbacks, Proceedings of the National Academy of Sciences of the United States of America, 105, 19336-19341, doi:10.1073/pnas.0810021105., 2008.
- Ollinger, S. V.: Sources of variability in canopy reflectance and the convergent properties of plants, New Phytologist, 189, 375-394, doi:10.1111/j.1469-8137.2010.03536.x, 2011.
- Ollinger, S. V., Reich, P. B., Frolking, S., Lepine, L. C., Hollinger, D. Y., and Richardson, A. D.: Nitrogen cycling, forest canopy reflectance, and emergent properties of ecosystems, Proceedings of the National Academy of Sciences, 110, E2437, 10.1073/pnas.1304176110, 2013.
  - Pacheco-Labrador, J., González-Cascón, R., Pilar Martín, M., and Riaño, D.: Understanding the optical responses of leaf nitrogen in mediterranean holm oak (Quercus ilex) using field spectroscopy, International Journal of Applied Earth Observation and Geoinformation, 26, 105-118, doi:10.1016/j.jag.2013.05.013, 2014.

- Peng, Y., and Gitelson, A. A.: Application of chlorophyll-related vegetation indices for remote estimation of maize productivity, Agricultural and Forest Meteorology, 151, 1267-1276, doi:10.1016/j.agrformet.2011.05.005, 2011.
- Peñuelas, J., Gamon, J. A., Fredeen, A. L., Merino, J., and Field, C. B.: Reflectance indices associated with physiological changes in nitrogen- and water-limited sunflower leaves, Remote Sensing of Environment, 48, 135-146, doi:10.1016/0034-4257(94)90136-8, 1994.

620

625

- Ramoelo, A., Skidmore, A. K., Cho, M. A., Schlerf, M., Mathieu, R., and Heitkönig, I. M. A.: Regional estimation of savanna grass nitrogen using the red-edge band of the spaceborne rapideye sensor, International Journal of Applied Earth Observation and Geoinformation, 19, 151-162, doi:10.1016/j.jag.2012.05.009, 2012.
- Reich, P. B., Walters, M. B., Kloeppel, B. D., and Ellsworth, D. S.: Different photosynthesis-nitrogen relations in deciduous hardwood and evergreen coniferous tree species, Oecologia, 104, 24-30, doi:10.1007/BF00365558, 1995.
  - Reich, P. B., Walters, M. B., and Ellsworth, D. S.: From tropics to tundra: Global convergence in plant functioning, Proceedings of the National Academy of Sciences of the United States of America, 94, 13730-13734, doi:10.1073/pnas.94.25.13730, 1997.
  - Reich, P. B., Ellsworth, D. S., Walters, M. B., Vose, J. M., Gresham, C., Volin, J. C., and Bowman, W. D.: Generality of leaf trait relationships: A test across six biomes, Ecology, 80, 1955-1969, doi:10.2307/176671 1999.
  - Reich, P. B.: Key canopy traits drive forest productivity, Proceedings of the Royal Society B: Biological Sciences, 279, 2128-2134, doi:10.1098/rspb.2011.2270, 2012.
  - Rodriguez-Galiano, V. F., Dash, J., and Atkinson, P. M.: Characterising the land surface phenology of Europe using decadal MERIS data, Remote Sensing, 7, 9390-9409, doi:10.3390/rs70709390, 2015.
  - Sardans, J., Rivas-Ubach, A., and Peñuelas, J.: Factors affecting nutrient concentration and stoichiometry of forest trees in Catalonia (NE Spain), Forest Ecology and Management, 262, 2024-2034, doi:10.1016/j.foreco.2011.08.019, 2011.
    - Sardans, J., and Peñuelas, J.: Tree growth changes with climate and forest type are associated with relative allocation of nutrients, especially phosphorus, to leaves and wood, Global Ecology and Biogeography, 22, 494-507, doi:10.1111/geb.12015, 2013.
  - Sardans, J., and Peñuelas, J.: Trees increase their P: N ratio with size, Global Ecology and Biogeography, 24, 147-156, doi:10.1111/geb.12231, 2015.
    - Schlemmer, M., Gitelson, A., Schepers, J., Ferguson, R., Peng, Y., Shanahan, J., and Rundquist, D.: 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, 2013.
- Schlerf, M., Atzberger, C., Hill, J., Buddenbaum, H., Werner, W., and Schüler, G.: Retrieval of chlorophyll and nitrogen in Norway spruce (Picea abies L. Karst.) using imaging spectroscopy, International Journal of Applied Earth Observation and Geoinformation, 12, 17-26, doi:10.1016/j.jag.2009.08.006, 2010.

Serrano, L., Peñuelas, J., and Ustin, S. L.: Remote sensing of nitrogen and lignin in Mediterranean vegetation from AVIRIS data: Decomposing biochemical from structural signals, Remote Sensing of Environment, 81, 355-364, doi:10.1016/S0034-4257(02)00011-1, 2002.

640

645

650

660

- Smith, B., Wårlind, D., Arneth, A., Hickler, T., Leadley, P., Siltberg, J., and Zaehle, S.: Implications of incorporating N cycling and N limitations on primary production in an individual-based dynamic vegetation model, Biogeosciences, 11, 2027-2054, doi:10.5194/bg-11-2027-2014, 2014.
- Smith, M. L., and Martin, M. E.: A plot-based method for rapid estimation of forest canopy chemistry, Canadian Journal of Forest Research, 31, 549-555, doi:10.1139/x00-187, 2001.
- Smith, M. L., Ollinger, S. V., Martin, M. E., Aber, J. D., Hallett, R. A., and Goodale, C. L.: Direct estimation of aboveground forest productivity through hyperspectral remote sensing of canopy nitrogen, Ecological Applications, 12, 1286-1302, doi:10.2307/3099972 2002.
- Smith, M. L., Martin, M. E., Plourde, L., and Ollinger, S. V.: Analysis of hyperspectral data for estimation of temperate forest canopy nitrogen concentration: Comparison between an airborne (AVIRIS) and a spaceborne (Hyperion) sensor, IEEE Transactions on Geoscience and Remote Sensing, 41, 1332-1337, doi:10.1109/TGRS.2003.813128, 2003.
- Stein, B. R., Thomas, V. A., Lorentz, L. J., and Strahm, B. D.: Predicting macronutrient concentrations from loblolly pine leaf reflectance across local and regional scales, GIScience and Remote Sensing, 51, 269-287, doi:10.1080/15481603.2014.912875, 2014.
- Sullivan, F. B., Ollinger, S. V., Martin, M. E., Ducey, M. J., Lepine, L. C., and Wicklein, H. F.: Foliar nitrogen in relation to plant traits and reflectance properties of New Hampshire forests, Canadian Journal of Forest Research, 43, 18-27, doi:10.1139/cifr-2012-0324, 2013.
  - Tian, Y. C., Yao, X., Yang, J., Cao, W. X., Hannaway, D. B., and Zhu, Y.: Assessing newly developed and published vegetation indices for estimating rice leaf nitrogen concentration with ground- and space-based hyperspectral reflectance, Field Crops Research, 120, 299-310, doi:10.1016/j.fcr.2010.11.002, 2011.
  - Townsend, P. A., Foster, J. R., Chastain Jr, R. A., and Currie, W. S.: Application of imaging spectroscopy to mapping canopy nitrogen in the forest of the central Appalachian mountains using hyperion and AVIRIS, IEEE Transactions on Geoscience and Remote Sensing, 41, 1347-1354, doi:10.1109/TGRS.2003.813205, 2003.
  - Townsend, P. A., Serbin, S. P., Kruger, E. L., and Gamon, J. A.: Disentangling the contribution of biological and physical properties of leaves and canopies in imaging spectroscopy data, Proceedings of the National Academy of Sciences of the United States of America, 110, 10.1073/pnas.1300952110, 2013.
  - Ullah, S., Si, Y., Schlerf, M., Skidmore, A. K., Shafique, M., and Iqbal, I. A.: Estimation of grassland biomass and nitrogen using MERIS data, International Journal of Applied Earth Observation and Geoinformation, 19, 196-204, doi:10.1016/j.jag.2012.05.008, 2012.

Vilà-Cabrera, A., Coll, L., Martínez-Vilalta, J., and Retana, J.: Forest management for adaptation to climate change in the Mediterranean basin: A synthesis of evidence, Forest Ecology and Management, 407, 16-22, doi:10.1016/j.foreco.2017.10.021, 2018.

675

680

685

Wang, Z., Wang, T., Darvishzadeh, R., Skidmore, A. K., Jones, S., Suarez, L., Woodgate, W., Heiden, U., Heurich, M., and Hearne, J.: Vegetation indices for mapping canopy foliar nitrogen in a mixed temperate forest, Remote Sensing, 8, doi:10.3390/rs8060491, 2016.

Wright, I. J., Reich, P. B., Westoby, M., Ackerly, D. D., Baruch, Z., Bongers, F., Cavender-Bares, J., Chapin, T., Cornelissen, J. H. C., Diemer, M., Flexas, J., Garnier, E., Groom, P. K., Gulias, J., Hikosaka, K., Lamont, B. B., Lee, T., Lee, W., Lusk, C., Midgley, J. J., Navas, M.-L., Niinemets, U., Oleksyn, J., Osada, N., Poorter, H., Poot, P., Prior, L., Pyankov, V. I., Roumet, C., Thomas, S. C., Tjoelker, M. G., Veneklaas, E. J., and Villar, R.: The worldwide leaf economics spectrum, Nature, 428, 821-827, doi:10.1038/nature02403, 2004.

Yao, X., Zhu, Y., Tian, Y., Feng, W., and Cao, W.: Exploring hyperspectral bands and estimation indices for leaf nitrogen accumulation in wheat, International Journal of Applied Earth Observation and Geoinformation, 12, 89-100, http://dx.doi.org/10.1016/j.jag.2009.11.008, 2010.

Zaehle, S., and Friend, A.: Carbon and nitrogen cycle dynamics in the O - CN land surface model: 1. Model description, site - scale evaluation, and sensitivity to parameter estimates, Global Biogeochemical Cycles, 24, doi:10.1029/2009GB003521, 2010.

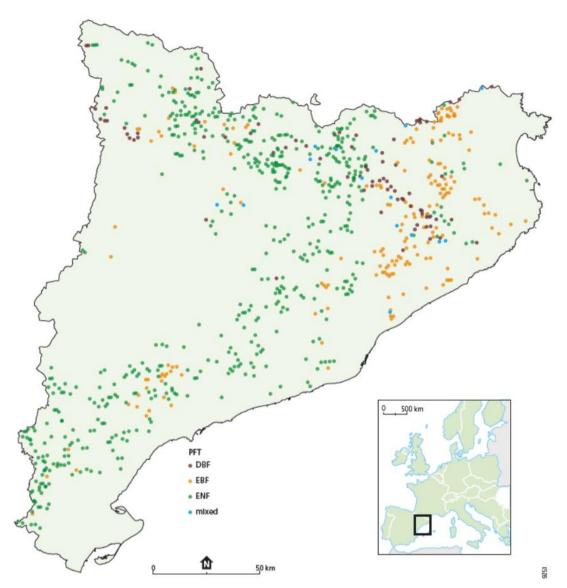


Figure 2. Map showing the forest plots (n = 846) location in the region of Catalonia, north eastern Spain. DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest.

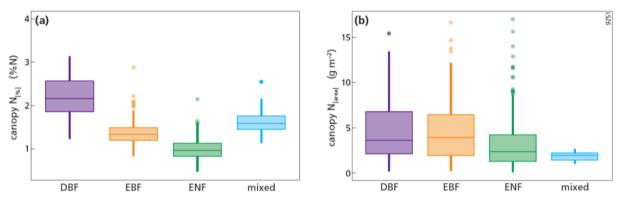


Figure 3. Boxplot of (a) canopy nitrogen (N) concentration (canopy  $N_{[\%]}$ , %N) for Deciduous Broadleaf Forest plots (DBF, n = 80), Evergreen Broadleaf Forest plots (EBF, n = 186), Evergreen Needleleaf Forest plots (ENF, n = 564) and mixed forest plots (mixed, n = 16); (b) canopy N content (canopy  $N_{[area]}$ , g m<sup>-2</sup>) for Deciduous Broadleaf Forest plots (DBF, n = 80), Evergreen Broadleaf Forest plots (EBF, n = 186), Evergreen Needleleaf Forest plots (ENF, n = 563) and mixed forest plots (mixed, n = 12);

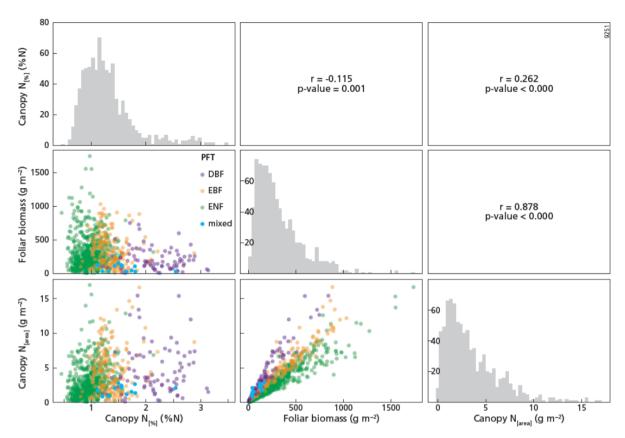


Figure 4. The upper right part of this figure shows the Pearson correlation matrix between canopy  $N_{[\%]}$  (%N), canopy  $N_{[area]}$  (g m<sup>-2</sup>) and foliar biomass (g m<sup>-2</sup>) variables for the whole dataset, n = 841. The diagonal presents the histograms of the variables on the x-axis, while the y-axis represents the number of counts. The lower left part of this figure represents the scatterplots between the variables. PFT = Plant Functional Type, DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest.

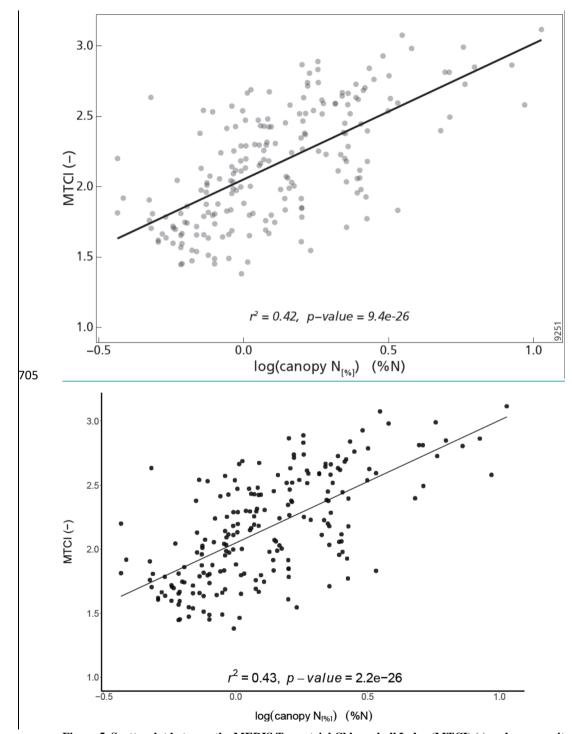


Figure 5. Scatterplot between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen concentration (canopy  $N_{[\%]}$ , %N) after resampling the datasets to 20 km-spatial resolution (n = 204).

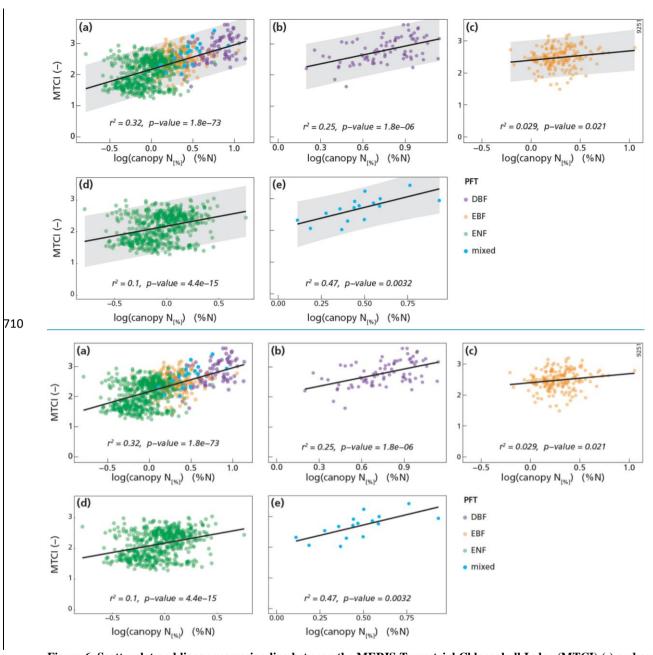


Figure 6. Scatterplot and linear regression line between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen (N) concentration (canopy  $N_{[\%]}$ , %N) for (a) whole dataset (n = 846); (b) Deciduous Broadleaf Forest plots (DBF, n = 80); (c) Evergreen Broadleaf Forest plots (EBF, n = 186); (d) Evergreen Needleleaf Forest plots (ENF, n = 564); (e) mixed forest plots (n = 16). PFT = Plant functional type. The grey shading represents the prediction intervals (95 %). Canopy  $N_{[\%]}$  variable was log transformed to fulfil linear model assumptions.

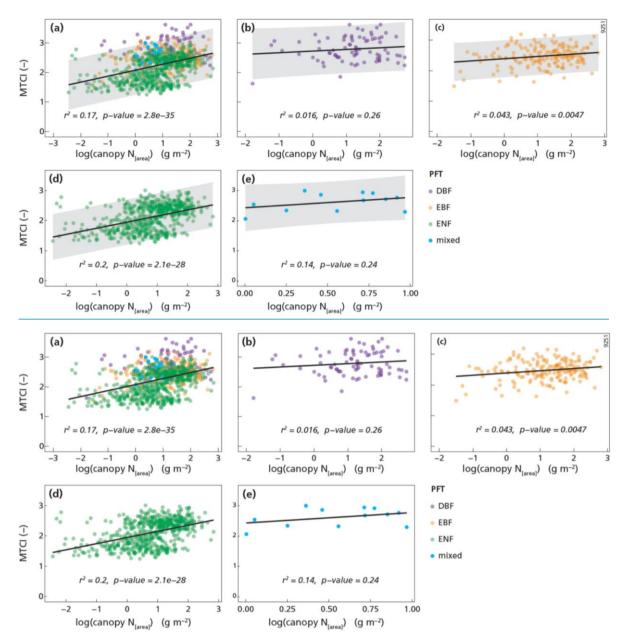


Figure 7. Scatterplot and linear regression line between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy N content (canopy  $N_{[area]}$ , g m<sup>-2</sup>) for (a) whole dataset (n = 841); (b) Deciduous Broadleaf Forest plots (DBF, n = 80); (c) Evergreen Broadleaf Forest plots (EBF, n = 186); (d) Evergreen Needleleaf Forest plots (ENF, n = 563); (e) mixed forest plots (n = 12). PFT = Plant functional type. The grey shading represents the prediction intervals (95 %). Canopy  $N_{[area]}$  variable was log transformed to fulfil linear models assumptions.

Table 54. Descriptive analysis of canopy nitrogen (N) concentration ( $N_{[\%]}$ , g 100g<sup>-1</sup>), foliar biomass (g m<sup>-2</sup>) and canopy N content ( $N_{[area]}$ , g m<sup>-2</sup>) by tree species. PFT = Plant Functional Type, DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest, min = minimum—value, max = maximum—value, mean = averaged—value, sd = standard deviation, a codominant plots refer to the plots where two tree species were dominant in the canopy, b foliar biomass data was lacking for five of the plots. Foliar biomass and canopy N content statistics are thus measured on a restricted number of plots.

		Abundance Canopy N <sub>[%]</sub> (g 100g <sup>-1</sup> )				Foliar biomass (g m <sup>-2</sup> )			Canopy N <sub>[area]</sub> (g m <sup>-2</sup> )						
Species	PFT	Number of plots	(% of total number of plots)	min	max	mean	sd	min	max	mean	sd	min	max	mean	sd
Castanea sativa	DBF	14	1.7	1.62	2.81	2.08	0.36	18.13	425.90	203.46	123.49	0.40	11.99	4.25	2.89
Fagus sylvatica	DBF	15	1.8	1.22	3.13	2.28	0.61	49.94	279.86	173.54	68.70	1.21	7.40	3.96	1.95
Pinus halepensis	ENF	240	28.4	0.56	1.57	0.90	0.19	9.58	827.80	197.23	145.54	0.09	7.29	1.77	1.33
Pinus nigra	ENF	37	4.4	0.56	1.28	0.89	0.19	32.25	923.98	294.29	224.32	0.23	8.87	2.67	2.18
Pinus pinaster	ENF	5	0.6	0.82	1.08	0.93	0.13	271.75	718.87	501.67	211.53	2.30	7.69	4.75	2.25
Pinus pinea	ENF	5	0.6	0.75	1.06	0.95	0.14	103.28	275.50	179.74	66.80	1.08	2.91	1.71	0.75
Pinus sylvestris	ENF	198	23.4	0.67	2.14	1.11	0.20	10.48	828.63	326.44	181.20	0.10	12.86	3.65	2.22
Pinus uncinata	ENF	69	8.2	0.46	1.33	0.87	0.19	183.59	1744.50	687.22	345.21	1.41	16.97	5.92	3.25
Quercus canariensis	DBF	3	0.4	1.97	2.78	2.25	0.46	122.11	197.85	160.32	37.87	2.41	5.51	3.71	1.61
Quercus faginea	DBF	4	0.5	1.49	2.11	1.82	0.31	10.34	419.14	233.47	187.01	0.17	8.83	4.64	4.09
Quercus humilis	DBF	9	1.1	1.53	3.11	2.41	0.42	56.12	337.33	142.65	92.11	1.21	8.64	3.33	2.19
Quercus cerriodes	DBF	17	2.0	1.44	2.80	2.07	0.37	12.97	834.68	262.24	237.49	0.29	15.42	5.06	4.31
Quercus ilex	EBF	160	18.9	0.81	2.87	1.32	0.26	16.63	1033.31	378.23	238.61	0.22	16.61	4.95	3.23
Quercus petraea	DBF	17	2.0	1.37	2.70	2.21	0.41	20.45	741.42	279.96	229.78	0.32	15.37	5.98	4.66
Quercus suber	EBF	23	2.7	1.25	2.08	1.55	0.21	26.26	219.05	110.49	55.65	0.40	4.34	1.72	0.96
Codominant <sup>a</sup>	mixed	30 (25) <sup>b</sup>	3.5	0.92	2.54	1.45	0.41	23.45	342.58	153.70	77.39	0.33	5.74	2.06	1.02

<u>Table 6.</u> Descriptive statistics of the number of plots per pixel, for different spatial resolution (km, pixel length). min = minimum, max = maximum, mean = average, sd = standard deviation.

C4:-114: (1)	Number of plots per pixel					
Spatial resolution (km)	<u>min</u>	max	mean	<u>sd</u>		
<u>5</u>	<u>1</u>	<u>6</u>	1.44	0.77		
<u>10</u>	<u>1</u>	<u>11</u>	2.19	1.53		
<u>15</u>	<u>1</u>	<u>15</u>	<u>3.11</u>	2.59		
20	1	22.	4.09	3.74		

Table 72. Observed linear regression equations between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen concentration ( $CN_{[\%]}$ , %N) for different subgroups. Number of plots (n), determination coefficient ( $r^2$ )<sub>2</sub>-and p-value and Relative Root Mean Square Error of cross-validation (RRMEcv). PFT = Plant Functional type, DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest.

group	<u>n</u>	linear regression	95% confidence interval intercept	95% confidence interval slope	<u>r</u> <sup>2</sup>	p-value	RRMSEcv
overall	846	$MTCI = 2.18 + 0.79 \log(CN_{[\%]})$	[2.15, 2.20]	[0.71, 0.87]	0.32	< 0.000	<u>17.0</u>
DBF	<u>80</u>	$MTCI = 2.07 + 0.95 \log(CN_{[\%]})$	[1.78, 2.36]	[0.59, 1.32]	0.25	< 0.000	12.7
EBF	186	$MTCI = 2.39 + 0.29 \log(CN_{[\%]})$	[2.31, 2.48]	[0.04, 0.54]	0.03	0.021	12.4
<u>ENF</u>	<u>564</u>	$MTCI = 2.13 + 0.61 \log(CN_{[\%]})$	[2.10, 2.17]	[0.46, 0.76]	0.10	< 0.000	<u>19.2</u>
mixed	<u>16</u>	$MTCI = 2.05 + 1.35 \log(CN_{[\%]})$	[1.63, 2.46]	[0.53, 2.17]	0.47	0.003	<u>12.4</u>

group	n	linear regression	95% confidence interval intercept	95% confidence interval slope	r <sup>2</sup>	<del>p-value</del>
overall	846	$MTCI = 2.18 + 0.79 \log(CN[\%])$	[2.148, 2.202]	[0.713, 0.867]	0.32	< 0.000
DBF	80	$MTCI = 2.07 + 0.95 \log(CN[\%])$	[1.782, 2.359]	<del>[0.585, 1.321]</del>	0.25	< 0.000
EBF	<del>186</del>	$MTCI = 2.39 + 0.29 \log(CN[\%])$	[2.311, 2.477]	<del>[0.044, 0.535]</del>	0.03	0.021
ENF	564	$MTCI = 2.13 + 0.61 \log(CN[\%])$	[2.100, 2.169]	<del>[0.463, 0.762]</del>	0.10	< 0.000
mixed	<del>16</del>	$MTCI = 2.05 + 1.35 \log(CN[\%])$	[1.634, 2.463]	<del>[0.534, 2.171]</del>	0.47	0.003

Table 83. Observed linear regressions equations between the MERIS Terrestrial Chlorophyll Index (MTCI) (-) and canopy nitrogen content (CN<sub>[area]</sub>, g m<sup>-2</sup>) for different subgroups. Number of plots (n), determination coefficient (r<sup>2</sup>), p-value and Relative Root Mean Square Error of cross-validation (RRMEcv) and p-value. PFT = Plant Functional type, DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest.

group	<u>n</u>	linear regression	95% confidence interval intercept	95% confidence interval slope	<u>r</u> <sup>2</sup>	p-value	RRMSEcv
Overall	<u>841</u>	$\underline{MTCI} = 2.08 + 0.20 \log(CN_{\underline{[areal]}})$	[2.04, 2.12]	[0.17, 0.23]	0.17	<0.000	18.7
DBF	<u>80</u>	$\underline{MTCI} = 2.72 + 0.06  log(\underline{CN}_{\underline{[area]}})$	[2.58, 2.87]	[-0.04, 0.15]	0.02	0.263	14.7
EBF	<u>186</u>	$\underline{MTCI} = 2.39 + 0.07 \log(\underline{CN}_{\underline{[area]}})$	[2.32, 2.46]	[0.02, 0.12]	0.04	0.005	12.4
ENF	<u>563</u>	$\underline{MTCI} = 1.94 + 0.20  log(\underline{CN}_{\underline{[area]}})$	[1.91, 1.99]	[0.17, 0.24]	0.2	<u>&lt;0.000</u>	18.2
mixed	<u>12</u>	$\underline{MTCI} = 2.43 + 0.34  log(\underline{CN}_{\underline{[area]}})$	[2.05, 2.82]	[-0.26, 0.95]	0.14	0.236	12.8

group	n	linear regression	95% confidence interval intercept	95% confidence interval slope	$\mathbf{r}^2$	<del>p-value</del>
Overall	841	$MTCI = 2.08 + 0.20 \log(CN[area])$	[2.036, 2.116]	[0.172, 0.234]	0.17	<0.000
DBF	80	$MTCI = 2.72 + 0.06 \log(CN[area])$	[2.576, 2.872]	[-0.043, 0.154]	0.02	0.263
EBF	186	$MTCI = 2.39 + 0.07 \log(CN[area])$	<del>[2.315, 2.464]</del>	[0.023, 0.123]	0.04	0.005
ENF	<del>563</del>	$MTCI = 1.94 + 0.20 \log(CN[area])$	[1.907, 1.989]	[0.168, 0.236]	0.20	<0.000
mixed	12	$MTCI = 2.43 + 0.34 \log(CN[area])$	[2.045, 2.815]	[-0.263, 0.949]	0.14	0.236

## 755 11 Appendix

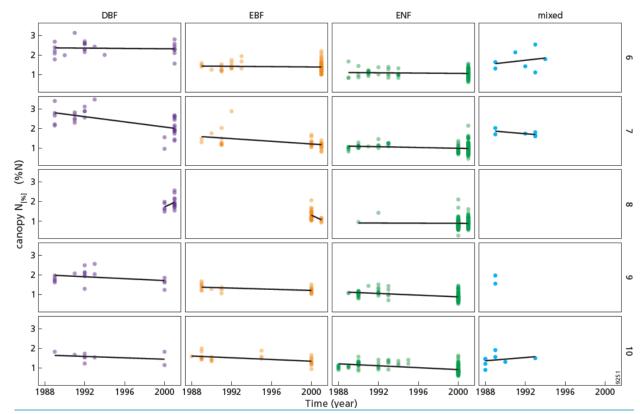


Figure A-2. Inter-annual variation of canopy  $N_{[\%]}$  (%N) for each month included in the analysis. The numbers 6-10 (right side of the figure, row numbers) refer to the month of June, July, August, September and October, respectively. DBF = Deciduous Broadleaf Forest, EBF = Evergreen Broadleaf Forest, ENF = Evergreen Needleleaf Forest, mixed = mixed forest. Each point represents an observation at a forest plot. Note that the forest plots were not sampled multiple times, hence the inter-annual variation encompasses both temporal variation and spatial variation.

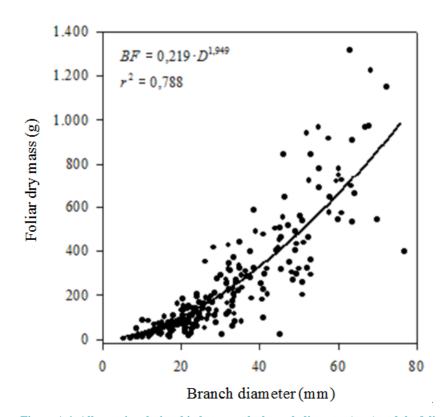


Figure A 1. Allometric relationship between the branch diameter (mm) and the foliar dry mass (g) for *Pinus sylvestris* (n = 208). This is an example of the relationships used to obtain the foliar biomass data of the forest plots. Modified from (Gracia et al., 2004).

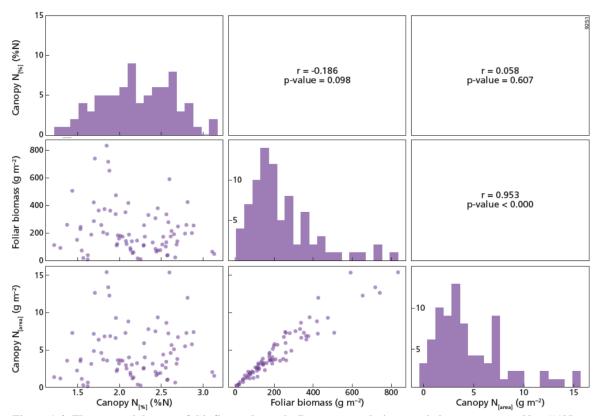


Figure A 3. The upper right part of this figure shows the Pearson correlation matrix between canopy  $N_{[\%]}$  (%N), canopy  $N_{[area]}$  (g m-2) and foliar biomass (g m-2) variables for deciduous broadleaf forest plots (DBF), n = 80. The diagonal presents the histogram of the variable on the x-axis, while the y-axis represents the number of counts. The lower left part of this figure represents the scatterplots between the variables.

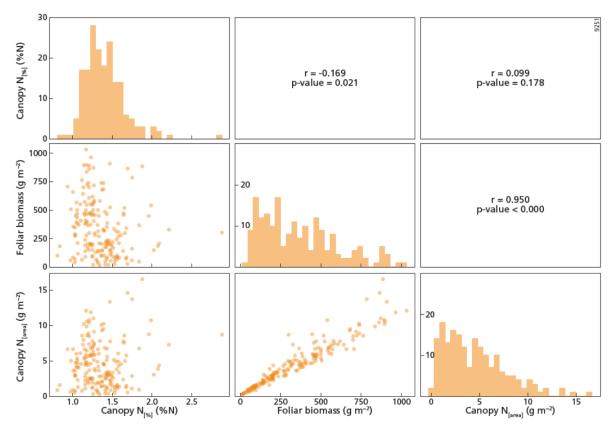


Figure A 4. The upper right part of this figure shows the Pearson correlation matrix between canopy  $N_{[\%]}$  (%N), canopy  $N_{[area]}$  (g m<sup>-2</sup>) and foliar biomass (g m<sup>-2</sup>) variables for evergreen broadleaf forest (EBF) plots, n = 186. The diagonal presents the histogram of the variable on the x-axis, while the y-axis represents the number of counts. The lower left part of this figure represents the scatterplots between the variables.

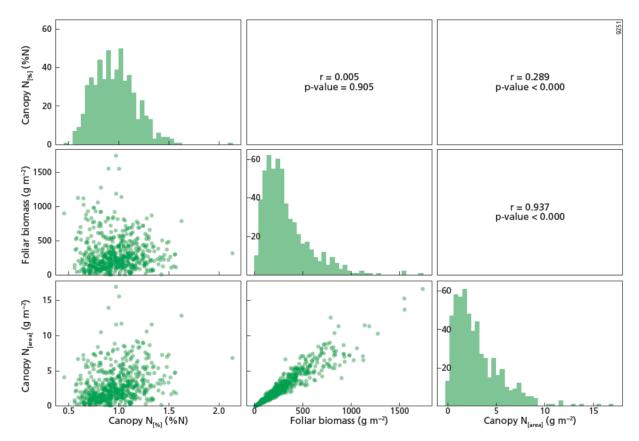


Figure A 5. The upper right part of this figure shows the Pearson correlation matrix between canopy  $N_{[\%]}$  (%N), canopy  $N_{[area]}$  (g m<sup>-2</sup>) and foliar biomass (g m<sup>-2</sup>) variables for evergreen needleleaf forest (ENF) plots, n = 563. The diagonal presents the histogram of the variable on the x-axis, while the y-axis represents the number of counts. The lower left part of this figure represents the scatterplots between the variables.

<u>Species</u>	Foli	Foliar biomass $= a \cdot DBH^b$					
	<u>a</u>	<u>b</u>	<u>n</u>	<u>r</u> <sup>2</sup>			
Castanea sativa	0.032	1.669	<u>86</u>	0.49			
Fagus sylvatica	0.026	<u>1.546</u>	<u>285</u>	0.66			
<u>Pinus halepensis</u>	0.037	1.656	2420	0.65			
<u>Pinus nigra</u>	0.022	1.870	1641	0.65			
<u>Pinus pinaster</u>	0.034	1.848	<u>169</u>	0.67			
<u>Pinus pinea</u>	0.014	2.029	<u>335</u>	0.72			
<u>Pinus sylvestris</u>	0.036	1.651	<u>2755</u>	0.66			
<u>Pinus uncinata</u>	0.087	1.410	<u>770</u>	0.62			
Quercus canariensis	0.120	1.322	<u>36</u>	0.57			
Quercus faginea	0.197	0.943	<u>170</u>	0.40			
Quercus humilis	0.047	1.462	<u>595</u>	0.59			
Quercus cerrioides	0.023	1.805	138	0.73			
Quercus ilex	0.063	1.576	<u>2151</u>	<u>0.60</u>			
Quercus petraea	0.014	1.888	<u>121</u>	0.73			
<u>Quercus suber</u>	0.026	1.446	<u>314</u>	<u>0.55</u>			

<u>Table A 3.</u> Descriptive statistics of the number of plant functional types (PFT) per pixel, by pixel spatial resolution (km). min = minimum, max = maximum, mean = average, sd = standard deviation.

Spatial resolution (km)	Number of PFT per pixel						
<u>Spatial resolution (Kili)</u>	min	max	mean	<u>sd</u>			
<u>5</u>	<u>1</u>	<u>3</u>	<u>1.08</u>	0.29			
<u>10</u>	<u>1</u>	<u>4</u>	<u>1.22</u>	0.48			
<u>15</u>	<u>1</u>	<u>4</u>	<u>1.34</u>	0.61			
<u>20</u>	<u>1</u>	<u>4</u>	1.45	0.69			

<u>Table A 4.</u> Descriptive statistics of the number of species per pixel, by pixel spatial resolution (km). min = minimum, max = maximum, mean = average, sd = standard deviation.

Creation accounting (Irms)	Number of species per pixel						
Spatial resolution (km)	min	max	mean	<u>sd</u>			
<u>5</u>	<u>1</u>	<u>4</u>	<u>1.14</u>	0.41			
<u>10</u>	<u>1</u>	<u>4</u>	1.38	0.67			
<u>15</u>	<u>1</u>	<u>4</u>	<u>1.58</u>	0.85			
<u>20</u>	<u>1</u>	<u>6</u>	<u>1.79</u>	1.07			

# 805 <u>Table A 5. Descriptive statistics of the number of sampling years per pixel, by pixel spatial resolution (km). min = minimum, max = maximum, mean = average, sd = standard deviation.</u>

Control masslytion (Irm)	Number of sampling years per pixel					
Spatial resolution (km)	min	max	mean	<u>sd</u>		
<u>5</u>	<u>1</u>	<u>2</u>	1.02	0.15		
<u>10</u>	<u>1</u>	<u>3</u>	1.07	0.26		
<u>15</u>	<u>1</u>	<u>3</u>	<u>1.10</u>	0.33		
<u>20</u>	<u>1</u>	<u>3</u>	1.14	0.40		