

Communication

A New Approach to Monitor Soil Microbial Driven C/N Ratio in Temperate Evergreen Coniferous Forests Managed via Sentinel-2 Spectral Imagery

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Abstract: Forests are key ecosystems for climate change mitigation, playing a pivotal role in C and N land sequestering and storage. However, the sustainable management of forests is challenging for foresters who need continuous and reliable information on the status of soil conditions. Yet, the monitoring of soils in temperate evergreen forests, via satellite data, is jeopardized by the year round prevailing heavily dense canopy. In this study, the Sentinel-2 spectral imagery derived normalized difference vegetation index (NDVI), proved to be a reliable tool to determine the C/N ratio in two managed pine-dominated forests, in southern Poland. Results showed a strong negative correlation between NDVI values and the on-site C/N ratios measured at the upper soil horizons in 100 and 99 randomly distributed sampling points across the Kup ($r^2 = -0.8019$) and Koniecpol ($r^2 = -0.7281$) forests. This indicates the feasibility of using the NDVI to predict the microbial driven soil C/N ratio in evergreen forests, and to foresee alterations in the vegetation patterns elicited by microbial hindering soil abiotic or biotic factors. Spatial/temporal variations in C/N ratio also provide information on C and N soil dynamics and land ecosystem function in a changing climate.

Keywords: remote sensing; soil properties; land vegetation index; Pinus sylvestris

1. Introduction

Coniferous temperate evergreen forests are key ecosystems for climate change mitigation, as they play a major role in carbon (C) and nitrogen (N) soil sequestration and storage [1,2]. Among soil properties, the C/N ratio is pivotal to soil biology and soil microbe community composition across soil types and climate zones [3]. The C-enriched root exudates in the rhizosphere of evergreen woody species are known to stimulate microbial N mineralization, thus maintaining the microbial C/N stoichiometric balance, via alterations in the microbiota taxonomic structure [4]. In forest ecosystems, the microbial necromass contributes to approximately 35% of the soil organic C content in the upper soil horizons [5,6]. While, soil and microbial N status plays a central role in the heterotrophic nitrifying activities, via the activity of soil N₂-fixing bacteria [7–9]. The uppermost soil horizons are also known to harbor a greatest bacterial diversity and fungal biomass, when compared to deepest horizons, involved in important biogeochemical processes such as the degradation of soluble carbohydrates and polysaccharides, as well as the metabolism of Ccompounds [10,11]. Under coniferous trees, the upper soil horizon also plays a critical role in organic C storage [2,12], although its sensitivity to climate change rising temperatures might transform its storage capacity into a net source of CO₂ [13].

In the case of managed evergreen pine forests, the maintenance of ecosystem resilience is crucial under climate change at multiple scales [10], as soil biology is directly modulated



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by the addition of synthetic fertilizers and the intensity of seasonal tree felling and replanting. As previously shown, intensive timber harvesting is known to hamper the soil C-sink capacity, concomitantly increasing the soil CO_2 and CH_4 emissions to the atmosphere [14]. Nevertheless, the sustainable forest-management is challenging for foresters who need continuous and reliable information on the status of soil conditions, since: (a) continuous on-site determinations of the soil C/N ratio across large forest sites is a laborious and time-consuming process, and (b) the monitoring via satellite data of soil properties in temperate evergreen forests, is jeopardized by the year-round dense canopy [15].

The normalized difference vegetation index (NDVI) selected for this study has been previously determined to be relevant for: (i) dendrochronological studies in boreal forests [16], (ii) the analyses of the degradation or loss of vigor of Romanian forests [17], (iii) the determination of the impact of anthropogenic activities on the aboveground biomass C dynamics [18], and (iv) significant improvements in forest conservation and sustainability [19], owing to the well-documented intimate relationships between soil biology and plant growth [20]. At the same time, freely available temporal, spatial, and spectral Sentinel-2 imagery has been successfully used for monitoring the impact of seasonal drought events on forest health [21,22], allowing a better sustainable management in terms of plantation inputs and production cost optimization [23,24]. Thus, the aim of this research was to test a new approach to address the use of Sentinel-2 imagery to estimate the soil microbial driven C/N ratios in the Kup and Koniecpol Forest districts of southern Poland, both of them known to be drought-prone [25].

2. Materials and Methods

2.1. Description of Forest Sites

This research was conducted in randomly scattered 100 sampling points across the Kup forest district (50°49'37" N, 17°52'59" E), and 99 sampling points across Koniecpol forest district (50°41'16" N, 19°46'16" E) in southern Poland (Figure 1). Both forests have a similar history of stands, growing season (212–215 days), accumulated annual precipitation (629–635 mm), average mean annual temperature (7.8–8.5 °C), soils developed from fluvioglacial sand, clay, and peat sediments, as well as a flat topography dominated by *Pinus sylvestris* L. The negligible differences between the Kup and Koniecpol forests in terms of stands, and edaphoclimatic prevailing conditions allowed: (a) the doubling of the studied area, without the side effect of varying soil and climatic parameters; and (b) the assumption of an uniform pine needles litter fall, undergoing similar decomposition rates [26] carried out by a decomposer microbial community displaying many redundant functions [27], modulated by a dominant plant species [28].



Figure 1. The sampling locations are marked with points. The two study sites are overlaid on the Poland map. The Kup (**left**) and Koniecpol (**right**) forest districts are highlighted.

2.2. Soil C and N Contents

At each sampling point in both forests, soils were collected up to a depth of 20 cm, during the summer of 2019. Soils were cleaned of stones and plant debris to be transported to the soil analytical laboratory at the University of Agriculture in Krakow. The total soil C and total N contents were determined in air dried subsamples sieved through a 2-mm mesh, using a LECO CNS True Mac Analyzer (Leco, St. Joseph, MI, USA). Average total C and N values were: (a) $25.92 \pm 14.92\%$ and $1.11 \pm 0.57\%$ in the Kup forest, and (b) $29.20 \pm 15.65\%$ and $1.22 \pm 0.68\%$ in the Koniecpol forest. The C and N values for each sampling point were used to calculate the C/N ratios.

2.3. Sentinel-2 Imagery and Vegetation Indices

Cloud-free Sentinel-2 imagery, taken at the same dates of the soil collection in individual sampling points at each forest site, were downloaded from the Copernicus Open Access Hub, as a Level-2A bottom-of-atmosphere. The normalized difference vegetation index (NDVI) was calculated using the near-infrared (NIR) and red edge (RED) reflectances, corresponding to the B05 and B08 bands in Sentinel-2 imagery, respectively (Table 1), both having 10×10 m spatial resolution, following the standard equation:

$$NDVI = \frac{NIR - RED}{NIR + RED}$$

Table 1. Sentinel-2 band parameters.

Band	Spectral Range (nm)	Pixel Size (m)	Name
B01	432–453	60	Atmospheric correction
B02	458–523	10	VIS-BLUE
B03	543–578	10	VIS-GREEN
B04	650–680	10	VIS-RED
B05	698–713	20	RED EDGE
B06	733–748	20	RED EDGE
B07	773–793	20	RED EDGE
B08	785–900	10	NIR
B8A	855-875	20	NIR narrow
B09	935–955	60	Water vapour absorption
B11	1565–1655	20	SWIR
B12	2100-2280	20	SWIR

The raster maps obtained were masked using a vector file with the boundary of the Kup and Koniecpol forest districts. The Pearson correlation (r^2) was used to analyze linear correlations between the Sentinel-2 imagery derived NDVI and the on-site measured C/N ratio, at a significant level of 95%. The data set was divided into two subsets by a pseudo-random approach, one for the model calibration and the other for validation with a proportion of 80% and 20%, respectively. This approach facilitates the replication of the results. For Kup forest, the training subset was n = 80, and the validation subset was n = 20. For the Koniecpol forest, the data in each subset were 79 and 20, respectively. A linear regression was performed based on the determined NDVI values and soil C/N ratios. After obtaining the linear models, the soil C/N ratio maps were created from the NDVI maps and compared with a natural color composition for each forest area (Figure 2). The bare soil, urban zones and other non-vegetated areas were left off in these maps.



Figure 2. Predicted C/N ratio and natural colour composition for comparing the forest areas.

3. Results

On-site analyses revealed non-significant differences in soil C/N ratios or NDVI values between both forests (Table 2), even though the statistical mean, minimum, and Q3 soil C/N as well as NVDI values were relatively higher in Koniecpol when compared to those in the Kup forest. The coefficients obtained from the regression model allowed the construction of an NDVI map linked to the soil C/N ratio map for the two forests. In Figure 3, geographical zones colored in yellow had higher NDVI values. The model performance was evaluated with the validation subset, obtaining the coefficient of determination for Kup forest ($R^2 = 0.8156$) and Koniecpol forest ($R^2 = 0.7760$) (Figure 4). The residuals assumptions were met in both models, with normality D'Agostino and Pearson's test (p < 0.05) of 0.8087 and 0.8706 for Kup and Koniecpol forests, respectively.



Figure 3. Calculated NDVI for the Kup (left) and Koniecpol (right) forest districts.

	Forest Districts		
Soil C/N Ratio	Кир	Koniecpol	
Mean ± STd	21.62 ± 5.78	23.45 ± 6.49	
Minimum	10.14	11.25	
Maximun	32.36	39.17	
Q1	17.66	18.00	
Q3	26.22	28.07	
NDVI Mean ± STd	0.76 ± 0.07	0.86 ± 0.06	

Table 2. Soil C/N ratio and Sentinel-2 derived Normalized Difference Vegetation Index (NDVI) determined in 100 sampling points in the Kup forest district and 99 sampling points in the Koniecpol forest district, in Southern Poland.



Figure 4. Regression analysis for the Kup (**top**) and Koniecpol (**bottom**) forest districts. The plots on the left show the measured C/N ratio, while the plots on the right show the predicted C/N.

4. Discussion

Worldwide, the Sentinel-2 imagery derived NDVI is the most commonly used vegetation index showing the strongest relationship with a large number of driving factors of vegetation changes [9]. The NDVI ranking between 0.6188 and 0.9823, could be considered as an indicator of the good plant-soil interactive conditions in both forest districts [29,30]. On the other hand, the soil C/N ratio is tightly associated with plant nutrient acquisition through various strategies such as forming mutualistic associations with N₂-fixing rhizobacteria and arbuscular mycorrhizal fungi, or the stimulus of soil microbial activity to mobilize nutrients from soil organic matter through root exudates [31–33]. It is interesting to note the significant positive correlation previously recorded between the C/N ratio and the activity of the prokaryote-excreted β -glucosidase enzyme directly involved in the C cycle [34] in soils of the Manowo Forest District, northern Poland [35]. In the present study, the soil C/N ratios ranged between 10 and 32 in both forests, coinciding with values reported for forests dominated by non N₂-fixing tree species [36].

Most important is the strong negative correlation detected between the Sentinel-2 imagery-derived NDVI and the on-site determined soil C/N ratios, confirming the value of this vegetation index as an indicator of the biological properties of the upper soil horizons

in temperate evergreen southern Poland's forests. This observation was further supported by the intimate relationship existing between soil nutritional levels and NDVI in vegetated areas [37–39]. According to Li et al. [40], NDVI values could replace field soil investigations, so facilitating the large-scale monitoring of soil quality after land consolidation. Moreover, the average soil C/N ratios above 21 recorded in both forests strongly suggests the need to minimize losses of soil-bioavailable N, which are known to limit soil C-sink activity, thus simultaneously constraining net primary productivity as well as the capacity for forests to respond dynamically to disturbance and environmental changes [41]. Whereas, soil C/N ratios >30 should alert foresters on the possibility of increasing soil N₂O emissions toward the atmosphere [42].

5. Conclusions

The present study indicates the feasibility of using Sentinel-2 spectral imagery to infer, in real time, soil biology status across large forest areas, based on the strong negative correlation between the on-site measured soil C/N ratio and NDVI values, at both forest sites. The high accuracy obtained with this approach supports the use of Sentinel-2-derived NDVI to predict the long term hindering effects of drought, snow melting temperatures, seasonal fires, pests and plant diseases, as well as the over felling of trees on soil dynamics and the sustainability of the forest ecosystem. This new approach for estimating the soil C/N ratio under coniferous evergreen trees can provide reliable information on the soil status for the decision-making process underlying the sustainable management of the forest. However, more research is needed to validate the effectiveness of NVDI as an indicator of soil C/N ratio under soil and environmental conditions different to those prevailing in Poland's Kup and Koniecpol forests.

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