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Warsaw, 21.02.2025

Review of Doctoral Dissertation

Author: Michal Antala

Title: Assessment of peatland vegetation physiology under manipulated environmental conditions by leaf and canopy level chlorophyll fluorescence measurements

The basis for conducting a review

The basis for the implementation of this review was the letter of prof. dr hab. inż. Mariusz Sojka, The Scientific Council of the Discipline of Environmental Engineering, Mining, and Energy Chairman. The work sent for review was carried out under the supervision of prof. UPP dr hab. inż. Anshu Rastogi and assistant supervisor dr inż. Marcin Stróżecki.

Basic information about the PhD candidate

Mr. Michal Antala was born in June 11 1996, and has completed the university master study programme at Slovak University of Agriculture in Nitra in Plant Nutrition and Protection in the field of study Agriculture and Landscape in 2020. This same year, he successfully applied for PhD studies at Poznań University of Life Sciences.

He has experience as an external staff at the Slovak University of Agriculture (from 07. 2020 to 09.2020). He has completed three research internships: Poznań (Poland), Freiburg (Germany), and Milano (Italy). He is also a laureate of the Erasmus+ program.

PhD candidate has already participated as a contractor or principal investigator in 5 research grants funded by various government and European agencies (National Research Science, EEPN). He is the author of 12 scientific publications published in internationally recognized journals (with an impact factor). He is also the author of one book chapter in a monograph published by Elsevier. He has participated in seven international scientific conferences (such as EGU). He is a member of 3 associations: American Geoscience Union, Ecological Society of America, and Society of Wetland Scientist.

General description of dissertation

The dissertation entitled **"**Assessment of peatland vegetation physiology under manipulated environmental conditions by leaf and canopy level chlorophyll fluorescence measurements" was prepared and presented for evaluation based on four articles, which constitute an integral part of the dissertation. Doctoral dissertation has been provided in printed and digital form and includes, among others:

1. Extended abstract of dissertation,
2. Publications,
3. Declaration of co-authors.

The title of the dissertation is well formulated and adequately expresses the research aim, which is to evaluate the physiological changes caused by warmer and drier conditions in peatland species at the leaf and canopy levels measured by active and passive chlorophyll fluorescence techniques. The Extended abstract consists of 6 relevant chapters. Chapter 1 is *Introduction*, chapter 2 is *Aims and hypotheses*, chapter 3 is *Material and methods*, chapter 4 is *Characteristic of obtained results*, chapter 5 is *Discussion*, and 6 is *Conclusions*. This part of the dissertation is 36 pages long. Generally, this part of the dissertation is at a very high and international scientific level with very well-elaborated results. The text is well-written, clear, logical, and professionally formulated. The structures in the text are generally proper and help the reader get an enhanced and fast orientation to the topic. Moreover, the Author used the appropriate number and quality of the bibliography and correctly quoted it in this thesis. Undoubtedly, the Author has deep theoretical knowledge and a good orientation in the investigated problem appropriately discussed in the dissertation.

The next part of the dissertation is four attached articles. All of them have been published in journals with an impact factor (IF). These are the following works:

1. Antala, M., Juszczak, R., van der Tol, Ch., Rastogi, A. (2022). Impact of climate change induced alterations in peatland vegetation phenology and composition on carbon balance. Science of the Total Environment. 827, 154294. https://doi.org/10.1016/j.scitotenv.2022.154294. **200 points; IF = 9.82.**

2. Antala, M., Juszczak, R., Rastogi, A., (2024). Nonphotochemical quenching does not alter the relationship between sun-induced fluorescence and gross primary production under heatwave. New Phytologist. 20312. https://doi.org/10.1111/nph.20312. **140 points; IF = 8.33.**

3. Antala, M., Rastogi, A., Cogliati, S., Stróżecki, M., Colombo, R., Juszczak, R. (2024). Sun-induced fluorescence spectrum as a tool for assessing peatland vegetation productivity in the framework of warming and reduced precipitation experiment. Remote Sensing of Environment. 301, 113921. https://doi.org/10.1016/j.rse.2023.113921. **200 points; IF = 11.14**.

4. Antala, M., Abdelmajeed, A.Y.A., Stróżecki, M., Krzesiński, W., Juszczak, R., Rastogi, A. (2024). Photosynthetic Responses of Peat Moss (*Sphagnum* spp.) and Bog Cranberry (*Vaccinium oxycoccos* L.) to Spring Warming. Plants 13 (22), 3246. https://doi.org/10.3390/plants13223246. **70 points; IF = 4.0**

The total IF of these papers is 33.29, and the number of points of the Ministry of Science and Higher Education is 610. The PhD candidate is the first author of all the papers. It can, therefore, be concluded that his participation is significant and entitles him to use it as the basis for awarding a doctoral degree.

The content of the thesis, its goals, methods, and structure fully comply with the requirements for doctoral dissertations and the principles adopted in technical sciences. The main topic of the work concerns the possibility of monitoring the vegetation of peatlands by using fluorescence. It should be emphasized that the subject of the work is very actual, as remote observation and accurate quantification of photosynthetic activity of peatland vegetation are unresolved problems, and often, the results of estimations are very biased. Sun-induced chlorophyll fluorescence (SIF) seems to be a tool that will make it possible to monitor the photosynthesis process more closely. Therefore, it should be recognized that the subject matter taken up by the PhD candidate is topical and of great utilitarian importance.

In the *Introduction* chapter, much space is devoted to the characteristics of the peatland ecosystems, mainly their ecological functions. The technique of chlorophyll fluorescence (especially remote measurements of SIF) was described. PhD candidate indicates that before using satellite-derived SIF for global photosynthesis assessment, the relationship between photosynthesis and SIF must be investigated for different ecosystems, including peatlands, and information provided by SIF has to be understood in the broader context of plant physiology.

Based on a literature review, PhD candidate showed that climate change is one of the key challenges of the 21st century, and its impacts, such as rising temperatures and increasing frequency of extreme weather events, affect peatland ecosystems, which play a crucial role in the global carbon cycle by storing vast amounts of carbon. Their functioning depends on vegetation composition, particularly on Sphagnum mosses, which maintain conditions

favorable for peat accumulation. However, climate change may disrupt the ecological balance of peatlands, leading to carbon release and reinforcing the greenhouse effect. The PhD candidate demonstrated that peatlands are underrepresented in climate models due to their remote locations and limited field studies. He thus highlighted the need for a better understanding of the relationship between the spectral properties of peatlands and their ecological functions, particularly in terms of carbon assimilation, which could enhance the use of satellite remote sensing for their monitoring.

The analysis of the current state of theoretical knowledge and practical solutions carried out by the PhD candidate allowed him to set the following goals:

1. Describe the changes in peatland vegetation composition and phenology under future climatic conditions and derive their implications for the carbon balance of peatlands based on the currently available literature.

2. Examine the potential of remote sensing in assessing the vegetation's physiological status and photosynthetic activity with a particular focus on SIF and peatland vegetation.

3. Assess the impact of warming alone and in combination with reduced precipitation on peatland vegetation physiology by remote sensing while taking into account vegetation communities of different compositions.

4. Investigate the influence of elevated temperature on different peatland vegetation plant species, focusing on their photosynthetic apparatus activity assessed by chlorophyll *a* fluorescence measurements.

In the opinion of the Reviewer, these aims were generally set correctly. After an adequately conducted introduction, there is no doubt that the aims set in the work are scientifically justified. I only have doubts about the first aim, which will be discussed later in the review.

Based on the analysis of research issues, the following research hypotheses were formulated:

1. Climate change will induce a shift in the peatland vegetation towards a higher abundance of vascular plants, which will have a more extended vegetation season due to warming. These changes will enhance the vegetation's photosynthetic rate, but due to increased respiration rate, peatlands will become weaker carbon sinks or even carbon sources.

2. Remote sensing metrics such as vegetation indices and SIF are robust proxies for peatland vegetation photosynthesis and can be used to track the physiological changes of vegetation in current and future warmer conditions.

3. Warming and reduced precipitation induce changes in peatland vegetation structure, phenology and physiology that are traceable as changes in greenness related vegetation indices or SIF.

4. Increased temperature impacts individual peatland PFGs differently, with vascular plants being impacted positively and mosses negatively.

In the chapter *Material and methods*, the PhD candidate provides information about the site of experiments. The majority of the research work has been conducted in the Rzecin peatland area, which is located in the western part of Poland. It should be emphasized that the research program itself is comprehensive. In 2017, two research sites (CL and CR) were established to assess the impact of warming and reduced precipitation on peatland vegetation. Each site consisted of nine experimental plots in three variants: control (C), warmed (W), and warmed with reduced precipitation (WP). Transparent chambers and infrared heaters provided heating, while WP plots had additional curtains to limit rainfall. Temperature and humidity measurements were taken every 30 minutes using thermo-hygrometers, and groundwater levels were recorded with piezometers. Photosynthetically active radiation was measured using a sensor mounted on a 3-meter tower. The research included spectral analyses (NDVI, PRI, NIRv) and SIF fluorescence, measured with the Piccolo Doppio system. CO_2 fluxes were assessed using a closed chamber method. Data were statistically analyzed in RStudio using ANOVA and Kruskal-Wallis tests. It should be noted that the research was carried out in cooperation with a very wide team of specialists from various countries, which is also reflected in the list of co-authors of individual publications.

Moving on to the work results, it should be noted that the PhD candidate showed that the assumed research goal was consistently pursued. The attached documentation, which the PhD student presents here, has been carefully prepared, and the statistical methods used in the research are correct. The results obtained by the PhD candidate showed that ericaceous shrubs, such as bog cranberry, benefit from spring warming by initiating photosynthetic activity earlier. The PSII structure of evergreen plants in temperate and boreal zones changes in winter to protect them from intense light at low temperatures, causing a decrease in their efficiency. Leaf-level measurements indicated that the rate of spring reactivation of reaction centers and PSII photochemistry depends on temperature and increases with experimental warming (W) despite measurements conducted during the warmest spring on record.

Peat mosses remain photosynthetically active even under snow cover as long as they are not frozen and continue growing during winter. The PhD candidate demonstrated that the

constant low dependence on non-regulated heat dissipation and high dependence on regulated dissipation align with previous studies. Since peat mosses do not require prolonged warm weather for photosynthetic activity, they derive relatively fewer benefits from warming than shrubs. The PhD candidate rightly concludes that the effective utilization of the photosynthetic potential by shrubs may contribute to the observed increase in their abundance in peatlands. The PhD candidate interprets increased SIF fluorescence of CR vegetation under W and WP conditions as an increase in vascular plant cover.

The results of this study suggest that warming has a more substantial impact on peatland communities that previously had sparser vascular plant cover. However, the dominance of the far-red peak in the SIF spectrum throughout the year suggests that a significant portion of the surface in the studied vegetation communities in C, W, and WP is occupied by evergreen shrubs. This study observed physiological changes in peatland vegetation at both the leaf and canopy levels. As previously mentioned, W caused an earlier start of photosynthetic activity in bog cranberry. Additionally, in spring, the SIF fluorescence of both studied plant communities exposed to W and WP increased more rapidly than in C. This suggests physiological and structural changes resulting from warmer and drier conditions.

The results of this study indicate that changes in the composition and phenology of peatland vegetation stem from plant physiology and further impact vegetation photosynthesis. Higher GPP values in CR vegetation during summer, caused by W and WP, provide the most direct evidence of these changes. As previously demonstrated, a more significant share of vascular plants in vegetation leads to stronger carbon assimilation at the season's peak. The extended growing season identified in this study suggests a further increase in the annual GPP value of peatlands under warmer conditions. SIF fluorescence measurements at different wavelengths and their relationship with GPP showed that the multi-wavelength regression approach yields more accurate and stable results than traditional remote sensing indices. The strong correlation between far-red SIF and GPP suggests that these metrics can effectively assess peatland plant photosynthesis dynamics. The PhD candidate correctly noted that under prolonged heatwaves, the relationship between SIF and GPP may become disrupted, primarily due to stomatal closure, which limits gas exchange but does not equally affect the light phase of photosynthesis. Therefore, thermal remote sensing may be a valuable complementary tool for monitoring plant physiology under stress conditions.

The PhD candidate summarized the results described above in chapter 6, *Conclusions*. The well-documented summary included in this chapter fully meets the work's set goals. In formulating this summary, the PhD candidate showed both criticism and due caution. Mr.

Michal Antala's doctoral dissertation has been correctly and carefully prepared for editing and meets the formal and substantive requirements for doctoral dissertations. As mentioned in the introduction of this review, an integral part of the dissertation is four articles published in international scientific journals with an impact factor (IF), which have certainly been peer-reviewed by high-class specialists. In my opinion, the material presented in these articles also meets the formal and substantive requirements for the doctoral degree award. This dissertation brings new cognitive and utilitarian elements, enriching knowledge in the field of the possibility of monitoring plant communities by sun-induced fluorescence. This work is a closed research cycle from the concept through careful selection of site, appropriate methods, and discussion of own results and their confrontation with the views of other authors.

Evaluation of the PhD dissertation

The study addresses the crucial issue of climate change and its impact on peatlands, which play a significant role in the global carbon balance. These findings are important for both scientific research and environmental policy. The combination of ecophysiological, phenological, spectral studies, and mathematical modeling makes this dissertation comprehensive, providing a multifaceted analysis of the issue. Using sun-induced fluorescence (SIF) and multi-wavelength regression to model GPP represents innovative approaches that can improve the accuracy of predictions regarding changes in peatland ecosystems. The results can be helpful in monitoring and protecting peatlands and for forecasting their future role as carbon sinks or sources in a changing climate. The dissertation references a wide range of studies, demonstrating a strong understanding of the current knowledge of peatland ecology.

The dissertation presents a solid theoretical foundation covering a broad range of topics related to climate change, peatland ecosystems, their role in the carbon cycle, and the impact of climate change on vegetation in these ecosystems. It provides a well-justified theoretical background, supported by numerous references to current scientific studies and literature, demonstrating a strong grasp of the subject. The researcher shows an understanding of climate change mechanisms, peatland functions, and remote sensing techniques, which allows for the formulation of theoretical bases for the research.

The dissertation demonstrates the ability to conduct independent scientific research on multiple levels. The author has defined the research objective and hypotheses and designed a comprehensive research approach that includes literature reviews and experimental studies. Advanced tools, such as remote sensing (SIF) and chlorophyll fluorescence methods, showcase proficiency in working with modern technologies. The developed experiments, including

studies on vegetation phenology changes, fluorescence measurements under different climatic conditions, and warming experiments, indicate the work results from meticulous and well-planned research.

The dissertation presents an original approach to the research problem. The application of remote sensing, particularly sun-induced fluorescence (SIF), to monitor peatland vegetation physiology in the context of climate change is an innovative solution. The author investigates the impact of climate change on peatland vegetation using advanced remote sensing methods, offering a novel perspective on assessing peatland ecosystem functioning. Additionally, the interactions between vascular plants and mosses, their responses to changing climatic conditions, and their influence on the carbon balance represent an innovative approach, considering the complex feedback mechanisms between vegetation groups and climate factors.

Critical remarks and questions:

1) The author has precisely defined the significance of peatlands as an ecosystem. The research objective has been correctly formulated. However, it could be further refined to include an investigation of the mechanism by which climate change (warming and reduced precipitation) affects the physiological processes of mosses. This is especially relevant since the measurements of chlorophyll fluorescence parameters allow for such an analysis.

2) Formulating utilitarian objectives in this study would have been possible.

3) In my opinion, Objective 1 is unnecessary. Defining the research problem based on the literature is a standard component of a PhD dissertation. I also consider placing subchapter 3.1 (*Literature review*) in chapter 3 (*Materials and methods*) as pointless, even if a review publication is attached to the dissertation.

4) In the extended abstract of the dissertation, the scope of whole work could be added as a subchapter.

4) Although all the studies have been published in renowned journals (and thus have been peer-reviewed by specialists), I am surprised by the use of the phrase *"dramatically decreased"* in one of the papers. A relationship is either statistically significant or not. Similarly, the phrase *"broken linearity"* is problematic—such a term does not exist in statistics.

5) Article II includes a supplement, which should be attached to the PhD dissertation if the article is part of this dissertation.

6) In the research methodology of Article III, a closed chamber was used to measure gas exchange. Why was this particular system chosen instead of an open one, and what are their differences?

7) In the doctoral candidate's opinion, what could climate change's effects (warming and lack of precipitation) be on the biodiversity of plant communities in peatlands?
8) The conclusions lack suggestions (proposals) for future research directions.
9) The *References* is a regular chapter and should have its number (in the case of this dissertation, it should be chapter number 7). Moreover, the page number of the next chapter in the Table of Contents (Attachments of published articles) is wrong, now it is 377.

Conclusions

The doctoral dissertation of Mr. Michal Antala is a complete scientific study based on multi-faceted research in which data was collected and processed using the latest methods. This work has both cognitive and practical values. The collected material and its critical discussion prove the PhD candidate's skills in the use of various research methods and statistical methods. The doctoral dissertation is a strong and valuable contribution to research on the impact of climate change on peatlands. It employs modern analytical methods and provides significant insights into changes in vegetation structure and productivity. The PhD candidate has demonstrated substantive knowledge and a good knowledge of professional literature in formulating the goals of the work and conclusions resulting from the experiments. Undoubtedly, the PhD candidate is a talented and independent researcher whose research is at a level that does not deviate from current European standards.

PhD thesis of Mr. Michal Antala meets all the requirements for doctoral dissertations by the Act of 20 July 2018 Law on Higher Education and Science. Considering my positive assessment, I am applying to the Scientific Council of the Discipline of Environmental Engineering, Mining, and Energy Chairman for admission to Mr. Michal Antala to the next stages of the doctoral dissertation.

I would like to submit a request to the Scientific Discipline Council for the distinction of the reviewed dissertation. The PhD dissertation deserves distinction due to its outstanding scientific value, innovative approach, and comprehensive research methodology. It addresses the critical issue of climate change and its impact on peatlands, essential for the global carbon balance. The study integrates ecophysiological, phenological, spectral analyses, and mathematical modeling, demonstrating an interdisciplinary and multifaceted approach to the subject. A key strength of the dissertation is the use of sun-induced fluorescence (SIF) and multi-wavelength regression to model Gross Primary Production (GPP), representing a cutting-

edge method for improving predictions of peatland ecosystem dynamics. This novel application of remote sensing significantly enhances the accuracy of ecosystem monitoring and forecasting, making a valuable contribution to environmental science and policy. The dissertation stands out for its originality, particularly in its application of remote sensing to monitor peatland vegetation in the context of climate change. The innovative exploration of vascular plant and moss interactions, their responses to climate variations, and their influence on the carbon balance further underscore the dissertation's novelty.

Warsaw, 21 February 2025