Author's response to Reviewers' comments #2

Manuscript reference number: **MS No.: egusphere-2025-292**

Manuscript title: Previous integrated or organic farming affects productivity and ecosystem N balance rather than fertilizer ^{15}N allocation to plants and soil, leaching, or gaseous emissions (NH_3, N_2O, and N_2)

Dear Editorial Team (egusphere), and Dear Eduardo Vázquez,

We sincerely appreciate your thorough and insightful review of our manuscript. We have carefully addressed your comments and made corresponding revisions to the manuscript. Thank you for giving us the opportunity to improve and resubmit our work. Please find below our detailed responses to each of your points.

Reviewer #2
1) I have reviewed the manuscript entitled "Previous integrated or organic farming affects productivity and ecosystem N balance rather than fertilizer ^{15}N allocation to plants and soil, leaching, or gaseous emissions (NH_3, N_2O, and N_2)." The study is highly relevant and has been very well executed using an elegant and robust methodological approach. Furthermore, the manuscript is clearly written, and the results are carefully analyzed and discussed. Therefore, I consider the manuscript worthy of publication, as it contributes significantly to our understanding of how agricultural management practices shape nitrogen cycling. ***Answer**: We sincerely thank you for taking the time to read and evaluate our manuscript. We are pleased that you consider the manuscript a significant contribution to the understanding of nitrogen cycling in relation to agricultural management practices.*
Despite a few minor corrections and suggestions (listed below), my main concern lies in the description and interpretation of the management practices, as well as how the experimental design may limit conclusions about the actual effects of those practices. ***Answer**: Thank you for your constructive feedback. We have incorporated the minor corrections and suggestions (see below, Minor Comments section) and addressed your primary concern as follows:* *We revised the Materials and Methods section (Lines 128-129) to better clarify the study's focus on the legacy effects of former management practices. Additionally, we added a new section in the Discussion (Section 4.5 Impact of Experimental Homogenization of management on Legacy Effects) that explicitly addresses how the experimental design limits the interpretation of management effects on nitrogen cycling.* *We also revised the Conclusion to reflect these limitations and present a more cautious interpretation of the results.*
As I understand it, although two contrasting management histories (organic farming, OF, and integrated farming, IF) are compared, both soils were managed identically during the experimental period (2021–2022). Thus, the study does not directly compare OF and IF, but rather the legacy effects of those systems. Although the authors acknowledge this limitation in the Materials and Methods (Lines 121–122) and in other parts of the manuscript (e.g., title, abstract [Lines 17–18], introduction [Lines 78–81], and conclusions [Line 605]), I believe a more explicit discussion is needed on how the homogenization of management during the experiment may have biased or limited the interpretation of OF and IF impacts on N cycling. ***Answer**: We sincerely appreciate this insightful observation. We agree that our study compares legacy effects of long-term organic (OF) and integrated (IF) management rather than their current practices, as both systems received identical management during the experimental period (2021-2022). While we did acknowledge this limitation in several manuscript sections (as noted), we agree*

that a more thorough discussion of its implications was needed. In the revised Discussion we added a section (4.5 Lines 647-661), we now explicitly address which reads as follows:

4.5 Impact of experimental homogenization of management on legacy effects

While the study aimed to investigate the legacy effects of organic farming (OF) and integrated farming (IF) systems on nitrogen cycling, it is important to recognize that both systems were managed identically during the experimental period (2021–2022). This homogenization of management practices, which included uniform crop rotations, tillage methods, and high cattle slurry application, may have limited the ability to draw direct comparisons between OF and IF systems in the experimental phase. As such, the observed differences in N cycling may be more reflective of past management legacies rather than current practices. Additionally, the timing of the crop assessments—green rye immediately after management homogenization and ryegrass two years later—could have further influenced the results, as the green rye was likely more influenced by previous management practices. Furthermore, the high rates of cattle slurry application, typical of IF systems but higher than those generally applied in OF systems, may have masked potential differences between the two systems in terms of nutrient cycling. Finally, the absence of legumes in the experimental crop rotations meant that their role in N cycling could only be inferred as a legacy effect, rather than being directly assessed. These factors should be considered when interpreting the findings, and future studies that include a more direct comparison of management systems with consistent practices and crop rotations, including legumes, would be valuable in further understanding the impacts of OF and IF on N cycling."

For instance, some observed differences across crop periods may result from the time elapsed since management homogenization. Green rye was evaluated immediately after management unification, while ryegrass was assessed two years later. Additionally, during the experimental phase, no inorganic N fertilizer was applied; only cattle slurry was used, and at high rates (136 kg N ha^{-1} in 2021 and 151 kg N ha^{-1} in 2022), similar to conventional IF practices, and notably higher than typical rates in organic systems. This might have masked key differences between the management systems. It is also important to note that legumes were not part of the rotation during the experimental phase, meaning their role can only be inferred as a legacy effect, rather than being directly assessed. I strongly recommend including a discussion on these aspects to help readers better understand the scope and limitations of the findings related to management system impacts.

***Answer**: Thank you for your comment. We agree with the reviewers and we have addressed this concern in the Discussion (Section 4.5, Lines 647-661), where we added a paragraph discussing the limitations of the experimental design, including the impact of homogenized management practices, the timing of crop evaluations, the high rates of cattle slurry applied, and the absence of legumes in the rotation. This discussion highlights how these factors may have influenced the observed results and how the study primarily reflects legacy effects rather than direct comparisons of organic and integrated farming practices.*

Minor Comments

- L16–17: The role of legumes in crop rotations is not directly evaluated in this study. I suggest removing or rephrasing this part of the abstract.

***Answer:** Thank for the comment. We agree that the role of legumes in crop rotation is not directly evaluated. The sentence is modified and reads as follows (lines 15-16):*

"Ecological intensification strategies in agriculture, including organic fertilization and diversified crop rotations, aim to reduce nitrogen (N) losses to the environment"

- L17–20: The lack of prior studies on this specific comparison does not alone justify the study. Consider strengthening the justification by emphasizing the relevance or potential impact of the findings.

Answer: *Thank you for this valuable suggestion. We agree that the justification for the study should go beyond identifying a knowledge gap. We have added the statement and specifically, we now emphasize that such insights are essential for optimizing nutrient use, reducing environmental impacts, and informing sustainable agricultural practices, which reads as follows (lines 19-21):*

"Understanding how these systems differ in their nitrogen dynamics is essential for improving nutrient management strategies, mitigating environmental impacts, and guiding sustainable agricultural practices."

- L54 and L93: Use consistent terminology, e.g., "symbiotic N_2 fixation."

Answer: *Thank you for pointing this out. We have revised the text to use the term "symbiotic N_2 fixation" consistently in both Line 54 and Line 93 to maintain clarity and uniformity. The sentences now read as follows (Line 55 and Line 98) .*

"….. associated fossil fuel consumption, due to their symbiotic N_2 fixing ability"

"However, nitrogen inputs from symbiotic N_2 fixation were…"

- L79–81: Clarify that the study evaluates the legacy effects of previous management practices, not their current application.

Answer: *Thank you for this important suggestion. We have revised the statement to clearly state that the study focuses on the legacy effects of past management practices rather than current management, which reads as follows (Lines 81-85):*

"Specifically, this study assesses how preceding management affects N balance and fertilizer N allocation by comparing sites with different management history. On the organic farming site (OF), legumes had been cultivated frequently, and external N was only occasionally added in the form of cattle slurry, whereas a combination of synthetic and organic fertilizers had been used on the integrated farming site (IF)."

- L88: What does "65618" refer to? Please clarify.

Answer: *Thank you for your comment. This is the postal code, which we have removed as coordinates are given. The sentence now reads (line 92):*

"The study was conducted in Selters, Germany (50°21'28.8"N, 8°15'47.4"E; elevation 310 m a.s.l.), where the average annual temperature and precipitation are 9.3 °C and 655 mm, respectively."

- L95: The phrase explaining integrated farming is redundant and unclear, revise for clarity.

Answer: *Thank you for the helpful suggestion. We have revised the sentence to remove redundancy and improve clarity. The updated sentence now reads (Lines 100-101):*

"The integrated farming (IF) site was managed using a combination of synthetic and organic fertilizers, with the aim of enhancing soil organic carbon (SOC) levels (Table A1)."

- L97–98: Was there any estimation of nitrogen inputs derived from symbiotic N_2 fixation during the previous management phase? Or at least the legume biomass production.

Answer: *Thank you for the comment. Unfortunately, no direct measurements or detailed records of symbiotic N_2 fixation or legume biomass production were available for the previous management phase. This limitation is now noted in the revised manuscript in the method section [Lines 98-99].*

"However, nitrogen inputs from symbiotic N_2 fixation were not measured prior to ours study. Therefore, these inputs are not included in the reported N input values (Khan et al., 2024, Table A1)."

-L121–122: I recommend explicitly noting the potential biases introduced by management homogenization here.

Answer: *Thank you for the insightful comment. We agree that management homogenization, while necessary to reduce confounding effects during the trial period, may introduce potential biases by masking legacy effects of previous land use. To address this, we have added the sentence to explicitly acknowledge this limitation. The sentence reads as follows (Lines 128-129):*

"However, we acknowledge that this management homogenization may mask legacy effects and introduce potential biases in interpreting differences between sites.**"**

- L133: Including photographs or diagrams of the experimental setup would improve reader understanding.

Answer: *Thank you very much. We have added details of the experimental design, including all parameters and the number of replicates for both organic and integrated farming.*

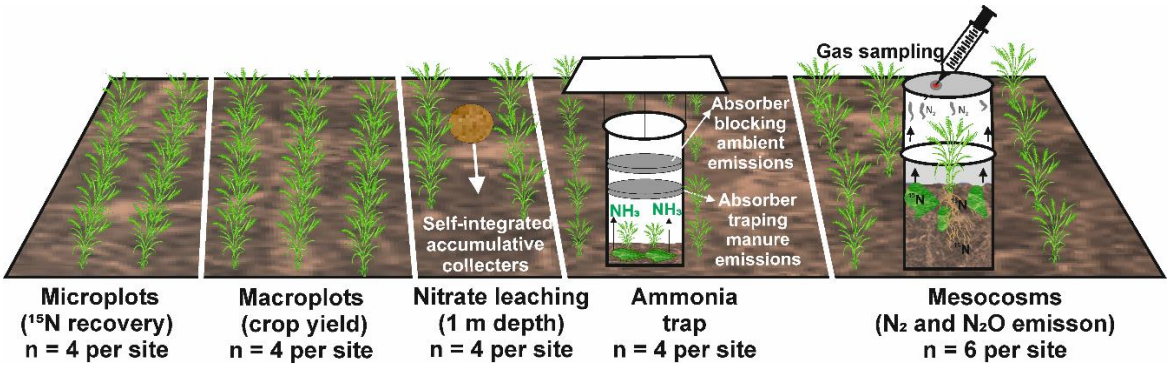

Figure 1: Schematic representation of the experimental units and associated measurements to assess the N balance and fertilizer N allocation. Microplots were established to assess ^{15}N recovery from soil and plants, macroplots to measure crop yield, self-integrated accumulative collectors (at 1-meter depth) to monitor nitrate

leaching, semi open chambers with absorbers for ammonia, and mesocosms to quantify gaseous N emissions (N_2 and N_2O) through gas sampling. The experiment included 4 replicates per site for microplots, macroplots, nitrate leaching, and ammonia traps, as well as 6 replicates per site for mesocosms.

- L353–354: "Silage maize" is mentioned twice, please correct.

Answer: *Thank you for pointing this out. We have broken the sentence into two parts to improve clarity and avoid redundancy. It now reads (Lines 370-372) :*

"In green rye, silage maize, and perennial ryegrass cultivation, 88, 74.8, and 151 kg N ha^{-1} cattle slurry were applied. For green rye and perennial ryegrass, cattle slurry was applied as top dressing in OF and IF (Fig. 4), while for silage maize, the slurry was incorporated."

- L402–403: Clarify "different", in what sense were they different? Which value was higher?

Answer: *Thank you for the comment. We clarified this in the following sentence (Lines 420-421):*

"In contrast, cumulative N_2 emissions were significantly different between OF and IF for the green rye and silage maize cultivation periods, with higher emissions under OF for green rye and under IF for silage maize."

- L402–413: Discuss how differences across managements and crops could be influenced by the time elapsed since management homogenization. Notably, differences were not observed in the last two crops, possibly due to longer homogenization time.

Answer: *Thank you for the suggestion. We have added a discussion addressing how differences between managements and crops may be influenced by the time since management homogenization. The discussion reads as follows (Lines 426–432):*

"One such factor could be the time elapsed since management homogenization. The same crop and fertilizer management were applied to both sites starting in October 2020, but legacy effects from the prior organic or integrated farming histories may still have influenced emissions, especially during the early cropping periods like green rye and silage maize. Over time, these legacy effects may have diminished, which could explain the lack of significant differences in N_2 emissions observed during the later crop (perennial ryegrass). Similarly, crop-specific factors may have interacted with site history. For example, lower plant N uptake on OF compared to IF in the green rye cultivation period may have led to a higher mineral N availability for microbial processes in the soil. Higher nitrate availability combined with the carbon sources of the organic fertilizer may have stimulated denitrification, particularly N_2 production (Senbayram et al., 2012; Samad et al., 2016). In contrast, by the time of the perennial ryegrass phase, soil microbial communities and N cycling dynamics may have adjusted to the unified management, leading to more comparable emissions. Alternatively, lower plant…."

- L407–411: The high nitrogen dose applied compared to the previously organic system, could have influenced the observed effects. This should be acknowledged.

Answer: *Thank you for this important point. We have acknowledged in the manuscript that the higher nitrogen dose applied to the previously organic system could have influenced the observed N_2 emissions and other effects, as follows (Lines 437–442):*

"Additionally, the relatively high N input applied during the experimental period compared to the historically low N input particularly under OF may have intensified the denitrification response, particularly in the early cropping periods. Thus, the complex interplay of historical management, time since management harmonization, crop type, and soil N cycle processes like plant N demand, the mineralization-immobilization cycle controlling N availability, and the effect of environmental conditions on the denitrification process (Butterbach-Bahl et al., 2002; Chen et al., 2019) led to variable N_2 emissions from both sites."

-L461: "There was no…"

Answer: *Thank you for pointing out. We have corrected the sentence which reads as follows (Line: 491).*

"There was no significant difference….."

-**L476–479**: This short paragraph should be more clearly connected to the previous one for better flow.

Answer: *Thank you for the suggestion. We have slightly revised and integrated the short paragraph to improve clarity and flow, which reads as follows (Lines 495-497):*

"Similarly, comparable NO_3^- leaching values on medium-texture soil were reported by Buchen-Tschiskale et al. (2023), who observed average nitrate leaching of 4% for trailing hose application or slurry injection of cattle slurry at an application rate of 71 kg N ha^{-1} for arable cropping systems."

- L479: As this is a hypothesis, rephrase the sentence to reflect speculative language.

Answer: *Thank you for the comment. We have revised the sentence to use more speculative language, now reading (Lines 498-499):*

"Thus, the medium soil texture of the sites in this study (Table 1) may have contributed to the observed low leaching rates."

- L599–601: Consider adding a recommendation for future studies to investigate both legacy and contemporary impacts of agricultural management practices.

Answer: *Thank you for the suggestion. We have added the recommendation for future studies which reads as follows (Lines 642-645):*

"To improve our understanding of long-term nutrient cycling, future studies should explicitly address both the legacy effects of past management practices and the direct impacts of current interventions. Such research will be crucial for disentangling time-dependent effects and guiding the design of resilient and sustainable farming systems."