23 November 2022

To the Reviewers:

We would like to thank the two reviewers of this manuscript for their careful consideration of our work and helpful suggestions for improvement. In the revised manuscript we are resubmitting, we have addressed every reviewer comment in the manner specified below. Note that all of our references to manuscript line number in this letter refer to the original manuscript submission.

***Reviewers' comments:***

***Reviewer #1:***

*This paper presents and discuss a new method for estimating Critical Nitrogen Dilution Curve for potatoes. Authors have pooled a large data set from different countries, Argentina, Canada and Belgium with different cultivars, and by using a Bayesian statistical approach they analysed the uncertainty of CNDC parameter determination and tried to infer on variation accross Genotype-Environment-Management conditions. The manuscript is very well written and very well organized. As said by authors the use of Bayesian method for CNDC uncertainty analysis is not fully originel, but they used this method very accurately and they proposed a new approach of partial pooling for a better analysis of G-E-M effects. So this manuscript is a very original and relevant contribution to crop N diagnosis problem. As I am not an expert in statistics, I cannot provide any comments on this part of the work. I guess that reviewing this manuscript by an expert in statistics and more particularly in Bayesian approach would be important. So our reviewing focus on agronomic and physiological interpretation.*

*The problem of "N dilution" process in crop having a strong "reserve" compartment as grain or tuber has been perfectly identified and discussed in introduction by authors. The "N dilution" model, originally developed on forage crop... only concern with plants in vegetative growth: producing only leaves and stems.... It was the reason why CNDC were limited in theory to flowering stage or just to early reproductive development when "grain biomass" was not too high... For taking into account grain or tuber growth... it should be necessary to have a two step N dilution (i) during vegetative growth ... with a given allometry coefficient "b1" reflecting the biomass allocation to "metabolic" and "structural" compartment...; and (ii) during grain or tuber filling reflecting C-N remobilisation from vegetative part and filling grain and tuber...with a more or less different value of allometry "b2" depending on the C-N ratio of grain or tuber accumulated. So "b2" being >> "b1" for grain and tuber accumulating preferentially starch (as for potatoes) or more or less = o r< b1 for grain accumulating preferentially proteins such as grain legumes... So it is clear that representing a single CNDC with a constant allometry "b"... while there is in fact a "break" in dilution process from "b1" to "b2".... is a problem... This problem is not so important statistically if b1 and b2 are not very differents... but becomes very importtant when they are very different as for potatoes. So the best way for showing this "break" in CNDC and to determine when this break occurs would be to represent CNDC in Log-Log term... So I suggest authors to illustrate that and to determine at which extent variation in the "break time" on the "crop biomass axis... would depend on G-E-M ? I think Giletto et al. (see their paper in EJA) have already well analysed this problem in Argentina? So it should be easy to this group of authors to deep this question. They can also refers to analogous work made on maize and wheat (ZHAO, B., ATA UL KARIM, S., T., LEMAIRE G.. DUAN, A., LIU, Z., GUO, Y., QIN, A., NING, D., LIU, Z., 2021. Exploring the source-sink relationship to quantify ear nitrogen accumulation in summer maize and winter wheat using critical nitrogen dilution curve. Field Crop Research, 274. https://doi.org/10.1016/j.fcr.2021.108332). By this way their comparison with "other crops" should be more complete because in their manuscript they compare CNDC of potatoes inclusing tuber filling process with other crops where CNDC was limited to "vegetative period"!!! So for potatoes, if G-E-M interaction has an impact on the onset of the change from b1 to b2 (as the start of tuber development)...as a consequence it should have an impact on the "average" CNDC fitted with a constant "b"!!! So it should be important to verify this hypothesis: has G-E-M an effect of both b1 or b2 separatly...? or has G-E-M has only an effect of the onset of change from b1 to b2 ? That would be a more fundamental question?*

*So my conclusion is that this excellent manuscript should be accepted for publication... But I suggest authors to improve its scientific value by adding some informations on "b1" and b2" for being able to discuss more strongly the hypothesis above.*

* We agree that the proposed approach suggestion by Reviewer 1 is an extremely valuable and a key question for future research to consider; however, we find that this analysis would fall outside the scope of this manuscript. Given the suggestion from Reviewer 2 to focus and reduce the length of this manuscript, we have limited our discussion of this suggestion from Reviewer 1 to a paragraph in a new Section 4.1.4. We look forward to addressing this research question in a subsequent publication.

*Details:*

* *Line 67: No, NNI is very sensible to any fertilization management...as it detect any effect on plant N nutrition status....*
* Change made here as suggested to clarify and correct statement.
* *Line 76: No, b is the ration between relative rate of %N decline (d%N)/(%N)dt and the relative rate of biomass accumulation dW/Wdt.... that is different of the rate of %N decline (d(%N)/dt*
* Change made here as suggested to clarify and correct statement.
* *Line 92: add "Acceleration" of dilution....*
* Change made here as suggested to clarify and correct statement.
* *Line 189: "reduce" is repeated two time....*
* Change made here as suggested to clarify and correct statement.

***Reviewer #2:***

*This study advances on the use of Bayesian hierarchical frameworks to develop critical N dilution curves introducing a partial pooling approach through random components. This could represent a useful alternative for comparing CNDCs across G × E x M conditions. Moreover, it could be further extended for developing critical N dilution curves of potato but also potentially of other crops. I found it interesting to read and review, which makes me think it would be very relevant for EJA journal.*

*I also found the paper excessively (and unnecessary) long in several sections, so my first main suggestion is to reduce the length of the manuscript reorganizing paragraphs and ideas. I identified below several sections and paragraphs where this could be done. Similarly, I would suggest reducing the number of figures considering the complexity of the methodology and number of panels. I believe the paper will have more impact if ideas (including figures) are more succinct.*

* We have addressed this main comment as described in the specific comments below.

*My second main suggestion is on one of the methodologies used and (at some point) recommended to evaluate uncertainty in the CNDC. This should be addressed before publication. See below specific comments related to this in LI423 & LI524.*

* We have addressed this main comment as described in the specific comments below.

*Specific comments:*

* *LI28: "was attributed to variation" is a statement that cannot be confirmed with this analysis due to the lack of factorial combinations of the G (maturity classes) x M (plant density) at each site (i.e., E). It would be better to claim "was hypothesized".*
* Change made here as suggested to clarify and correct statement.
* *LI59-62: no need to go back to the rate-response approach as the paper is not about it, could be removed.*
* Change made here as suggested to remove this reference.
* *LI82-114: I think this section can be largely summarized in a single paragraph. This paper is more about the methodology of fitting and quantifying uncertainty in CNDC, so only a brief overview of the dilution theory + use in potato is needed.*
* This paper is focused both on the methodology of fitting and quantifying uncertainty in CNDC as well as understanding the particular mechanisms of N dilution for potato across G x E x M factors. The comments by Reviewer 1 highlight the need for this key background information in the Introduction section, with respect to understanding the mechanism of dilution across G x E x M factors. Therefore, we have not made this change as suggested by Reviewer 2.
* *LI117-118: this is a good example of the type of "expensive" writing used along the paper that makes it hard to read... "Previous development of CNDCs for potato has been conducted using a non-uniform set of statistical methods and with limited quantification of uncertainty in either the range of plausible %Nc values or the fitted parameter values themselves" can be rephrased with the same meaning by "Previous CNDCs for potato have been developed with different statistical methods and limited quantification of their uncertainty." Simplifying sentences would not only reduce the length of the paper but also increase the impact of each message. I would recommend considering this point when re-organizing ideas.*
* Change made here as suggested to simplify this statement.
* *LI126-127 & 133-134: These three paragraphs can be combined into one, no need for break lines*
* Change made here as suggested to consolidate paragraphs.
* *LI141: "linear plateau was designed to discriminate against" Not clear. In the paragraph above, it was claimed that the linear plateau cannot address these exact two points?*
* Change made here as suggested to delete this phrase to clarify and correct statement.
* *LI166-192: Should be combined into a single paragraph.*
* Change made here as suggested to consolidate paragraphs.
* *Tables 2, 3 and 4 can be combined into one. Table 1 can be combined within Table 2-3-4 and Table 5. Will then need only two tables.*
* Change made here to combine Tables 2, 3, and 4 into a new Table 2. Thank you for the excellent suggestion for simplification! It is not clear, however, how further combining Table 1, Table 2-3-4, and Table 5 can be done to simplify data presentation; therefore, we have kept three tables, instead of two as suggested.
* *LI273: Is confusing referring here as "experimental" data, considering the first set of sites were classified as "Experimental" vs "Prev. published". Can consider the use of terms here.*
* Change made here as suggested to clarify and correct statement.
* *Section 2.1.2. I suggest using the Tables to report detailed information of these experiments, considering detailed information has been already reported in previous publications. Can use text to report only data that is not in Table, such as location site.*
* Changes have been made to Section 2.1.2. and Table 5 as suggested. Another excellent suggestion by the reviewer for simplification!
* *LI336-347: Is this paragraph needed? Most of this has been mentioned in the introduction. Same with figure 1, I think it is not a critical figure of the manuscript and could be removed?*
* Changes have been made as suggested to remove this paragraph and Figure 1.
* *LI391: What about replicates? How they were treated in the model?*
* Changes have been made to clarify that models were fit using treatment-level means and replicates were not included in model hierarchical structure. Data at replicate level was not available for all experimental trials.
* *LI395: Convergence checks? I assume they were conducted, please add.*
* Change made here to clarify and describe methods used.
* *LI397: "biologically or physically impossible predictions" not sure what that means.*
* Change made here as suggested to clarify and explain statement.
* *LI401: What about priors for the random effects variances? it looks to me that the priors from Table 6 are very informative and could constrains parameters to a short range of variation, but according to the results, there seems less restriction on priors for the random effects? This can be tested through a sensitivity analysis with less informative prior distributions.*
* Change made here as suggested to clarify and explain the need for informative priors for random effect variances, specifically related to ensuring joint prior predictive distribution was biologically and physically plausible as well due to propagation of variance in joint prior predictive distribution in hierarchical models.
* *LI405: This is great addition and authors should be congratulated for this.*
* We thank the reviewer for their recognition of this!
* *LI423: I am not sure I understood this, but this might not be correct. Did you fit a new model (and frequentist?) to the data estimated by the parameters of the 90% credible limits of the Bayesian curves? There is circularity in this approach, and not sure why it was done.*
* The reviewer’s interpretation of our method as described above is correct. This step was done to create parameterized estimates of the 90% credible region. While the 90% credible region as determined from the posterior distribution of the fitted Bayesian hierarchical model is most appropriate to use, it is very difficult to communicate the 90% credible region in this manner (i.e., non-parameterized, requires very large model object). Without parameterized estimates of the upper and lower boundary of the 90% credible region, then it is very difficult to propagate uncertainty in %Nc to derivative computations (i.e., calculating NNI). This is discussed in Section 4.3. of the original manuscript. Additional description of the methods has been added to this section.
* *LI436: The definition of delta%Nc is not clear. This is a very complex and long sentence, but critical to understand the paper. What is "the difference between the 0.50 quantile for %Nc and the various methods to quantify uncertainty (i.e., 90% credible region for %Nc, CNDCup & CNDClo, and estimates of credible region for %Nc using 90% credible interval for parameters a and b)"?*
* Changes were made here to clarify the definition of ∆%Nc in Sections 2.3.2., 2.3.3., and 2.3.4.
* *LI448-455: Another case of "expensive" writing.*
* Change made here as suggested to simply this statement.
* *LI479: Figure 1 again? Check numbering of all figures.*
* It appears that numbering of figures was affected by an error in the manuscript upload process. This has been corrected for the revised manuscript, and numbering of all figures and tables has been checked as well as updated based on consolidation and removal of tables and figures as suggested by the reviewer.
* *LI525: Figure 4: why 15 individual draws are represented in red? Please clarify this analysis.*
* Change has been made here to remove Figure 4b and references to this figure, for the purpose of conciseness and clarity.
* *LI542: Why is quite uninformative? I do not agree with this analysis of comparing methods to measure uncertainty of the CNDC because there are basically different things being compared. If there is interest in quantifying uncertainty of the %Nc, credible intervals for the %Nc should be analysed. If there is interest in quantifying uncertainty in the parameters of the CNDC, credible intervals for a and b parameters. This is the advantage of the Bayesian framework through the obtained posterior distributions.*
* Change made to clarify the approach used and conclusion of findings of this approach, following from changes made in Sections 2.3.2. and 2.3.3.
* *LI612: If there were significant differences, wasn't it also biased? Can claim that was less biased.*
* Change made here as suggested to clarify and correct statement.