

For review article

Response to Reviewer 2 Comments

1. Summary

I extend my sincerest gratitude for the time and effort you have dedicated to reviewing our manuscript. Your insights are invaluable to us. Below, you will find a comprehensive list of our responses to your comments, along with the respective revisions and corrections meticulously highlighted in the track changes feature within the re-submitted document.

We have taken great care to address each point with the aim of enhancing the clarity, accuracy, and overall quality of our work. We trust that these amendments will meet with your approval and bring us closer to the publication of our research.

Thank you once again for your invaluable feedback.

2. Questions for General Evaluation	Reviewer's Evaluation	Response and Revisions
Is the work a significant contribution to the field?		<i>[Please give your response if necessary. Or you can also give your corresponding response in the point-by-point response letter. The same as below]</i>
Is the work well organized and comprehensively described?		
Is the work scientifically sound and not misleading?		
Are there appropriate and adequate references to related and previous work?		
Is the English used correct and readable?		

3. Point-by-point response to Comments and Suggestions for Authors

Comments 1: *The manuscript "Identification of the Pyrus CDPK Pan-genomic Family and Analysis of Its Response to Venturia nashicola in Pear" describes a relevant study but must be thoroughly improved before being publishable. In title, "Pyrus" is unnecessary because the "Pear" term is included. Then, when Pyrus is mentioned, correction to Pyrus spp. must be done because various species of the genus Pyrus were analyzed.*

Response 1: We sincerely thank the reviewer for this constructive feedback. We have revised the manuscript accordingly to address these points:

1.Title: We have removed the redundant "Pyrus" from the beginning of the title as suggested. The revised title now reads: "Identification of the CDPK Gene Family in Pear

(Pyrus spp.) and Analysis of Its Response to Venturia nashicola". We believe this new title is more concise and accurate.

2.Pyrus spp. nomenclature: Throughout the manuscript, we have carefully reviewed all instances where the genus Pyrus is mentioned in the context of our multi-species analysis. We have corrected these to the accurate designation "Pyrus spp." to properly reflect that our study encompasses multiple species within the genus, as rightly pointed out by the reviewer.

We appreciate these valuable suggestions, which have significantly improved the precision and clarity of our manuscript.

Comments 2: *In Introduction, no general or specific objectives are explicit. Instead, a brief presentation of the overall manuscript is given in the last paragraph. The aims of the study must be clearly presented, especially considering that some experimental research was done in tobacco, not in pear. Also, specific objectives related to in silico, in vivo and in vitro studies must be presented. Indeed, there are some experiments with pear calli described in Results but nor mentioned in Materials and Methods.*

Response 2: We sincerely thank the reviewer for these critical comments regarding the clarity and completeness of our study objectives and methodological descriptions. We have thoroughly revised the manuscript to address these concerns as follows:

1.Explicit research objectives in Introduction: We have added a dedicated paragraph at the end of the Introduction section that clearly states both the general aim and specific objectives of our study. These now explicitly encompass our multi-level approach including: In silico analyses (pan-genome construction, phylogenetic analysis, etc.)、 In vitro experiments with pear calli、 In vivo validation in tobacco model system

2.Methodological completeness: We have expanded the Materials and Methods section to include detailed descriptions of: The pear calli culture protocols

We believe these significant revisions have greatly improved the transparency and completeness of our manuscript by clearly articulating our research objectives and ensuring all described methodologies are properly documented in the relevant sections. Thank you for these valuable suggestions that have substantially enhanced our manuscript.

Comments 3: *Particularly in Materials and Methods, the assessment of normality distribution ought to be done to properly apply parametric statistics such as the t-test.*

Response 3: We thank the reviewer for this important methodological suggestion. We fully agree that testing for normality is an essential step before applying parametric statistical tests. Accordingly, we have revised the Materials and Methods section to explicitly state that the normality of the data distribution was assessed using the Shapiro-Wilk test prior to conducting t-tests. This addition ensures that our statistical analyses are properly validated and meet the necessary assumptions for parametric testing. We appreciate this comment, which has helped improve the rigor of our statistical methodology.

Comments 4: *In Results, section 3.4 actually corresponds to Discussion. Legends to Tables and Figures must be improved to achieve self-contained meaning.*

Response 4: We sincerely thank the reviewer for these valuable observations, which have significantly improved the organization and clarity of our manuscript.

1.Section 3.4 Reorganization: We agree with the reviewer that the content of the original section 3.4 was interpretive in nature and more suitable for the Discussion. Consequently,

we have moved this section to the Discussion part of the manuscript. The Results section now focuses solely on the objective presentation of our findings.

2.Improvement of Figure and Table Legends: We have thoroughly revised all legends for our figures and tables. Each legend now provides a more comprehensive and self-contained summary, including essential details on the experimental context, methodology (where pertinent), and a clear interpretation of the key findings presented. We believe these revisions have enhanced the logical flow of our manuscript and the clarity of our data presentation. Thank you for these insightful suggestions.

Comments 5: *Discussion and eventually Conclusions (absent in present version) sections must be redacted to respond the also absent general and specific objectives, which in fact must be added in Introduction, as previously indicated.*

Response 5: We sincerely thank the reviewer for this critical comment regarding the structure and focus of our manuscript. We have carefully revised the paper to address these important concerns as follows:

1.Introduction Revision: We have added a dedicated paragraph at the end of the Introduction section that clearly states the general and specific objectives of our study, as previously recommended.

2.Addition of a Conclusions Section: We have created a new, standalone Conclusions section that summarizes the main findings of our study, emphasizes their scientific and practical significance.

We believe these significant revisions have greatly improved the logical flow, clarity, and scholarly rigor of our manuscript. The study's aims and outcomes are now presented with greater transparency and structure. We are grateful for these suggestions, which have substantially enhanced the quality of our work.

Comments 6: *English must be deeply revised. Words as varieties are (lane 213), where a (lane 306), as examples, must be revised. Additionally, there are lost and spare spaces among word along the entire text.*

Response 6: We sincerely thank the reviewer for carefully reading our manuscript and pointing out these language and formatting issues. We have thoroughly revised the entire manuscript to address these concerns:

1.We have corrected the specific errors mentioned ('wherea' to 'whereas') and conducted a comprehensive spell-check throughout the document to identify and correct similar errors.

2.We have carefully reviewed the entire text to ensure proper spacing between words and sentences, removing any unnecessary spaces and adding missing spaces where needed.

3.Additionally, we have engaged a professional English editing service to perform a complete language revision of the manuscript to ensure proper grammar, syntax, and academic style throughout.

We appreciate the reviewer's attention to these important details, which has significantly improved the overall quality and readability of our manuscript.

4. Response to Comments on the Quality of English Language

Point 1: Please deeply revise.

Response 1: We sincerely thank the reviewer for this critical comment regarding the English language quality of our manuscript. We fully acknowledge that the initial submission required significant linguistic improvement. In response to this comment, we

have undertaken a comprehensive and deep revision of the entire manuscript to ensure the clarity, accuracy, and professionalism of the English language. To achieve this, we have taken the following concrete steps:

1. Professional Editing Service: The manuscript has been thoroughly edited and polished by a professional scientific editing service .

2. Meticulous Author Review: Following the professional editing, we, the authors, have carefully reviewed the entire document again. We have double-checked all technical terms, gene nomenclature (e.g., *Pyrus spp.*, *Venturia nashicola*), and scientific expressions to ensure they are precise and consistent throughout the text.

3. Focus on Clarity and Flow: We have paid particular attention to improving sentence structure, grammar, punctuation, and overall narrative flow. We have rephrased awkward constructions, eliminated ambiguities, and ensured that the logical progression of ideas is clear and easy to follow.

We believe that these extensive revisions have significantly enhanced the readability and overall quality of the manuscript. We hope that the current version now meets the high standards required for publication in your esteemed journal. We thank the editor and reviewer for the opportunity to improve our work.

5. Additional clarifications

[Here, mention any other clarifications you would like to provide to the journal editor/reviewer.]