

**Title: "Screening of transcription factors interaction with *LDOX* promoter to regulate the anthocyanin biosynthesis in peach"**

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### **Strengths of the Manuscript**

- The introduction provides a thorough background on the topic, covering the importance of peach, anthocyanin biosynthesis, and the role of transcription factors.
- The introduction cites relevant literature to support the research, demonstrating a good understanding of the field.
- The introduction clearly states the research question and objectives, providing a direction for the study.
- The materials and method section provides a detailed description of the methodologies used, including plant materials, Y1H screening, NGS analysis, and bioinformatics analysis.
- This section also specifies the reagents and equipment used, which can help with reproducibility.
- The data analysis methods used, including phylogenetic analysis and qRT-PCR is also enough for precise results.
- The results section provides a comprehensive analysis of the interaction between MYB1R1 and the LDOX promoter, including Y1H assay, NGS analysis, and bioinformatics analysis. This section also provides a detailed analysis of the MYB1R1 protein, including its physicochemical properties, secondary and tertiary structure, and potential interacting proteins.
- Gene expression analysis includes MYB1R1 in various tissues, providing insights into its potential role in regulating anthocyanin synthesis.
- The discussion provides a thorough review of the relevant literature on anthocyanin synthesis, LDOX, and MYB transcription factors.
- The authors clearly hypothesize that MYB1R1 interacts with the LDOX promoter to regulate anthocyanin synthesis in peach petals.
- The discussion effectively integrates the experimental results with the existing literature to support the authors' conclusions.
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### **Weaknesses of the manuscript**

- The introduction is quite long and could be condensed to focus on the most essential information. Some sentences are dense and could be broken up for easier reading. The introduction could benefit from smoother transitions between paragraphs and ideas.
- The methodology section is quite long and could be condensed to focus on the most essential information. Some information, such as the description of the kits and reagents used, is repetitive and could be summarized.
- Some methods, such as the choice of specific software or parameters, could be justified with more detail.

- The section includes a large amount of data, which could be overwhelming for readers. Consider highlighting the most important findings.
- The discussion is quite long and could be condensed to focus on the most essential points. Some sentences repeat information that has already been mentioned, which could be avoided to make the discussion more concise.
- Some statements, such as the role of LDOX promoter DNA methylation 450-451 are speculative and could be further investigated.

### **Suggestions for improvement**

- Identify the most critical information and focus on those points to make the introduction more concise. Use transitional phrases to connect ideas between paragraphs and improve the flow of the text. Consider defining key terms, such as "variegation" and "anthocyanin biosynthesis," to make the text more accessible to readers without a strong background in the field.
- Summarize repetitive information: Consider summarizing repetitive information, such as the description of kits and reagents used, to make the section more concise.
- Consider providing more justification for methodological choices, such as the choice of specific software or parameters, to make the section more transparent.
- Consider organizing the section into subheadings, such as "Identification of MYB1R1 as a potential regulator of LDOX," "Analysis of MYB1R1 protein properties," and "Gene expression analysis of MYB1R1."
- Consider highlighting the most important findings, such as the interaction between MYB1R1 and the LDOX promoter, and the potential role of MYB1R1 in regulating anthocyanin synthesis.
- Consider organizing the discussion into subheadings, such as "Regulation of Anthocyanin Synthesis," "Role of MYB1R1," and "Future Directions."
- Emphasize the most important findings and avoid repetitive information.
- Clearly outline the future research directions, such as investigating the role of LDOX promoter DNA methylation.

### **Overall comments**

Overall, the introduction provides a comprehensive overview of the research topic and sets the stage for the study. With some revisions, it could be even more effective in engaging readers and conveying the research question and objectives.

The Materials & Methods section provides a detailed description of the methodologies used, which can help with reproducibility. With some revisions, it could be even more effective in conveying the research methods and results.

The Results section provides a comprehensive analysis of the interaction between MYB1R1 and the LDOX promoter, and its potential role in regulating anthocyanin synthesis. With some revisions, it could be even more effective in conveying the research findings.

The discussion provides a comprehensive overview of the research findings and their implications for understanding anthocyanin synthesis in peach petals. With some revisions, it could be even more effective in conveying the research conclusions.

A handwritten signature in black ink, appearing to read 'Shazia Sakhi', with a stylized flourish at the end.

**Dr. Shazia Sakhi**

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of Swat