



SDI Review Form 1.6

Journal Name:	Current Journal of Applied Science and Technology
Manuscript Number:	Ms_CJAST_42299
Title of the Manuscript:	Molecular Detection of Antimicrobial Peptide Genes and Identification of Novel Antifungal Compounds of Bacillus sp. against Fusarium oxysporum f. sp cubense Causing Wilt of Banana
Type of the Article	Original Research Article

General guideline for Peer Review process:

This journal's peer review policy states that **NO** manuscript should be rejected only on the basis of '**lack of Novelty**', provided the manuscript is scientifically robust and technically sound. To know the complete guideline for Peer Review process, reviewers are requested to visit this link:

(<http://www.sciencedomain.org/page.php?id=sdi-general-editorial-policy#Peer-Review-Guideline>)



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PART 1: Review Comments

Reviewer's comment	Author's response to Reviewer's comment	Author's comment (if agreed with reviewer, correct the manuscript and highlight that part in the manuscript. It is mandatory that authors should write his/her feedback here)
Compulsory REVISION comments	Abstract Aims is clear and concise, abstract is with relevant and informative matches with the rest of the article. Line 10: Please insert "biocontrol" before "strategy" Line 12: Please delete "of which" and insert "20 strains," What is missing: Design of the experiment Introduction Literature is brief and sufficient to cover the information about the study conducted. Author clearly mentioned research question and knows about the subject under investigation based on the literature reviewed. Line 25: Please delete "important fruit crops" before commercially. Line 27: Please delete "It is a vital fruit crop and become the largest cultivation in India". Vital is equal to important which is stated in then previous sentence. Line 29: Please insert a "," after "Hans." as the sentence is continued. Lines 30-32: Please check the rewritten sentence as pasted below for these lines "Application of fungicides and resistant cultivars are the only management techniques available for controlling fusarium wilt in Banana (Saravanan et al., 2003; Geetha et al., 2005) Line 32: Please replace "Moreover," with "The" Line 33: Please replace "the need" with "look" and replace "alternating" with "alternative" Line 34: Please insert "s" to "method" to make "methods" Line 34: Please replace "contain as several as" with "carries" and insert "that are in use for control of soil born plant pathogens." after "... 2014) by deleting "." Please expand the term "AMP" , will it mean amplified or something else? Line 36: Please replace "Chung et al.," with "Chung et al.," Line 37-38: Please delete the entire sentence "Indication of quite a while." Line 38-40: Please insert "through bacterial cell culture" after "metabolites" and before "with". Please replace "one determinant of their capacity to control plant diseases" with "proven way to control plant pathogens" Line 41: Please delete " were investigated" and replace "its" with "their" as well as add "s" to "effect" to make "effects". Line 42-43: Please replace "In this perspective" with "Therefore" and replace "explored" with "considered". Line 44: Please replace "An objective" with "The objectives" and delete "s" from "includes" to make it "include ," Weakness: 1) Important fruit crop, but missing its value of importance as a fruit crop to human nutrition point of view or economic point (by export or domestic consumption) of view for getting income to the country. Production constraint in terms of production quantity and income statistics were missing to represent the amount of economic loss due to wilt disease. 2) Poor sentence construction skills, needs improvement Methods Subject selection is clear, study methods are reliable and valid., but methods are described in an organised manner to be consistent with the procedure followed as referenced. There is no flow with clear definition of variables and measures. Line 48: Please (Foc) Line 49: Please specify the tissue of the plant part used in banana for isolation of pathogen. Line 49-51: Please specify the methods followed with references and with brief description of the procedure Line 53: The sentence best fit in line 49. But requires a reference to justify the pathogen isolation procedure Line 54: Please indicate the sample from which the genomic DNA was isolated following the reference methodology of Knapp and Candle, 1996. Line 54- 55: Please delete "gene was characterized with" and insert "intervening sequence specific" before "BCF1" Line 56: Please insert "were used to confirm bacterial strain as Bacillus species" after "primer pair" Suggested Correction: Please combine paragraphs under line 48 and line 52. Delete the subhead of line 48	As per the reviewer comments line numbers 10, 12, 25, 27, 29, 30, 32, 33, 34 36, 38, 41, 42 and 44 has been carried out. The sentence have been corrected as per reviewer suggestions. Line numbers 48 to 55 have been made. Results, discussion and conclusion has been rewritten as per reviewer suggestions.



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	as it is repeated again in line 52. First describe about sample selection, pathogen isolation from sample, then culturing the pathogen, then sequencing to identify the genes coding for antifungal activities and to confirm bacterial culture with antibiotic activity. Also please mention the target DNA sequence length in base pairs, whether it is 546bp or more than that. Then describe about the antifungal activity for pathogen control and antibiotic compounds effective against it along with GCMS analysis. Line 93: Why "B18" was only chosen as most effective strain of <i>Bacillus cereus</i>? when there are other strains showing positive response, how did you select, is it based on the retention time of antifungal compounds through GCMS analysis/chromatograph fig 2 Strength: 1) Excellent methodology and procedures were chosen for the investigation. 2) Line 61-86: Series of PCR primers and conditions to make multiple copies of the bacterial gene that can synthesize specific antibiotic with literature support. 3) The research is done with suitable methods which are technically standard. Major Weaknesses 1) Pathogen isolation, incubation methods were not described in the appropriate section and with standard references. Over all, methods need thorough revision 2) Incubation temperature were mentioned as 27+ or -2 degrees in line 50 for 5 days, line 58 for 7 days and line 88 for 7 days at 28+ or -2 degrees. Please specify the incubation time, purpose, and the organism. 3) Line 87-91: The source of collection of crude metabolites, the purpose and procedure is missing though two recent references were cited. Results and Discussion The data well controlled and not robust, provided relevant and current references during discussion. Results and discussions were based on actual facts and figures. There is no limitation or bias in statements observed. Statistical analyses is required for this paper for table 1 where results are compared to a control to check for the effectiveness of the strain for antifungal activity. Suggested Corrections Line 103: Please specify the pathogen, whether it is fungi or bacteria. or the main "FOC". Please correct the errors of incubation, the term pathogen by indicating bacteria, foc , or fungi, as in banana there is bacterial wilt and fungal wilt. The investigation title is fungal wilt caused by FOC. Line 104: Please correct the error "20 27" in incubation temperature. Line 105-107: The sentence is wrong in subject "the pathogen was also molecular characterized through PCR sequencing the 18S–28S rRNA" without any description of the procedure of sequencing and method and the organism. Line 110: What is control in the investigation, as information about the control is not specified anywhere in methods or discussion Line 110: replace "sequenced" with "sequence" Line 111: Please replace "so" with "to be" Line 112: Please insert "407 mm2 area of" before "the" and replace "F. oxysporium f.sp cubens in vitro by 407.00 mm2" with "FOC invitro" Line 115: The number in the parenthesis "(5)" if indicates number of amplified genes, then rewrite the sentence as " Strains B18 and B4 had five AMP genes responsible for biosynthesis of antibiotic genes." But the table 2 is displaying more than these two strains like B2,B5,B7,B8,B17,B19., besides B18 and B4. Line 114: Iturin C is not prevalent, showing negative response, Please check the table again and write the results. Line 115 to till the end of the discussion: Please see the comment 2 of the weakness of this section Tables and Figures: Table 1: Title should be above the contents of the table, missing information on statistical significance and for symbols used "a to j" and "*" as well as for the values in parenthesis in column four. Table 3: Information about Rt is missing Figure 1: Quantity of sample loaded in each well, Standard ladder name and size is missing in the labelled M lane for the reference to compare the "546 bp" matching size of sample on the ladder. It is a very small size fragment should appear above five markers of 100bp ladder. The image is of PCR amplified 16 S r RNA bands were matching with the region on the ladder between lightly stained fifth and brightly stained sixth bright marker. If it is 100 bp ladder, then it is correct. Weaknesses 1) The subject is introduced for the first time without any detailed description of the methods or procedure	
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	followed and the organism (fungal/bacterial/banana) For Example in line 106 "the pathogen was also molecular characterized through PCR sequencing the 18S–28S rRNA" sequenced 2) The first fifteen lines are related to results obtained and are not completely described in a systematic way connecting the concepts of cell culture, identification of antibiotics and the purpose of PCR sequencing for amplification of bacterial fragments with amplicons of antifungal activity and GCMS technique for identification of chromatographic peaks with best retention time (Rt) of effective strain with antifungal compounds. Majority of discussion is filled with previous reports of similar findings and explanations provided for the antibiotic activity. 3) The discussion is lacking information on results presented, comparisons to previous the results obtained for each and every distinct result obtained through present investigation and there were no suggested future lines of research for improvement or continuation. 4) Needs thorough revision of the manuscript as the research was conducted with lots of scientific effort and knowledge. 5)An appropriate statistical analyses were carried out , but not presented for tables as necessary, neither discussed in the text. Conclusion: Conclusion require more input with results obtained. It covers aim and some results. Line 142: Please insert "Management of " before "Fusarium" and delete "foc" and delete "is causing Fusarium wilt of banana and its". As it is know fact, not a conclusion. Line 143: Please replace "Management is difficult to realize which usually relies of" with usually relies on". Line 144: Please insert "conducted" before "to" and insert "role and efficacy of" after "the" Line 145-147: Please incorporate the suggested rewritten sentence without changing the meaning of the original text written by author and was pasted below. "GC/MS analysis of crude metabolite extract of the effective Bacillus cereus strain B18 revealed active antifungal compounds responsible for inhibition of foc. Results showed amplified (AMP) genes from all Bacillus spp. Therefore the antifungal and antibacterial compounds identified through present investigation would help to control the FOC pathogen in Banana. Over All 1) please avoid repetition of subheads, terms or sentences or information in the entire text including conclusion. 2) Please add future lines of research to the conclusion. Also please add the main conclusion of the investigation, as your investigation involves use of biological activity of metabolites to control pathogens as one of the biological control methods suggested for controlling plant pathogens. References References are relevant, recent including key studies of investigation conducted and cited correctly in both text and reference section. Line 149: Please follow the staandard rule for the reference: Ashwini Kumar should be used as "Kumar, A" if there is no last name. Please follow the same rule for correcting author names in line 198 Line 158,160,186, 189, 202,209,212: No need to use "and" between names of authors before the title of the reference, especially for the science domain publications. Peptide genes are in the title, four antifungal compounds are presented in one table, but not the peptide sequence or genes discussed in relation to the disease and control OVER ALL : NEEDS THOROUGH REVISION AND ORGANISATION OF THE CONTENT WITH ADDITIONAL INPUT OF DISCUSSION AS NECESSARY	
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Minor REVISION comments		
Optional/General comments		