

Review Form 1.6

Journal Name:	Journal of Advances in Microbiology
Manuscript Number:	Ms_JAMB_85810
Title of the Manuscript:	Screening of Selected Soil Environments of Jos North Local Government Area for Lipase-Producing Fungi
Type of the 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:

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PART 1: Review Comments

	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	<p>1- Regarding malt extract agar, I did not find the composition of the medium in item 2.3. In addition, I recommend the use of PDA (Potato-dextrose-agar) medium for fungal growth.</p> <p>In this same item, to obtain a pure culture, the ideal would be to perform a monosporic isolation. Here is the methodology: Nelson, P.E.; Touson, T.A.; Marasas, W.F.O. Fusarium Species, an. Illustrated Manual for Identification; Pennsylvania State University Press: University Park, PA, USA, 1983.</p> <p>If you haven't done this isolation, I recommend deleting the word "pure" and replacing it with something like: ...to obtain morphologically separate cultures.</p> <p>2- In item 2.4, in addition to these tests, I would add the rhodamine B test, considered the most sensitive to determine the presence of true lipase. The substrate used is olive oil. Rhodamine (fluorescent compound) in the presence of free fatty acid, if complexes and under UV light, releases fluorescence, indicating the substrate degradation zone. In this work there is a description of this medium: SANTOS, F.C. DOS ; DE CASTRO, F.F. ; APOLONIO, T.M. ; YOSHIDA, L. ; MARTIM, D.B. ; TESSMANN, D.J. ; BARBOSA-TESSMANN, I.P. . Research Article Isolation, diversity, and biotechnological potential of maize (Zea mays) grains bacteria. GENETICS AND MOLECULAR RESEARCH, v. 18, p. GMR18320, 2019</p> <p>For other tests, such as tributyrin for non-specific esterase activities, I recommend this one: GOPINATH, S. C. B., et al. Strategies to Characterize Fungal Lipases for Applications in Medicine and Dairy Industry. BioMed Research International, 2013</p> <p>3- In item 2.5, during the days of incubation of the fungus, were they incubated under agitation? Was there also a 12-hour photoperiod?</p> <p>About the presence or absence of agitation, this is related to greater aeration of the growth environment and greater contact of the fungus with the substrates and other elements of the medium. This can make the fungus grow more or less, we just can't compare mycelium size with enzyme production. Agitation can help the fungus to release the enzyme into the medium, or not.</p> <p>Photoperiod is related to mimicking a natural situation in the laboratory, providing a condition that is similar to that found in nature, where there is basically light for 12 hours.</p> <p>4- When working with enzymatic activity, there are many details to note:</p> <ul style="list-style-type: none"> - The incubation environment is one of them. For 200ml of medium, the ideal would be to inoculate in 500ml flasks. These same 200ml in 250ml flasks, leaves little room for the fungi to "breathe". My recommendation (to spend less) would be 25ml of medium in 125ml bottles, or 50ml of medium in 250ml bottles. In addition, I recommend testing the addition of some substrate for the enzyme in the culture medium. The medium used in this work does not present any carbon source. I will leave an article for you to see the influence of a carbon source in the culture medium on enzyme production. 	

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	<p>DOS SANTOS, FABIANE CRISTINA; DE OLIVEIRA, MARCO AURELIO SCHULER; SEIXAS, FLAVIO AUGUSTO VICENTE; BARBOSA-TESSMANN, IONE PARRA. A Novel Cellobiohydrolase I (CBHI) from <i>Penicillium digitatum</i>: Production, Purification, and Characterization. <i>APPLIED BIOCHEMISTRY AND BIOTECHNOLOGY.</i>, v.192, p.257 - 282, 2020.</p> <ul style="list-style-type: none">- For the first enzyme assays, determining the best substrate for the reaction is also ideal. And show this data too, mainly with statistical analysis (triplicates). I recommend these articles here: <p>CASTRO, FAUSTO F.; PINHEIRO, ANA B. P. ; GERHARDT, EDILEUSA C. M. ; OLIVEIRA, MARCO A. S. ; BARBOSA-TESSMANN, IONE P. . Production, purification, and characterization of a novel serine-esterase from <i>Aspergillus westerdijikiae</i>. <i>JOURNAL OF BASIC MICROBIOLOGY</i>, v. 58, p. 131-143, 2018</p> <p>DE CASTRO, FAUSTO FERNANDES; PONCHIO PINHEIRO, ANA BEATRIZ ; BEATRIZ NASSUR, CAROLINA ; PARRA BARBOSA-TESSMANN, IONE . Mycelium-bound lipase from a locally isolated strain of <i>Aspergillus westerdijikiae</i>. <i>BIOCATALYSIS AND AGRICULTURAL BIOTECHNOLOGY</i>, v. 10, p. 321-328, 2017</p> <ul style="list-style-type: none">- One of the cultivation optimization data for enzyme production was also presented (3 days), but it was only written at the end of the work. It would be interesting to present this data in another way, as a graph, for example, since it was done in triplicate. And add to the methodology.	
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<p>Minor REVISION comments</p>	<ol style="list-style-type: none"> 1- Every time you add numbers, standardize with the long number first, or just leave the numeral itself. This happens throughout the entire text. Example: 10 (ten) minutes OR 10 minutes. 2- Some English corrections, as in “spectrophotometrically” – spectrophotometrically 3- In the legend of table 3, I recommend putting the signs in parentheses (+) and (-) 4- In item 2.5, the fungal growth medium was not described before incubation with The Bushnell Haas broth. I believe it is the MEA but it is not mentioned in the text. Also, as there is no use of the mycelium to determine activity, the crude extract can be filtered through filter paper, without the need to sterilize 5- In determining lipase activity, were the reactions performed in triplicate? If not, I highly recommend it - this to validate the data. 6- In item 3.2, how was the amount of fungi in the samples determined? The results were presented but the methodology was not. 7- In item 3.5, I would replace the word "fermentation" with incubation. 8- In some references, check that the volume numbers of the journal are all standardized 	
<p>Optional/General comments</p>	<p>As the need to optimize production for better production of lipase has been demonstrated, perhaps the focus of the work could be only on qualitative analyses and leave a second work for quantitative analyses. The work would be more interesting by testing the different means of lipolytic activity with the fungi found. Add figures/photos of the different results and comparison tables of halos formed.</p>	

PART 2:

	<p>Reviewer’s comment</p>	<p>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)</p>
<p>Are there ethical issues in this manuscript?</p>	<p><i>(If yes, Kindly please write down the ethical issues here in details)</i></p>	

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