

International Journal of Recycling of Organic Waste in Agriculture

p-ISSN : 2195-3228 e-ISSN : 2251-7715

BUKTI KORESPONDENSI IJROWA

International Journal of Recyling of Organic Waste in Agriculture (IJROWA)

Judul : Cultivation of straw mushroom (Volvariella volvacea) on oil palm empty fruit bunch growth medium

- > SUBMIT (21 Mei 2018)
- REVISI JURNAL TAHAP 1 (16 Agustus 2018)
- REVISI JURNAL TAHAP 2 (14 Januari 2019)
- REVISI JURNAL TAHAP 3 (16 Maret 2019)
- ➤ TERBIT DI IJROWA (11 April 2019)

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IRWA-D-18-00104 - Submission Confirmation

From: International Journal of Recycling of Organic Waste in Agriculture (IRWA) (em@editorialmanager.com)

To: striyono2001@yahoo.com

Date: Monday, May 21, 2018, 02:13 AM GMT+7

Dear Mr. Triyono,

Thank you for submitting your manuscript, "Cultivation of Rice Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch (EFB) Growth Medium", to International Journal of Recycling of Organic Waste in Agriculture

The submission id is: IRWA-D-18-00104 Please refer to this number in any future correspondence.

During the review process, you can keep track of the status of your manuscript by accessing the journal's website.

Your username is: striyono2001 If you forgot your password, you can click the 'Send Login Details' link on the EM Login page at <u>https://irwa.editorialmanager.com/</u>

With kind regards, Springer Journals Editorial Office International Journal of Recycling of Organic Waste in Agriculture

IRWA-D-18-00104 - Revise before review

From: International Journal of Recycling of Organic Waste in Agriculture (IRWA) (em@editorialmanager.com)

To: striyono2001@yahoo.com

Date: Saturday, June 2, 2018, 11:29 AM GMT+7

CC: <u>agusharyid65@gmail.com</u>, <u>marelitelaumbanua@gmail.com</u>, <u>dermiyati.1963@fp.unila.ac.id</u>, <u>jlumbanraja53@gmail.com</u>

Dear Mr. Triyono,

I have read your manuscript, "Cultivation of Rice Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch (EFB) Growth Medium", submitted to International Journal of Recycling of Organic Waste in Agriculture

Before it can be sent out for review, I would request that you carry out the corrections below.

Please submit your revised manuscript by accessing the journal's website.

Your username is: striyono2001 If you forgot your password, you can click the 'Send Login Details' link on the EM Login page at <u>https://irwa.editorialmanager.com/</u>

Please note: when uploading your revised files, please make sure only to submit your editable source files (i. e. word, tex).

We are looking forward to receiving your revised manuscript before 17 Jul 2018.

Thank you very much.

With kind regards, Payam Najafi Director – in – Chief International Journal of Recycling of Organic Waste in Agriculture

COMMENTS TO THE AUTHOR:

1. The manuscript should be revised for English language before evaluation. It is better that a native person or some sites help you for this point.

You can refer the link <u>http://www.editorialmanager.com/homepage/ms/springer-msservices.html</u> for information regarding editing service.

2.Please provide a structured abstract of 150 to 250 words which should be divided into the following sections:

Purpose (stating the main purposes and research question) Methods Results Conclusions The manuscript hasn't Conclusions.

3. The doi of articles should be added in references.

4. You should introduce the full keywords, not the abbreviation.

5.We recommend using of new articles which have been published in this journal recently.

IRWA-D-18-00104 - gentle reminder

From: International Journal of Recycling of Organic Waste in Agriculture (IRWA) (em@editorialmanager.com)

To: striyono2001@yahoo.com

Date: Tuesday, July 3, 2018, 11:46 AM GMT+7

Ref.: Ms. No. IRWA-D-18-00104

Cultivation of Rice Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch (EFB) Growth Medium International Journal of Recycling of Organic Waste in Agriculture

Dear Mr. Triyono,

When checking our records, we noticed that the due date for your revision on IRWA-D-18-00104 is approaching.

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IRWA-D-18-00104R1 - clarification needed

From: International Journal of Recycling of Organic Waste in Agriculture (IRWA) (em@editorialmanager.com)

To: striyono2001@yahoo.com

Date: Tuesday, July 17, 2018, 10:42 AM GMT+7

Journal Name : International Journal of Recycling of Organic Waste in Agriculture Manuscript Number : IRWA-D-18-00104R1 Article Title : Cultivation of Rice Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch (EFB) Growth Medium

Dear Mr. Triyono,

We have received your revised manuscript entitled "Cultivation of Rice Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch (EFB) Growth Medium".

During the initial check we have noticed a discrepancy in the authors list between the previous version and this version.

Currently the author list is as follows:

Sugeng Triyono, Ph.D.; Agus Haryanto, Ph.D.; Mareli Telaumbanua, Ph.D; Dermiyati Dermiyati, Ph.D.; Jamalam Lumbanraja, Ph.D.; Filip To, Ph.D

Could you please clarify this change, and also let us know if all co-authors are aware of this?

Please fill in the attached form and upload it together with your revision. Please note that all contributing authors should sign the form.

I have returned the submission to you via the system, and it can be found in the folder 'Submissions Sent Back to Author'.

Please re-submit your manuscript together with your clarification.

Your username is: striyono2001 If you forgot your password, you can click the 'Send Login Details' link on the EM Login page at <u>https://irwa.editorialmanager.com/</u>

We will assign your revision to the Editors after having received the fully filled out form.

The Editors will decide to continue with your revision, based on the information provided.

If we do not hear back from you within 21 days your paper will be considered as rejected.

Thank you very much.

With kind regards,

Ashi Asokan JEO Assistant International Journal of Recycling of Organic Waste in Agriculture



Authorship+form_imprints various - pre-acc.pdf 369.2kB

IRWA-D-18-00104R4 - Manuscript Sent Back

From: International Journal of Recycling of Organic Waste in Agriculture (IRWA) (em@editorialmanager.com)

To: striyono2001@yahoo.com

Date: Saturday, March 23, 2019, 04:22 PM GMT+7

Dear Mr. Triyono,

Your submission entitled "Cultivation of Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch Growth Medium" has been received.

Before we can further process it you are kindly requested to make the following corrections to meet the journal's requirements (please also refer to the Instructions for authors):

Remove the author name from the file name (SugengTriyono_Article_11 J Recycling Org Wastes (7).doc)

Please log onto Editorial Manager as an author.

Your username is: striyono2001 If you forgot your password, you can click the 'Send Login Details' link on the EM Login page at <u>https://www.editorialmanager.com/irwa/</u>

Go to the menu item 'Submissions Sent Back to Author', and click on 'Edit Submission'. If no changes are to be made in the metadata, please go to the submission step 'attach files', and upload your corrected submission. Build the PDF, view your submission, and approve the changes.

Thank you for submitting your work to this journal.

With kind regards,

Harini Senthil JEO Assistant International Journal of Recycling of Organic Waste in Agriculture

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Section 1: Please provide the current title of manuscript

(For journals: Please provide the manuscript ID, title and/or DOI if available.) (For books: Please provide the title, ISBN and/or DOI if available.) Manuscript ID no. in case of unpublished manuscript: IRWA-D-18-00104R1

DOI in case of published manuscript:

ISBN (for books):

Title: CULTIVATION OF RICE STRAW MUSHROOM (Volvarealla Volvacea) ON OIL PALM EMPTY FRUIT BUNCH (EFB) GROWTH MEDIUM

Section 2: Please provide the previous authorship, in the order shown on the manuscript before the changes were introduced. Please indicate the corresponding author by adding (CA) behind the name.

| | First name(s) | Family name | ORCID or SCOPUS id, if available |
|------------------------|---------------|--------------|---------------------------------------|
| 1 st author | SUGENG | TRIYONO (CA) | https://orcid.org/0000-0001-6678-9198 |
| 2 nd author | AGUS | HARYANTO | https://orcid.org/0000-0002-8954-4463 |
| 3 rd author | MARELI | TELAUMBANUA | - |
| 4 th author | DERMIYATI | - | https://orcid.org/0000-0001-5888-6084 |
| 5 th author | JAMALAM | LUMBANRAJA | https://orcid.org/0000-0002-4694-3640 |
| 6 th author | | | |
| 7 th author | | | |

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Change of authorship request form (pre-acceptance)

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WE ASKED A HELP TO DR. FILIP TO, TO REVISE THE MANUSCRIPT FOR ENGLISH LANGUAGE AND SOME SUBSTANTIAL POINTS WHEN THE MANUSCRIPT NEEDED TO BE REVISED, AT THE FIRST REVISION. WE THINK THAT HIS CONTRIBUTION WAS MEANINGFUL AND HE DESERVES FOR BEING COAUTHOR., WE AGREED TOADD HIM ON THE AUTHOR LIST AT THE SECOND SUBMISSION.

WE APOLOGIZE FOR THIS INCONVENIENCE

Section 4: Proposed new authorship. Please provide your new authorship list in the order you would like it to appear on the manuscript. Please indicate the corresponding author by adding (CA) behind the name. If the corresponding author has changed, please indicate the reason under section 3.

| | First name(s) | Family name (this name will appear in full on the final publication and will be searchable in various abstract and indexing databases) |
|------------------------|---------------|--|
| 1 st author | SUGENG | TRIYONO (CA) |
| 2 nd author | AGUS | HARYANTO |
| 3 rd author | MARELI | TELAUMBANUA |
| 4 th author | DERMIYATI | DERMIYATI |
| 5 th author | JAMALAM | LUMBANRAJA |
| 6 th author | FILIP | то |
| 7 th author | | |

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New acknowledgements:

N/A

New Disclosures (financial and non-financial interests, funding):

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None

New Author Contributions statement (if applicable per the journal policy): out of state collaborator of necessaria in verstigatos

State 'Not applicable' if there are no new authors.

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| 3 rd author | MARELI | TELAUMBANUA | I agree to the proposed new authorship shown in section 4 /and the addition/removal*of my name to the authorship list. | Reind | LAMPUNG UNIVERSITY | 19/7/2018 |
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| 6 th author | FILIP | то | I agree to the proposed new authorship shown in section 4 /and the addition/zemoval*of my name to the authorship list. | pili | MISSISSIPPI STATE UNIVERSITY | 19/7/2018 |
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IRWA-D-18-00104R1 - clarify authorship change

From: Ashi Asokan (em@editorialmanager.com)

To: striyono2001@yahoo.com

Date: Tuesday, August 7, 2018, 03:47 PM GMT+7

Journal Name : International Journal of Recycling of Organic Waste in Agriculture Manuscript Number : IRWA-D-18-00104R1 Article Title : Cultivation of Rice Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch (EFB) Growth Medium

Dear Mr. Triyono,

Could you please respond to our previous request to clarify the change in authorship in your revision?

Please re-submit within 10 working days or, if you are not able to do so, send us a response by return email.

Your submission can be found in the folder 'Submissions Sent Back to Author'.

Please re-submit your manuscript together with your clarification.

Your username is: striyono2001 If you forgot your password, you can click the 'Send Login Details' link on the EM Login page at <u>https://irwa.editorialmanager.com/</u>

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Thank you very much.

With kind regards,

Ashi Asokan JEO Assistant International Journal of Recycling of Organic Waste in Agriculture

IRWA-D-18-00104R1 - clarify authorship change

From: Ashi Asokan (em@editorialmanager.com)

To: striyono2001@yahoo.com

Date: Thursday, August 16, 2018, 01:36 PM GMT+7

Journal Name : International Journal of Recycling of Organic Waste in Agriculture Manuscript Number : IRWA-D-18-00104R1 Article Title : Cultivation of Rice Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch (EFB) Growth Medium

Dear Mr. Triyono,

Could you please respond to our previous request to clarify the change in authorship in your revision?

Please re-submit within 10 working days or, if you are not able to do so, send us a response by return email.

Your submission can be found in the folder 'Submissions Sent Back to Author'.

Please re-submit your manuscript together with your clarification.

Your username is: striyono2001 If you forgot your password, you can click the 'Send Login Details' link on the EM Login page at <u>https://irwa.editorialmanager.com/</u>

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If we do not hear back from you within 10 days your paper will be considered as rejected.

Thank you very much.

With kind regards,

Ashi Asokan JEO Assistant International Journal of Recycling of Organic Waste in Agriculture

International Journal of Recycling of Organic Waste in Agriculture Cultivation of Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch Growth Medium --Manuscript Draft--

| Manuscript Number: | IRWA-D-18-00104R2 | | | | |
|--|--|--|--|--|--|
| • | | | | | |
| Full Title: | Cultivation of Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch Growth Medium | | | | |
| Article Type: | Original Research | | | | |
| Funding Information: | Kementerian Riset, Teknologi dan Pendidikan Tinggi (ID) (1640/UN26.21/KU/2017; 062/SP2HL/LT?DRPM/2018) | | | | |
| Abstract: | Abstract Purpose The research aimed to study effects of size reduction and composting duration of EFB (Empty Fruit Bunches) on straw mushroom production, and to examine the doses of fertilizers commonly used among farmers. Methods The experiment was done in two stages. The first stage was done for identifying optimum physical parameters of EFB, and the second stage focused on enhancing the performance of EFB which was chosen from the first experiment. Experiments in a Randomized Completely Block Design (RCBD) with a 3x3 factorial treatment were carried out in both stages of the study. The first stage had 3 levels of aggregate sizes factor (S) and 3 levels of composting duration factor (C) of EFB. The second stage had 3-levels of NPK factor (N) and 3 levels of organic fertilizer factor (O). Results The whole stalk of EFB produced the highest productivity of 2458.47±1015.23 g m-2. The supplement of fertilizers increased EFB decomposition rate, productivity to 2950.24±208.50 g m-2, and nutritive values (particularly for protein content of 41.00±3.79%). Averaged BCE (Biological Conversion Efficiency) also improved from 3.61±1.22 to 6.56±0.46%. Conclusions EFB did not need to be cut into smaller pieces, and should not be composted for more than 8 days, because there was a tendency to decrease yield. Supplemental fertilizers increased decomposition rates of EFB as well as yield and nutritive values of straw mushroom. The BCE can potentially be improved by increasing the dosages of fertilizers, use of micronutrients, or use of enzyme- producing agents that capable of delignification. Keywords: Agricultural waste management, Arduino, Biological Conversion Efficiency, Controlled environment, Fermentation, Lignocelluloses | | | | |
| Corresponding Author: | Sugeng Triyono, Ph.D. Lampung University Bandar Lampung, Lampung INDONESIA | | | | |
| Corresponding Author Secondary Information: | | | | | |
| Corresponding Author's Institution: | Lampung University | | | | |
| Corresponding Author's Secondary Institution: | | | | | |
| First Author: | Sugeng Triyono, Ph.D. | | | | |
| First Author Secondary Information: | | | | | |
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| | Mareli Telaumbanua, Ph.D | | | | |
| | Dermiyati Dermiyati, Ph.D. | | | | |
| | Jamalam Lumbanraja, Ph.D. | | | | |
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| | Filip To, Ph.D |
|---|--|
| Order of Authors Secondary Information: | |
| Author Comments: | Thank you for the chance of publication |
| Response to Reviewers: | COMMENTS FOR THE AUTHOR: Reviewer #1: The manuscript describes the optimisation of media form EFB for the cultivation of rice straw mushrooms. Findings are of interest as it represents one of the ways to deal with the increasing environmental wastes. This manuscript also, however, suffers from several technical issues. Data presentation (first vs. second batch and separation into three parts vs. complete fruiting body) needs to be improved and data are usually presented in the form of mean and SD. The authors may also want to compare and contrast their results with those reported in the literature. How does their findings advance our knowledge regarding the optimal substrate for the cultivation of rice straw mushroom? Response to Reviewer #1: Data presentations have been improved. Some relevant literatures have been compared. Appreciate. Reviewer #2: The manuscript was studied and following observations are being made |
| | i) The English is not up to the standard throughout the manuscript. Response: have been trying to be better. The English have been revised. ii) The mushroom is popularly known as straw mushroom not the rice straw mushroom. Response: It has been revised iii) Its not clear that how 9 trays of 75 cm length can be adjusted in room size of 6 meter length because 9 trays are making it 675 cm if no space is given between trays. Response: The three-layer racks consisted two rows (right and left), 1.5 m in between giving an access for operational. So there are 5 trays in one side and 4 trays in the other side for every layer (there are 3 layers). Calculated total length is 75 cm x 5 = 375 cm max. The rack is given 4 m long. (P 2 line 18-21) |
| | iv) Space in front, back and sides of racks has n ot been mentioned. Response: the rack was 4 m long and 3 m wide, giving 1.5 m for front space, and 0.5 m for right, left, and back spaces. (P 2 line 21-22) v) In mushrooms its not transplanting rather it is inoculation or mixing. Response: appreciate. it has been revised vi) Meinture content of the EER in the beginning of the experiment is missing. |
| | vi) Moisture content of the EFB in the beginning of the experiment is missing. Response: revised. moisture content is mentioned on p 6 line 11 vii) The quantity of the inorganic and organic fertilizers used is per sack or what? Its not clear. Response: an experimental unit is initially in a sack when 100 kg EFB substrate is prepared for composting. Each sack of EFB substrate is then transferred to a growing tray in the mushroom house when pasteurization is ready, then the experimental unit is in the growing tray. The every dosage of treatment combination is for every experimental unit. (p 4 line 1-11). |
| | viii) Commercial spawn was used @ 70 g/bed. Is it sufficient enough? Response: As written on the label, the volume of spawn was 3.500 cc (=1.5 kg) per bag. One bag was used for three beds. Yes that is mistaken. Actually we spent 9 bags of seed for 27 beds. Revision in the manuscript has been made. |
| | ix) It has been mentioned that pasteurization is for removal of nitrogen which is not true. Response: thank oyu. It has been revised (p4 line 16-17) |

x) Requirement of light and fresh air is not mentioned any where, which are crucial for indoor cultivation of this mushroom.

Response:

On P 2, preparation section, it is mentioned that the perimeter of house is covered by net, semi-transparent tarpaulin and transparent plastic. Sun light penetrate the seal but not to be excessive. This is what normally done by local farmers. The description is added, but light for night time is not given.

P2 line 24-25. There are two units of electric fans, one for air intake placed at lower part of the slab (at the rear), and the other is for exhaust placed at the upper part (above the door fig 1a). Those fans are mainly to control air temperature and RH, regulated by an automatic device. Room air should be replaced when the fans are on, particularly at around noon and midnight to early morning. Description is added. xi) Fig 3 is not required.

Response: Fig 3 has been deleted

xii) Spelling mistakes are there at several places like for physico-chemical, fruit body, etc.

Response: it have been corrected

xiii) The points suggested for improvement have not been incorporated.

Response: discussion section is improved.

xiv) The yield data of second experiment i.e. treatment wise is missing.

Response: the tables of the data have been synchronized.

xv) Statistical analysis is missing.

Response: Statistical analysis in each experiment is clarified. But comparisons between the two experiments were done descriptively.

xvi) The content of the three different substrates before and after pasteurization with respect to cellulose, hemi-cellulose and lignin is missing. Response: The contents were compared between raw EFB and after mushroom cultivation. The different contents were considered as the changes of cellulose, hemicelluloses, and lignin during global processes of fermentation, pasteurisation, and mushroom growth.

xvii) Relevant references are missing.

Response: some relevant references have been added.

xviii) Discussion part is very poorly written

Response: it has been improved

xix) Overall yield level is very low.

Response: The yield was in fact comparable to what the local farmers have done. There were several factors affecting the low BCE, one of which is high bulk density of EFB. Some potential methods for improving the yield have been added.

Reviewer #3: The subject is interesting because it deals with the processing of an agro-industrial residue in a value-added product; however, the text needs a further information:

It's necessary to indicate, at the beginning of the experiment, that the residue was outside the mill and was submitted to the weather. It is a composting! (line14, p 7) Response: the EFB waste was taken from nearest palm oil mill. As in normal situation, the EFB is just dumped outside the mill. Farmers collected the EFB waste for growing the mushroom. The EFB collected by farmers is normally not fresh, but about one month on the pile, not intended composting. The information has been added on the beginning.

Give details about the composting!? Line12 p3 Response: the description has been improved and may be understandable.

The spawning rate of 7 for 1000 is too low (70g spawn/100kg EFB), is there any error?(line 22 p 4)

Response: that is mistaken. We actually spent 9 bags for 27 beds, making a bag of 1.5 kg for three beds, or 500 gram for 100 kg substrate.

What means am.? (line33 p4) Response: sorry, it has been corrected. I mean harvest is done at 5:00 a.m.

| | Correct the title of tab.2 (line36 p 6) Response: appreciate. |
|--|--|
|--|--|

COMMENTS FOR THE AUTHOR:

Reviewer #1: The manuscript describes the optimisation of media form EFB for the cultivation of rice straw mushrooms. Findings are of interest as it represents one of the ways to deal with the increasing environmental wastes. This manuscript also, however, suffers from several technical issues. Data presentation (first vs. second batch and separation into three parts vs. complete fruiting body) needs to be improved and data are usually presented in the form of mean and SD. The authors may also want to compare and contrast their results with those reported in the literature. How does their findings advance our knowledge regarding the optimal substrate for the cultivation of rice straw mushroom?

Response to Reviewer #1:

Data presentations have been improved. Some relevant literatures have been compared. Appreciate.

Reviewer #2: The manuscript was studied and following observations are being made

i) The English is not up to the standard throughout the manuscript. Response: have trying to be better. The English have been revised.

ii) The mushroom is popularly known as straw mushroom not the rice straw mushroom.

Response: It has been revised

iii) Its not clear that how 9 trays of 75 cm length can be adjusted in room size of 6 meter length because 9 trays are making it 675 cm if no space is given between trays.

Response: The three-layer racks consisted two rows (right and left), 1.5 m in between giving an access for operational. So there are 5 trays in one side and 4 trays in the other side for every layer (there are 3 layers). Calculated total length is 75 cm x 5 = 375 cm max. The rack is given 4 m long. (P 2 line 18-21)

iv) Space in front, back and sides of racks has n ot been mentioned. Response: the rack was 4 m long and 3 m wide, giving 1.5 m for front space, and 0.5 m for right, left, and back spaces. (P 2 line 21-22)

v) In mushrooms its not transplanting rather it is inoculation or mixing. Response: appreciate. it has been revised

vi) Moisture content of the EFB in the beginning of the experiment is missing. Response: revised. moisture content is mentioned on p 6 line 11

vii) The quantity of the inorganic and organic fertilizers used is per sack or what? Its not clear.

Response: an experimental unit is initially in a sack when 100 kg EFB substrate is prepared for composting. Each sack of EFB substrate is then transferred to a growing tray in the mushroom house when pasteurization is

ready, then the experimental unit is in the growing tray. The every dosage of treatment combination is for every experimental unit. (p 4 line 1-11).

viii) Commercial spawn was used @ 70 g/bed. Is it sufficient enough?



Response:

As written on the label, the volume of spawn was 3.500 cc (=1.5 kg) per bag. One bag was used for three beds. Yes that is mistaken. Actually we spent 9 bags of seed for 27 beds. Revision in the manuscript has been made.

ix) It has been mentioned that pasteurization is for removal of nitrogen which is not true.

Response: thank oyu. It has been revised (p4 line 16-17)

x) Requirement of light and fresh air is not mentioned any where, which are crucial for indoor cultivation of this mushroom. Response:

On P 2, preparation section, it is mentioned that the perimeter of house is covered by net, semi-transparent tarpaulin and transparent plastic. Sun light penetrate the seal but not to be excessive. This is what normally done by local farmers. The description is added, but light for night time is not given.

P2 line 24-25. There are two units of electric fans, one for air intake placed at lower part of the slab (at the rear), and the other is for exhaust placed at the upper part (above the door fig 1a). Those fans are mainly to control air temperature and RH, regulated by an automatic device. Room air should be replaced when the fans are on, particularly at around noon and midnight to early morning. Description is added.

xi) Fig 3 is not required.

Response: Fig 3 has been deleted

xii) Spelling mistakes are there at several places like for physico-chemical, fruit body, etc.

Response: it have been corrected

xiii) The points suggested for improvement have not been incorporated. Response: discussion section is improved.

xiv) The yield data of second experiment i.e. treatment wise is missing. Response: the tables of the data have been synchronized.

xv) Statistical analysis is missing.

Response: Statistical analysis in each experiment is clarified. But comparisons between the two experiments were done descriptively.

xvi) The content of the three different substrates before and after pasteurization with respect to cellulose, hemi-cellulose and lignin is missing. Response: The contents were compared between raw EFB and after mushroom cultivation. The different contents were considered as the changes of cellulose, hemicelluloses, and lignin during global processes of fermentation, pasteurisation, and mushroom growth. xvii) Relevant references are missing.

Response: some relevant references have been added.

xviii) Discussion part is very poorly written

Response: it has been improved

xix) Overall yield level is very low.

Response: The yield was in fact comparable to what the local farmers have done. There were several factors affecting the low BCE, one of which is high bulk density of EFB. Some potential methods for improving the yield have been added.

Reviewer #3: The subject is interesting because it deals with the processing of an agro-industrial residue in a value-added product; however, the text needs a further information:

It's necessary to indicate, at the beginning of the experiment, that the residue was outside the mill and was submitted to the weather. It is a composting! (line14, p 7)

Response: the EFB waste was taken from nearest palm oil mill. As in normal situation, the EFB is just dumped outside the mill. Farmers collected the EFB waste for growing the mushroom. The EFB collected by farmers is normally not fresh, but about one month on the pile, not intended composting. The information has been added on the beginning.

Give details about the composting!? Line12 p3 Response: the description has been improved and may be understandable.

The spawning rate of 7 for 1000 is too low (70g spawn/100kg EFB), is there any error?(line 22 p 4)

Response: that is mistaken. We actually spent 9 bags for 27 beds, making a bag of 1.5 kg for three beds, or 500 gram for 100 kg substrate.

What means am.? (line33 p4) Response: sorry, it has been corrected. I mean harvest is done at 5:00 a.m. Correct the title of tab.2 (line36 p 6) Response: appreciate.

Cultivation of Straw Mushroom (*Volvarealla volvacea*) on Oil Palm Empty Fruit Bunch Growth Medium

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1 Introduction

Indonesia's palm oil mills generate a large amount of oil palm (*Elaeis guineensis*) and EFB (Empty Fruit Bunches) that are currently discarded as waste. Each ton of fresh EFB processed produces about 0.21 ton (21%) of crude palm oil (CPO), 0.05 ton (5%) of PKO (Palm Kernel Oil) and the remainder are solid wastes consisted of EFB (23%), fibers (13.5%), and shells (5.5%) (Poeloengan et al. 1993). In 2015, Indonesia's oil palm plantation area was around 11.3 million ha, producing about 31.3 million tons of FFB, and generated about 7.2 million tons of EFB (Ministry of Agriculture 2017). Although some of the EFB are used as organic mulch in gardens, the majority of them are not utilized (wasted). Many private palm oil mills receive EFB directly from farmers, and the EFB is simply discarded around the factory as waste, polluting the environment.

Composting and redistributing EFB into plantations may be a good way of managing EFB waste from palm oil mills, but only a few of them practice this management scheme because it is considered not cost effective. A more traditional method of EFB utilization is for cultivation of straw mushrooms in individual farms. Farmers use EFB collected from palm oil mills as a growth medium (called as substrate) for cultivating mushrooms directly without any pretreatment, resulting in lower BCE (Biological Bonversion Efficiency).

Edible mushrooms are high value products, they have a delicious taste, soft texture, and the crude protein content is about 25.9-28.5% (Sunandar 2010), and they have been cultivated for long time (Sánchez 2004). Although the cultivation of mushrooms as nutritious food (Cheung 2013) has been widely practiced (Chang and Wasser 2017), the production rate remains below consumers' demand. The demand for mushroom in Indonesia in 2010 was about 25 tons per day, but its production was only 15 tons per day (Hendritomo 2010). Globally, the world's production of edible mushroom has increased in the past decade from 23,559 tons in 2006 to 40,906 tons in 2016 (Faostat 2016). Developing methods of using EFB as a growth medium for straw mushroom can potentially improve the supply of mushroom in palm oil producing regions of the world to meet the strong demand.

There have been a number of research efforts in the use of EFB for cultivating oyster mushroom (*Pleurotus ostreatus*) including the following cited literatures: Tabi et al. (2008); Rizki and Tamai (2011); Sudirman et al. (2011); Kavitha, et al. (2013); and Marlina et al. (2015). Some works on EFB as a biofertilizer have been published including: Kananam, et al. (2011); Hayawin, et al. (2012); Hoe et al. (2016), and EFB works in bioenergy (Sudiyani et.al. 2013; Chang 2014; Tye et al. 2014; Muryanto et al. 2015; Zulkiplea et al. 2016). Agricultural wastes from cotton (Sukendro et al. 2001; Rajapakse 2011), coffee husk (Mayun 2007), oil palm fiber (Onuoha et al. 2009; Ichsan et al. 2011), coconut husk (Rahmanda, 2014), banana leaves (Belewu and Belewu, 2005) or their mixtures have been investigated as growth media of straw mushroom. Straw mushroom cultivation using EFB substrate has been reported by Thiribhuvanamala et al. (2012). In their research, straw mushroom was cultivated on various agricultural solid wastes including oil palm bunch waste, but there was no mention of any praperation process of the oil palm bunch.

Published research showed that proper pretreatment of mushroom growth media is required in order to improve productivity. The pretreatments include: composting, addition of supplemental nutrients and pasteurization. It was reported that composting of substrate for a period of time (Vargas and Hepperly 1986) prior to pasteurization softened the EFB and helped in the breaking down of lignin (de-lignifications) (Wood and Leatham 1983). There is no standard operating procedure in determining the duration for composting EFB media and whether composting is mandatory. Consequently, some farmers do compost EFB growth media, while others do not. The duration of composting cotton waste has been shown to have a significant impact on the productivity of straw mushroom (Sukendro, et al. 2001). Arifestiananda et al. (2015) found that a five-day duration for composting rice straw was the most optimum composting time, based on total weight and number of fruit body harvested.

Physical properties of the media including their crystallinity, surface area, porosity, and particle size were also known to affect mushroom growth (Pandey 2003 and Viniegra-González et al. 2003). For Oyster mushroom, EFB were mostly chopped into smaller sizes to prepare it as a substrate (Sudirman et al. 2011; Kavitha, et al. 2013). Reducing the physical size of EFB prior to its application as the substrate of straw mushroom may have positive effects on production because it results in larger total particle surface area. The spent EFB may also be more practical for further usage as a biofertilizer, as an aggregate to mix with other compost materials (Nugroho et al. 2012; Dermiyati et al. 2015), for dietary supplement of animal feed (Fazaeli et al. 2014; Fard et al. 2014), or for use as a biogas feed stock (Williams et al. 2001).

58 EFB contains 23.7-65.0% cellulose, 20.58-33.52% hemicellulose, 14.1-30.45% lignin (Sreekala 59 et al. 1997, Abdullah et al. 2011, Omar et al. 2011, Apetorgbor et al. 2015). The high cellulose content

is what makes EFB a potential for use as a substrate for cultivating straw mushroom (Zakhary et al. 3 1984), and the reducing its size may also improve the BCE. Other ingredients such as chicken manure, rice bran, lime, and organic or inorganic fertilizers are often added into the growth media during preparation for mushroom cultivation (Zakhary et al. 1984). These supplements were intended to enrich nutrient contents of the mushroom substrate (Fasidi and Akwakwa 1996; Banik and Nandi 2004). Purpose of this research is to explore the effects of size reduction and composting duration of EFB growth medium on the mushroom production, and to investigate the doses of fertilizer supplements commonly used among farmers.

Materials and Methods

Preparation

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This research was conducted in the months of April - October 2017 at the Experimental Station of the Faculty of Agriculture, Lampung University, Indonesia, in a 4 m wide, 6m long, and 5m high mushroom house. The structure was constructed on a concrete slab foundation with 1 m high brick walls on the lower part as shown on Fig. 1a. The upper part of the structure was made of steel frames, and the perimeter was covered with netting, semi-transparent tarpaulin, and 14% UV transparent plastic to prevent excessive direct sunray from penetrating into the room. This setting was what normally done by local farmers. Woven asbestos roof top was used and the ceiling was made of 5 mm plywood to reduce the effect of excessive heat penetrating the roof.

There were two racks with 3 layers of wooden shelves in each (Fig. 1b), positioned inside the mushroom house with 1.5 m spacing between the racks for human access. Each layer of the shelf held 9 trays of growing beds, and there were a total of 27 trays. The dimensions of the trays were 75x75 cm² square and 25 cm depth (Riduwan et al. 2013). Total space occupied by the racks was 4 m long x 3 m wide, giving about 1.5 m for front space, 0.5 m for right, left, and back spaces of the mushroom house. The shelves were separated by a sheet of tarpaulin to prevent leachate contamination between an upper bed and the bed below it. The mushroom house was equipped with 2 units of 35 cm diameter fans for intake air and exhaust (each 350 Watt), 1 unit of circulation fan, 4 units of water spray nozzles, and 1 unit of electric heater (700 Watt) to control temperature and humidity of the room. Room air was replaced with the fresh air whenever the intake and exhaust fans were on, particularly when temperature was high around noon and RH was high during midnight to early morning. The temperature and humidity of the mushroom house were monitored and controlled automatically by a microcontroller (An Arduino Mega-2560) with data recording peripheral. Twenty units of DHT-22 (from Aosong) sensors were used, with 18 units of the sensors located in a regularly spaced manner inside the room, one sensor was put at the attic, and the last one was put outside of the mushroom house. The temperature and RH inside the mushroom house were maintained at the optimum condition of averages in temperature and humidity of 28-33°C (Reyes 2000) and 80-95% (Thiribhuvanamala et al. 2012) respectively.



Fig. 1 a. Mushroom house and b. Selves of bedding trays

1 Experiment Setup and Implementations

The study was carried out in two sequential experiments. The first experiment was to evaluate the effect of reduced sizes of EFB medium and composting duration on the mushroom production. The second experiment was to leverage the findings of the first experiment and to examine the dosage of additional inorganic and organic fertilizers normally used by local farmers for the mushroom production.

7 The EFB waste was transported from nearest palm oil mill which usually dumped the waste 8 outside the mill. The EFB used in this study was the same as normally collected by local farmers for 9 mushroom cultivation. The EFB taken by farmers is not fresh, but it is already about one month on the 10 pile. Supplemental materials such as rice bran, chicken manure, and lime were collected from available 11 sources. Inorganic and organic fertilizers were bought from a nearest farm shop. Commercial F3 straw 12 mushroom seed was purchased from a producer.

13 The First Experiment

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The experiment used a Randomized Completely Block Design (RCBD) with a 3x3 factorial arrangement in 3 replicates. The two factors were the size reduction, and composting durations of EFB. The EFB size reduction (S) factor had 3 levels: EFB stem as small part (S1), EFB hump as moderate part (S2), EFB whole stalk (S3) (Fig. 2). The composting durations (C) factor had 3 levels: 2 days (C1), 5 days (C2), and 8 days (C3). Each treatment combination was replicated/blocked (R) vertically in 3 layers of the shelves, so there were 27 experimental units. The data set was analyzed by using Analysis of Variance and followed by LSD multiple comparison tests.



Fig. 2 Treatment of reduced size EFB for the straw mushroom substrates

The EFB was firstly processed and cut into nine groups at one time, consisting of three groups of stem (S1), three groups of hump (S2), and three groups of whole stalk or left un-cut (S3). Each group of 100 kg EFB was put into a sack, and soaked in water for over night, then removed from the water and drained. Each sack of the moistened EFB was poured out from the sack and thoroughly mixed with supplemental materials: chicken manure, rice bran, and lime. The dosages of supplemental materials were: 80 kg chicken manure, 70 kg rice bran, and 60 kg lime per 1000 kg of EFB for each sack of the mixture. Each resulting mixture was put back into each sack and was composted in the sack for 8 days (C3). From this procedure, we had three treatment combinations of S1C3, S2C3, and S3C3 in three replicates.

Three days later, other nine groups of EFB were processed with the same procedure, and composted for 5 days (C2). From this procedure, we have other three different treatment combinations of S1C2, S2C2, and S3C2 in three replicates. Again, after three days, other nine groups of EFB were also processed with the same procedure, and composted for 2 days (C1). From this procedure, we have other three different treatment combinations of S1C1, S2C1, and S3C1 in three replicates too. After 2 days later, all 27 sacks of the composted EFB were harvested and transferred to the growing beds in the mushroom house randomly. By doing this procedure, we can harvest composted EFB at the same time but different maturity as in designed treatments.

1 The Second Experiment

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 The second experiment used a Randomized Completely Block Design (RCBD) with a 3x3 factorial arrangement in 3 replicates. The factor was a complete inorganic fertilizer (14%N, 14%P, 14%K) and the second factor was a commercial liquid organic fertilizer. The inorganic fertilizer factor (N) had 3 dose levels: 25 g (N₁), 50 g (N₂), 75 g (N₃), and the organic fertilizer factor (O) had 3 dose levels: 5 cc (O_1) , 10 cc (O_2) , and 15 cc (O_3) . Doses of N_2 and O_2 were according to what normally used by local farmers. The treatment combination was replicated vertically in 3 shelf layers (Blocks), so there were a total of 27 experimental units, each contained 100 kg of mixed substrates. Data set was analyzed by using Analysis of Variance and followed by LSD multiple comparison tests.

The second experiment was conducted after the first experiment had been completed (after the mushroom had been harvested and the data sets had been analyzed). The second experiment was set up using the most optimal treatment combination of EFB size and composting duration determined from the first experiment, with the addition of inorganic and organic fertilizers. The EFB bulk materials were initially split into 27 groups each weighted for 100 kg and put it into a sack (a sack became an experimental unit later on) at a time. All sacks of EFB materials were soaked in water overnight then removed and drained. Each sack of EFB was poured and added with lime, rice bran, and chicken manure with the same doses as in the first experiment. Inorganic fertilizer (14%N, 14%P, 14%K) and organic fertilizer (a commercial liquid organic fertilizer) were diluted in 2L water and spaved onto the media according to the designed treatment. Each group of the materials was mixed thoroughly then put back into each sack. The 27 sacks of the prepared substrates were composted for the optimal duration determined from the first experiment. After the composting had completed, all the 27 sacks of the growth media were transferred randomly to the growing beds in the mushroom house according to the rules designed for the experiment. The media were ready to be pasteurized and then inoculated.

24 Pasteurization and Spawning Processes

To pasteurize the mushroom house, water in three 200-liter drums were brought to boil outside the mushroom house; all openings (doors and vents) were shut tightly. The steam was piped into the mushroom house until the temperature inside the house reached 60-70°C, and the temperature was maintained for approximately 5 hours. The purpose of the pasteurization was to kill pathogenic organisms and to suppress wild fungal spores (Kurtzman 2010) that could compete with the cultivated mushrooms.

After 5 hours of steaming, the temperature of the pasteurized media was allowed to naturally cool to ambient temperature (about 28-32°C). The growth media was then moistened with water spray. Commercial F3 mushroom seed of 500 g was spread evenly on the surface of each bed, then the door and ventilations of the mushroom house were closed tightly for 4 days at ambient environmental condition.

36 Maintenance and Mushroom Harvesting

Four days after seeding, the automatic control device was activated to control and record the temperature and humidity of the mushroom house. The temperature was maintained around 28-33°C by means of exhaust fans which were activated when the room temperature was above the set point. An electric heater was used to provide heat when the temperature dropped below set point. The humidity was maintained at about 80-95% by using mist spray nozzles and exhaust fans. The beds of growth media were shielded by tarpaulin sheet to minimize the effect of water spray. The growth media were hand sprayed every other day to maintain the moisture in the growth media.

The first harvest was done at days 8-10 after spawning, when the mushrooms were at egg/button
stage, before the opening of the cap of the fruit body. The fruit body was picked at around 4.00 to 5.00
a.m. The follow up harvests were done whenever the fruit body was at egg stage.

47 Parameters and Measurements

The number of fruit body harvested was weighed and counted, the diameter and height of fruit body were measured by using a caliper. Proximate analyses (crude protein, crude fiber, fat in the fruit body) were determined by the standard methods as described by Helrich (1990); Carbohydrate was calculated by subtracting the total of protein, fiber, and fat from initial weight. Water and ash contents of the fruit body were determined gravimetrically. Cellulose, hemicelluloses, and lignin contents of EFB before and after used as mushroom growth media were measured by using Chesson method as

described by Datta (1981). Biological Conversion Efficiency (BCE) was calculated by taking the ratio
 of fresh yield to dried weight of the growth medium.

Results and Discussion

Temperature and Relative Humidity

The room temperature and relative humidity (RH) of the mushroom house were recorded every 15 minutes, starting from the fifth day of spawning to the end of harvesting. The settings used were 28-33°C (Reyes et al. 1998) for temperature and 80-95% for RH. The data showed that the averages of temperature and RH were 28.07 ± 2.25 °C and $93.89\pm7.06\%$ respectively. The temperature average was near the lower set point level, while the average RH was near the upper set point value. The controllability of temperature and RH are sufficient and did not pose any observable problem to the growth of the mushrooms (Fig 3). The appearance of wild fungi was observed, corroborating the findings of Vargas and Hepperly (1986) who stated that competitors or wild fungi were more antagonistic at low temperature (27°C) than at 35°C. Some species of wild mushrooms observed in these experiments was not identified because it was not a focus of this study.



Fig 3 a. Developing Mycelium (Vegetative phase), b. Fruit body formation (Reproductive phase)

19 Yield of straw mushroom fruit body

In the first experiment, interaction effect between EFB size and composting duration on fruit body yield was found to be not significant (p > 0.05). The effect of EFB composting duration on the fruit body yield was not significant either. The size reduction of the EFB however significantly affected some parameters (total weight, number, and diameter) of fruit body (p < 0.05). Reducing the size of EFB from S3 to S2 and S1 significantly decreased the total weight of fruit body, thus reducing BCE as shown in Table 1. Total number, individual weight, and diameter of button were also negatively affected by reduced sizes of EFB, while height of button was not affected.

Table 1 Effect of size reduction and composting duration of EFB on yield of mushroom in the first
 experiment

| Parameters | Fac | ctor I (Reduced Sizes of | of EFB) | Factor II (O | A | | |
|---|--------------------------|----------------------------|----------------------|----------------------|-------------------------|------------------------|--------------------|
| Parameters | S1 | S2 | S 3 | C1 | C2 | C3 | Average |
| Total weight (g m ⁻²) | 853.73±317.71 c | 1567.21±441.181 b | 2458.47±1015.23 ª | 1975.51±1039.00 a | 1563.06±993.94 a | 1340.84±692.77 ª | 1626.47±547.7 8 |
| BCE (%) | 1,90±0.71° | $3,48\pm0.98^{b}$ | 5,46±2.26ª | 4.39±2.31 ª | 3.47±2.21 ª | 2.98±1.54 ª | 3.61±1.22 |
| Number (buttons m ⁻²) | 87.90±34.46° | 153.0.9±45.65 ^b | 218.86±82.75ª | 176±81.14 ª | 155±74.69 ª | 128±61.69 ^a | 153.28±44.17 |
| Button weight (g button ⁻¹) | 10.20 ±2.44 ^b | 10.28±0.82 ^b | 11.20±1.27ª | 10.87±1.45 ª | 9.91±1.27 ª | 10.79±2.11 ª | 10.52±0.46 |
| Button height (mm) | 33.61 ± 3.22^{a} | 38.59±2.23ª | 39.62±4.45ª | 37.71±4.72 ª | 36.32±4.53 ª | 37.78±3.76 ª | 37.27±2.10 |
| Button diameter (mm) | 23.73±1.58.b | 26.86±1.01ª | 27.72±2.11ª | 26.45±2.04 ª | 26.10±2.77 ^a | 25.76±2.41 ª | 26.11±1.35 |

29 *) means with different letters are significantly different (p<0.05)

The effect of substrate composting duration factor (C) on all parameters was not significant. 3 Arifestiananda et.al. (2015), using substrate of rice straw, found that composting duration of 5 day was significantly different from composting durations of 10 and 15 days in term of total weight and number of fruit body harvested. In This research, even statistically not significant, the weight and number of fruit body parameters tended to noticeably decrease with composting duration. The levels of composting durations (C1, C2, and C3) may be too narrow. This fact suggested that single EFB substrate may not be composted for longer than that in this study because it can potentially decrease mushroom vield.

Based on the reduced sizes of EFB aggregate, the highest productivity of 2458.47 ± 1015.23 g m⁻², was found in S3. This productivity is higher than that usually produced by farmers (about 1700 g m⁻², based on personal communication). For initial EFB water content of 55%; however, the highest BCE was $5.46\pm2.26\%$, slightly lower than what farmers have (about 6.35%). This is because weights of EFB used per m² bed are different. Treatment of S3 also produced the highest total number of fruit body (218.86\pm82.75) and highest weight of individual fruit body (11.20±1.27). Height of individual fruit body in S3 was not different, but in term of diameter the fruit body from S3 was still bigger than in S1. Therefore, it could be said that treatment of S3 was the best over S1 and S2.

17 The fact that reduced size of EFB aggregate decreased productivity was in contrary to the 18 hypothesis because smaller aggregate of growth substrate had larger total surface area and was 19 supposed to be more conducive to the productivity of the mushroom. This may indicate that other 20 factors in smaller aggregate growth media played a role in productivity, an issue needing investigation 21 in the future. It was observed that S1 had the tendency of being more compact or dense as compared to 22 the other two treatments (S2 and S3).

Based on literature, straw mushroom productivity is dependent on growth media, environment, climate, and cultivation systems. Thiribhuvanamala et al. (2012) stated that the variability in mushroom productivity may be attributed to certain physico-chemical factors such as temperature, O₂, CO₂, aeration, wetness and compactness of the beds. He reported that more compact substrate beds gave $\overline{27}$ better production because compact bed might have experienced homogenous moisture level and bed temperature between the layers, which would have facilitated better proliferation of the mycelium, production of more pinheads and buttons with ultimate increase in yield. Apetorgbor et.al. (2015) also found that plantain leaf bundle substrate (supposed to be more compact) was more productive than chopped loose plantain leaf with BCEs of 25% and 15.7% respectively for oil palm mushroom (different strain from straw mushroom in this study). With cotton waste and oil palm fiber substrates, the BCEs were even worse (0.3% and 1.6% respectively). In contrast, BCE (26.7%) of chopped rice straw was higher than BCE (3.7%) of rice straw bundle. Apetorgbor et.al. (2015) mentioned that aeration and drainage of substrates, other than temperature and humidity, could be addressed to the variations of productivity. The compactness of the substrate should be accompanied by better porosity among the aggregates, so aeration and drainage were favorable. In this study, S3 aggregate (like a banana leaf bundle) was more compact, but probably had better aeration and better drainage among the stalks, so produced higher BCE. This finding suggested that EFB did not need to be chopped to smaller sizes if utilized as mushroom media.

The whole stalk EFB was further tested in the second experiment, using five day composting. The second experiment was to determine the optimum doses of inorganic and organic fertilizers added. The results showed that none of the yield parameters was statistically significant (p>0.05) as shown on Table 2. The normal doses of inorganic and organic fertilizers (N2 and O2) as mentioned above were in fact taken from what normally practiced by local farmers. The doses of inorganic and organic fertilizers in the experiment included 50% below normal (N1 and O1) and 50% above normal (N3 and O3), and there was no significant difference on all yield parameters (p>0.05). When compared to the results on the first experiment, all parameters were relatively higher (with no statistical analyses) except for diameter of fruit body. Yields ranged from 2697.61±913.64 to 3177.88±1089.12 g m⁻², BCEs ranged from 5.99±2.03 to 7.06±2.42%, number of fruit body ranged from 278.12±83.73 to 340.54±135.01 buttons m⁻², and individual weight ranged from 8.76±1.58 to 10.41±1.18 g button⁻¹. The productivity was in general improving upon fertilizer addition if compared to the first experiment. The BCEs of some treatments were also relatively higher than what farmers have.

54 Table 2 Effect of fertilizer supplements on yield of mushroom in the second experiment

| Donomotoro | Fa | Factor I (Doses of NPK) Factor II (Doses of Organic Fertilizer) | | | | A | |
|-----------------------------------|----------------|---|----------------|----------------|-----------------|---------------|----------------|
| Parameters | N1 | N2 | N3 | 01 | O2 | O3 | Average |
| Total weight (g m ⁻²) | 3177.88±1089.1 | 2931.75±590.5 | 2727.51±1143.2 | 3173.73±1207.6 | 2697.61±913.6 | 2992.99±699.7 | 2950.24±.208.5 |
| BCE (%) | 7.06±2.42 | 6.52±1.31 | 6.06±2.54 | 7.05±2.68 | 5.99 ± 2.03 | 6.65±1.56 | 6.56±0.46 |
| Number (buttons m ⁻²) | 304.00±89.48 | 297.09±56.01 | 319.21±137.56 | 340.54±135.01 | 278.12±83.73 | 301.63±53.67 | 306.77±21.18 |

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| Button weight (g button ⁻¹) | 10.41±1.18 | 9.87±0.70 | 8.76±1.58 | 9.64±2.13 | 9.56±0.88 | 9.85±0.79 | 9.68±0.54 |
|---|------------|-------------|-------------|------------|-------------|------------|-----------------|
| Button height (mm) | 34.82±3.27 | 33.29±31.99 | 28.97±30.05 | 28.94±2.1 | 30.50±31.98 | 28.97±3.07 | 3.16±0.10 |
| Button diameter (mm) | 23.52±1.71 | 23.08±1.29 | 22.72±1.41 | 23.60±1.73 | 22.73±0.95 | 22.99±1.60 | $2.30{\pm}0.04$ |

|) | When compared to some literatures; however, the BCEs found in this research were considered |
|--------|--|
| , , | 1 |
|) | low to some extent. This may be attributed to other physico-chemical properties of EFB, one of which |
| - | is density, but may need to be investigated further. Bulk density of fresh EFB is more likely higher than |
| 5 | that of any other substrate such as cotton waste, rice straw, sugarcane bagasse, and banana leaf. The |
|) | high bulk density was supposed to contribute to lower BCE. The relatively highest productivity of |
| 7 | 3177.88±1089.12 g m ⁻² (BCE of 7.06±2.42%) in this research was comparable to the research finding |
| 3 | done by Zakhary et.al. (1984), who demonstrated that productivity of straw mushroom ranged from |
|) | 309 g m ⁻² (BCE=1.5%) on single substrate of paddy straw to 5029 g m ⁻² (BCE=14.7%) on mixed |
|) | substrates of orange juice waste, sugar cane, bagasse, horse manure, and mollases. Other research done |
| | by Rajapakse (2011) produced the lowest straw mushroom productivity by 1730 g m ⁻² on paddy straw |
| 2 | substrate to the highest straw mushroom productivity by 5380 g m ⁻² on cotton waste substrate. The |
| 5 | productivity in this research was even better than what Vargas and Hepperly (1986) found (465 g m ⁻² , |
| ŀ | BCE=3.16) on straw mushroom using substrate of sugarcane bagasse. Compared to finding of Belewu |
| 5 | and Belewu (2005) (2500 g m ⁻² of straw mushroom cultivated on substrates of banana leaf bundle), the |
|) | productivity was still better. But BCE found by Belewu and Belewu (2005) was 15.21%, which is |
| , | much higher than in this study. |

More inspiring research finding is in the published work of Thiribhuvanamala et al. (2012). Using supplement of micronutrient based boosters; they found BCE of 20.8±0.95% for straw mushroom cultivated on single substrate of 4 kg oil palm bunch waste (supposed to be the same property as in this research). In spite of different environment, local climate, and techniques, which $\overline{22}$ affect mushroom productivity (Chang and Wasser 2017), the doses of fertilizers used in this research (and also used by farmers) seemed too low (referring to Reves (2000) who used dose of 500g NPK to cultivate straw mushroom on 100kg paddy straw). The BCEs found this research was still considered low, and was potential to be enhanced. Enhancement of BCE may be through applications of higher fertilizer doses, micronutrient based boosters, or using mixtures of substrates from other agricultural wastes. Final choice; however, needs to be practical vet beneficial to local farmers.

Physicho-chemical compositions of straw mushroom fruit body

The physico-chemical composition of mushroom fruit body harvested was analyzed based on crude protein, fat, crude fiber, carbohydrate, water, and ash contents. In the first experiment, there was no significant interaction effect between S (Size reduction of EFB) and C (Composting duration of EFB) factors (p<0.05) on the physico-chemical composition of fruit body (Table 3). Factor of S was significant, but factor of C was not. Protein was the only parameter that was significantly affected by S. The data showed that reducing the sizes of EFB from S3 to S2 and S1 tended to increase the protein contents (from 29.67% to 38.27%). Fat, crude fiber, water, and ash contents were not significantly different among the treatments of S and C. In the first experiment, there was no fertilizer added to the substrate to enhance nutritional values of mushroom, but rice bran, chicken manure, and lime.

40 Table 3 Effect of size reduction and composting duration of EFB on physicho-chemical composition 41 of mushroom (%) in the first experiment

| or musicount (70) in the mist experiment | | | | | | | |
|--|----------------------|-----------------------|-------------------------|---|------------------|-------------------|-----------------|
| Parameters - | Factor I (| Reduced Sizes of | of EFB) | Factor II (Composting Durations of EFB) | | | A |
| Farameters - | S 1 | S2 | S 3 | C1 | C2 | C3 | Average |
| Crude protein | 38.27 ± 8.47^{a} | $32.47{\pm}6.92^{ab}$ | 29.67±7.17 ^b | 35.02±6.07 | 34.33±6.12 | $31.07{\pm}11.41$ | 33.47±3.08 |
| Fat | $4.14{\pm}1.50$ | 3.45 ± 0.96 | 3.52 ± 7.17 | 3.23±0.53 | $4.49{\pm}1.64$ | 3.39 ± 0.98 | 3.71 ± 0.50 |
| Crude Fiber | 7.19 ± 2.38 | 7.57 ± 1.90 | 8.46±3.20 | 7.59 ± 2.35 | 6.82 ± 2.20 | 8.81±2.79 | 7.74±0.76 |
| Karbohydrate | 40.56±9.35 | 47.15±8.43 | 49.26±8.01 | 44.60±7.25 | 44.19 ± 9.27 | $48.18{\pm}10.94$ | 45.66±3.19 |
| Water content* | 90.36±0.35 | 89.87±1.05 | 90.08±1.30 | 90.43±0.86 | 89.91±0.96 | 89.97±1.10 | 90.10±0.24 |
| Ash | 9.83±1.30 | 9.35±1.58 | 9.09±1.57 | 9.56±1.31 | 10.17 ± 1.04 | 8.55±1.63 | 9.42±0.57 |

^{42 *)} Wet basis

In the second experiment, NPK and organic fertilizer were added to enhance both productivity and quality of mushroom. Result showed that none of the psychochemical compositions of fruit body was significantly affected by interaction effect between N and O (Table 4). Simple effects of N (NPK fertilizer) and O (Organic fertilizer) were not significant either. As discussed above, fertilizer additions were in fact too low, that even increase of 50% did not significantly increased productivity. However,

fertilizer addition in the second experiment remarkably enhanced nutrient contents of mushroom 3 relative to that in the first experiment, particularly for crude protein and crude fiber. This is in line with what Jeznabadi et al. (2016) found, that supplement combinations could enhanced both production and quality of mushrooms. On the average, crude protein increased from average of 33.47±3.08% to $41.00\pm3.79\%$, and crude fiber increased from $7.74\pm0.76\%$ to $16.06\pm1.55\%$. Fat, water content, and ash did not much change, while carbohydrate automatically decreased because it was calculated based on protein, fat, and fiber contents. Crude protein content reported by Zakhary et al. (1984) was 30.8% which was lower than that found in this research. Fat, crude fiber, carbohydrate, water, and ash content; however, were in agreement with what Zakhary et al. (1984) reported. Belewu and Belewu (2005) found crude protein (10.25%) and crude fiber (9.23%) contents of straw mushroom cultivated on treated banana leaves which is still lower than found in this research. In conclusions, the supplemented fertilizers noticeably improved both productivity and nutritive values of straw mushroom.

Table 4 Effect of fertilizer supplements on physicho-chemical composition of mushroom (%) in the second experiment

| Doses of NPK | | | Doses of Organic Fertilizer | | | Avanaga |
|------------------|---|---|--|---|--|--|
| N1 | N2 | N3 | 01 | O2 | O3 | Average |
| 42.28±6.69 | 44.94±7.71 | 35.79±8.09 | 39.66±8.57 | 38.16±6.11 | 45.18±8.83 | 41.00±3.79 |
| 4.52 ± 1.18 | 5.02 ± 1.39 | 5.75 ± 1.22 | 5.14 ± 1.51 | 4.87 ± 1.26 | 5.28 ± 1.31 | 5.10 ± 0.41 |
| 14.87 ± 6.58 | 16.70 ± 4.78 | 16.60 ± 7.46 | 13.76 ± 6.52 | 18.19 ± 7.09 | 16.21±4.51 | 16.06±1.55 |
| 28.12±12.85 | 23.63±10.11 | 31.88 ± 14.32 | $32.36{\pm}14.18$ | 28.61±11.69 | 22.65 ± 10.90 | 27.88 ± 4.05 |
| 91.00±0.60 | 91.50±0.58 | 90.73±0.87 | 90.71±0.88 | 91.21±0.66 | 91.31±0.60 | 91.08±0.32 |
| 10.21±1.63 | 9.72±1.42 | 9.99±1.83 | 9.07±1.83 | 10.17±1.53 | 10.67 ± 0.98 | 9.97±0.54 |
| | $\begin{array}{c} 42.28 \pm 6.69 \\ 4.52 \pm 1.18 \\ 14.87 \pm 6.58 \\ 28.12 \pm 12.85 \\ 91.00 \pm 0.60 \end{array}$ | 42.28±6.69 44.94±7.71 4.52±1.18 5.02±1.39 14.87±6.58 16.70±4.78 28.12±12.85 23.63±10.11 91.00±0.60 91.50±0.58 | 42.28±6.69 44.94±7.71 35.79±8.09 4.52±1.18 5.02±1.39 5.75±1.22 14.87±6.58 16.70±4.78 16.60±7.46 28.12±12.85 23.63±10.11 31.88±14.32 91.00±0.60 91.50±0.58 90.73±0.87 | 42.28±6.69 44.94±7.71 35.79±8.09 39.66±8.57 4.52±1.18 5.02±1.39 5.75±1.22 5.14±1.51 14.87±6.58 16.70±4.78 16.60±7.46 13.76±6.52 28.12±12.85 23.63±10.11 31.88±14.32 32.36±14.18 91.00±0.60 91.50±0.58 90.73±0.87 90.71±0.88 | $\begin{array}{cccccccccccccccccccccccccccccccccccc$ | $\begin{array}{cccccccccccccccccccccccccccccccccccc$ |

) Wet basis

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Chemical decomposition of EFB

Before being used as mushroom growth media, EFB's contents of cellulose, hemicelluloses, and 21 lignin were measured and the results are presented on Table 5. According to Bledzki and Gassan (1999) cellulose is a linear condensation polymer consisting of d-anhydroglucopyranose units. Hemicellose comprise a group of polysaccharide (excluding pectin). Lignins are complex hydrocarbon 24 polymer with both aliphatic and aromathic constituents. In the first and the second experiments, initial contents of cellulose, hemicelluloses, and lignin were considered the same and averaged. The cellulose 25 26 27 content was much lower than that (65%) reported by Sreekala et al. (1997). This may be because the EFB used in this research was about 1 month dumped in a pile outside a mill. But the cellulose content was higher than reported by Kavitha et al. (2013). The content of hemicelluloses was also less than 30.5% as reported by Apetorgbor et al. (2015), but (including lignin) still in the ranges as reported by Omar et al. (2011) and Chang (2014). The variations of the components may depend on plant variety, plantation management, locations, climate, and freshness of the EFB waste.

Table 5 Initial EFB's chemical composition

| Composition (%) | First Experiment | Second Experiment | Average |
|-----------------|------------------|-------------------|---------|
| Cellulose | 39.00±1.01 | 39.29±1.10 | 39.15 |
| Hemicelluloses | 20.96±1.09 | 22.31±1.14 | 21.64 |
| Lignin | 24.08±1.53 | 24.16±1.05 | 24.12 |

After being used for mushroom growth media, contents of cellulose, hemicelluloses, and lignin of Spent EFB (post experiment) were also analyzed. Statistical analyses on the chemical composition were performed. In the first experiment, ANOVA analysis showed that interaction effects between S and C on cellulose, hemicelluloses, and lignin were not significant (p > 0.05). Simple effect of S on cellulose content was significant, but not significant on hemicelluloses, and lignin. Simple effect of C on cellulose, hemicelluloses, and lignin were not significant. Table 6 presents treatment combination (S and C) on final cellulose, hemicelluloses, and lignin contents of the spent EFB as the results of LSD comparison analyses. Mean of cellulose content in S_1 (33.22%, averaged combinations with C1, C2, C3) was significantly higher than that in S_2 (30.56%, averaged combinations with C1, C2, C3) and S_3 (28.60%, averaged combinations with C1, C2, C3). This indicated that decomposition rates of

cellulose in S3 was higher than that in S1, and could be correlated to the mushroom yield where S3 produced the highest productivity, followed by S2 and S1. Chang and Wasser (2017) stated that substrate decomposition was started from the composting process (Phase I fermentation). This stage is vital for mushroom cultivation because, in this stage, complex organic carbon of substrates is decomposed to simpler forms that will be available to mushroom growth. Humic acid is one important product of EFB transformation during fermentation process (Amir et al. 2010).

Table 6 Effects of size reduction and composting duration of EFB

on chemical composition of spent EFB in the first experiment

| Treatment Combination | Cellulose (%) | Hemicelluloses (%) | Lignin (%) | |
|-----------------------|--------------------------|----------------------|----------------------|--|
| S1C1 | $33,42 \pm 3,15^{a}$ | $20,44 \pm 0,92^{a}$ | $19,82 \pm 1,28^{a}$ | |
| S2C1 | $30,00 \pm 0,23^{b}$ | $19,17 \pm 1,03^{a}$ | $19,08 \pm 0,10^{a}$ | |
| S3C1 | $28,28 \pm 2,81^{b}$ | $18,88 \pm 1,45^{a}$ | 19,23± 0,31ª | |
| S1C2 | $31,96 \pm 4,32^{a}$ | $19,60 \pm 0,45^{a}$ | $20,84 \pm 2,86^{a}$ | |
| S2C2 | $30,54 \pm 3,49^{b}$ | $18,73 \pm 0,25^{a}$ | $19,22 \pm 1,18^{a}$ | |
| S3C2 | 28,69± 1,33 ^b | $19,47 \pm 0,38^{a}$ | $19,80 \pm 0,70^{a}$ | |
| S1C3 | $34,29 \pm 1,92^{a}$ | $19,54 \pm 0,43^{a}$ | $19,88 \pm 0,51^{a}$ | |
| S2C3 | $31,15 \pm 1,19^{b}$ | $19,25 \pm 0,43^{a}$ | $20,04 \pm 1,03^{a}$ | |
| S3C3 | $28,83 \pm 1,10^{b}$ | $19,33 \pm 0,99^{a}$ | $19,95 \pm 0,67^{a}$ | |
| Average | 30,79± 2,12 | $19,38 \pm 0,40$ | $19,76 \pm 0,54$ | |

*) means with different letters are significantly different at p<0.05

In the second experiment, ANOVA analyses showed that interaction effect between N and O factors on cellulose and hemicelluloses contents of the spent EFB were significant (p < 0.05), but not significant on lignin (Table 7). Combinations of N1O2, N2O2, N3O2, N2O3, N3O1, and N3O3 (label d) resulted in lower contents of cellulose and hemicelluloses. This could be interpreted that decomposition rates of the EFB in those treatments were higher than in the other combinations. For lignin, none of the combinations was significantly different. If compared to the first experiment, all cellulose, hemicelluloses lignin contents of spent EFB were notably lower. Chang and Wasser (2017) state that breakdown of raw ingredients by microorganisms in fermentation process is more dynamics in conducive environment. Mohamed et al. (2016) state that in the fermentation process, microorganisms propagate and the nutritive substances of the substrate are accumulated in form of protein and other useful compounds. Therefore, effects of fertilizer supplement to the EFB substrate on improving fermentation dynamics in this research was quite clear.

Table 7 Effects of fertilizer supplements on chemical composition of spent EFB

| in the second experiment | | | |
|--------------------------|--------------------------|-------------------------|--------------------------------|
| Treatment Combination | Cellulose (%) | Hemicelluloses (%) | Lignin (%) |
| N101 | $25.91{\pm}~1.39^{ab}$ | 16.08 ± 0.98^{a} | 12.82 ± 2.71^{a} |
| N2O1 | $25.32{\pm}~1.72^{ab}$ | 14.35 ± 1.24^{abc} | $14.37{\pm}2.13^{a}$ |
| N3O1 | 20.90 ± 1.05^{cd} | $11.58{\pm}~0.06^{d}$ | 11.64 ± 2.10^{a} |
| N1O2 | $20.57{\pm}~1.93^{d}$ | 12.26 ± 1.49^{cd} | 12.62 ± 1.82^{a} |
| N2O2 | $23.71{\pm}~1.55^{abcd}$ | 13.46 ± 1.12^{bcd} | $12.47{\pm}~1.02^{a}$ |
| N3O2 | $24.04{\pm}~2.89^{abcd}$ | 13.63 ± 2.72^{abcd} | 12.52 ± 2.27^{a} |
| N1O3 | $26.91{\pm}~0.71^{a}$ | 15.33 ± 0.88^{ab} | $15.63{\pm}~0.40^{\mathrm{a}}$ |
| N2O3 | $20.89{\pm}~1.01^{cd}$ | $11.39{\pm}~0.74^{d}$ | 12.26 ± 1.34^{a} |
| N3O3 | $23.36{\pm}\ 2.18^{bcd}$ | 13.72 ± 1.34^{abcd} | 13.00 ± 1.69^{a} |
| Average | 23.51 ± 2.32 | 13.53 ± 1.60 | 13.04 ± 1.22 |

*) means with different letters are significantly different at p<0.05

30 Chemical composition in the spent EFB was the rest after fermentation, pasteurization, and 31 mushroom growth. But cellulose, hemicelluloses, and lignin all mainly changed during the EFB 32 fermentation (Wan Razali, et al. 2012). In the second experiment, on the averages, cellulose decreased

by 39.94%, hemicelluloses by 37.46%, and lignin by 45.94% from the initial status. According to Bledzki and Gassan (1999), hemicellulose is the weakest component because the structures are made of polysaccharides, amorphous structures, containing monomer sugar, and are subject to the most extensive degradation. While lignin resists biodegradation and only undergoes partial biotransformation (Jouraiphy et al. 2005). Even known as the hardest component to be degraded, lignin had the highest decomposition percentage among the other two components. This may be because initial lignin content of EFB in this study was relatively high.

Final contents of cellulose, hemicelluloses, and lignin in the spent EFB were on the averages 23,51±2,32%, 13,53±1.60%, and 13.04±1.22% respectively. The mushroom productivity (the BCE) as mentioned before was quite low if compared to what Thiribhuvanamala et al. (2012) reported. Our hypothesis is that the straw mushroom productivity can be increased by means of further lowering the final contents of cellulose, hemicelluloses, and lignin in the spent EFB. One potential method may be by doing better composting process of EFB, but this may not be a simple task. Wan Razali, et al. (2012) found cellulose, hemicelluloses, and lignin contents of 26.2%, 15.4%, and 33.3% respectively in the final compost of EFB fiber mixed with supplemental materials after 3 month composting. Contents of the three components; however, are still higher than in this study. Apetorgbor et al. (2015) also found higher contents of those components in the spent oil palm fruit fiber. Rice straw and cotton waste, which are known as the best substrates for straw mushroom, had much lower final lignin contents (4.8% and 0.2% respectively) as reported by Apetorgbor et al. (2015). Some methods, such using supplemental micronutrients (Thiribhuvanamala et al.2012), adequate fertilizers (Reyes 2000), mixed materials with low C-N ratios (Zakhary et.al. 1984), better aeration and drainage (Apetorgbor et.al. 2015), can be potential used to improve EFB composting techniques. Usage of laccase producing Trichoderma to breakdown lignin during substrate fermentation as reported by Bagewadi et al. (2017) was also potential option to apply.

26 Conclusion

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28 Finding in this research suggested that EFB did not need to be cut into smaller pieces if utilized as straw mushroom substrate. Aeration and drainage in composting process of the substrate and in the growing bed (in addition to O₂, CO₂, temperature, and humidity of the environment) may need to be given extra attention. The EFB should not be composted for more than 8 days, because there was a tendency to decrease yield. Supplemental fertilizers increased decomposition rates of EFB, as well as yield and nutritive values of straw mushroom. The fact that BCE was still considered low compared to other methods, there are still room for improvements in straw mushroom cultivation using EFB, such as by increasing dosages of fertilizer, supplementing with micronutrients, using mixed substrates, or incorporating enzyme-producing agents capable of stimulating delignification process.

36 Acknowledgment

We greatly appreciate the support of the Indonesian Ministry of Research, Technology and Higher
Education for the funding this research. We are grateful for the tireless contributions of the following
individual: Windri Meiawan, Herza Wirasaputra, Aditya Hari Prabowo, Muhammad Muslihudin, Linda
Fauziah, and Dian Nova Ayu Pulung.

Compliance with ethical standards

43 Conflict of interest: The authors declare that there is no conflict of interest with this research.

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View Letter

| Date: | 13 Jan 2019 |
|-------|--|
| To: | "Sugeng Triyono" striyono2001@yahoo.com |
| cc: | "Agus Haryanto" agusharyid65@gmail.com, "Mareli Telaumbanua" marelitelaumbanua@gmail.com, "Dermiyati Dermiyati" dermiyati.1963@fp.unila.ac.id, "Jamalam Lumbanraja" jlumbanraja53@gmail.com, "Filip To" fto@abe.msstate.edu |
| From: | "International Journal of Recycling of Organic Waste in Agriculture (IRWA)" harini.senthil@springernature.com |

Subject: Your Submission IRWA-D-18-00104R2

Dear Mr. Triyono,

We have received the reports from our advisors on your manuscript, "Cultivation of Straw Mushroom (Volvarealla volvacea) on Oil Palm Empty Fruit Bunch Growth Medium", submitted to International Journal of Recycling of Organic Waste in Agriculture

Based on the advice received, I have decided that your manuscript can be accepted for publication after you have carried out the corrections as suggested by the reviewer(s).

Below, please find the reviewers' comments for your perusal. You are kindly requested to also check the website for possible reviewer attachment(s).

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Thank you very much.

With kind regards, Payam Najafi Director – in – Chief International Journal of Recycling of Organic Waste in Agriculture

COMMENTS FOR THE AUTHOR:

Reviewer #1: I think the authors addressed most of the gueries satisfactorily.

Reviewer #2: The authors have tried to improve the text as per the suggestions of the reviewers. However, still there are several points which need attention.

In abstract section, the method part has not been spelled out correctly.
 In keywords what is Arduino?

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(iv) Spelling mistake at page 7 i.e. Biological Bonversion instead of Biological Conversion Efficiency. Similar spelling mistake for physico-chemical, it has been spelled as phychochemical.

v) Data on mushroom production as per FAOSTAT 2016 is not accurate, please check.

vi) At page 10 it has been mentioned that inorganic fertilizer was added @ 25 g, 50 g and 75 g, while organic fertilizer @ 5 cc, 10 cc and 15 cc. This is in what quantity of substrate? vii) Effect of substrate size and composting period seems to be significant, however, it has been mentioned as non significant, please check.

vii) How substrate size reduction leads to increase in protein content? Please clarify.

ix) How composting period has not affected the final content of the cellulose, hemicellulose and lignin in spent EFB, any explanation?

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To: striyono2001@yahoo.com

Date: Monday, January 14, 2019, 11:53 AM GMT+7

CC: "Agus Haryanto" <u>agusharyid65@gmail.com</u>, "Mareli Telaumbanua" <u>marelitelaumbanua@gmail.com</u>, "Dermiyati Dermiyati" <u>dermiyati.1963@fp.unila.ac.id</u>, "Jamalam Lumbanraja" <u>jlumbanraja53@gmail.com</u>, "Filip To" <u>fto@abe.msstate.edu</u>

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Date: Saturday, March 16, 2019, 02:32 PM GMT+7

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Dear Mr. Triyono,

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However, before your paper can be forwarded to our Production Department, you are requested to make the corrections as suggested by Payam Najafi

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Thank you very much.

With kind regards, Payam Najafi Director – in – Chief International Journal of Recycling of Organic Waste in Agriculture

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