

1. Proses Submit

Journal of Research in Pharmacy

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Dear Suci Suciati,

Thank you for submitting your manuscript entitled "Evaluation of Cholinesterase Inhibitory Activity of Cassia Species" to Journal of Research in Pharmacy . Your manuscript will first be evaluated by the editors and if it meets the Journal's standards, will be forwarded to referees for scientific review. You will be able to follow the stage of your manuscript in the review process through the author center to which you will have access with your user name and password. You can use the author center for revisions and new submissions.

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2. Proses Review

Hasil Review Tahap 1

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Suggestions

1. Reviewer Comments

In the manuscript entitled "Evaluation of Cholinesterase Inhibitory Activity of Cassia Species" the results of some in vitro investigations have been presented. The importance of the selected topic is unquestionable, since Alzheimer's disease is a progressive neurodegenerative disorder, which is the most common cause of dementia, and the number of people suffering from AD is expected to increase each year in the lack of effective treatment. Medicinal plants, natural products in general represent a significant source of secondary metabolites with beneficial pharmacological effects including potential anticholinesterase activity.

Major points

What was the yield for ethanol extraction? What about fractions obtained after the liquid-liquid partition? Which is the fraction where most of the alkaloids are concentrated?
What do we know about the pharmacological activity of compounds other than alkaloids found in Cassia sp. ?

There are some grammatically incorrect and/or confusing sentences, which needs to be revised for e.g.:

Page 2, line 12: „These plants not only used as ornamental trees but also have great economic importance as tanning material as well as its utilization in traditional medicine.”

Page 2, line 16: „Metabolites reported from Cassia species include anthraquinone, terpenoid, xanthone, flavonoid as well as alkaloid which posses promising biological activity...”

Page 5, Conclusion section: „Cassine and spectraline were identified in the active fractions by LC-MS/MS may responsible for the cholinesterase inhibitory activity”

Some minor points:

Abstract, line 3: use „cholinesterase” instead of „Cholinesterase”

Abstract, line 13: delete „nad” and insert „and”

Section Plant collection, extraction and fractionation, penultimate line: delete „nad” and insert „and”

Page 5, Line 2: delete „nad” and insert „and”

2. Reviewer Comments

There are many spelling and grammatical errors that affect the intelligibility of the article. Errors on the abstract are marked. Others are recommended to be corrected by the authors.

Near about hundreds of Cassia species are present so the title of the manuscript can be revised according to the number of species studied or where it grows.

The source from which the last sentence of the first paragraph was taken in the introduction section, is incorrect. The allegation made in this sentence is not included in the stated source.

The reason for investigating the cholinesterase inhibitory effects of Cassia species and the hypothesis of the study should be explained.

The names of the Cassia species should be checked and given with the author abbreviations. Also the epithets of the species should be checked. Cassia spectabilis DC. is a synonym of Senna spectabilis (DC.) H.S. Irwin & Barneby and Cassia siamea Lam. is a synonym of Senna siamea (Lam.) H.S. Irwin & Barneby. Therefore, two of the study materials should be considered and discussed as belonging to a different genus.

In plants containing anthraquinones, the stoge conditions and storage period are important factors. Therefore, the statement "The leaves were air-dried for several days" in section 4.2 should be detailed.

It was written that the ethanolic extract was separated by a liquid-liquid partition with n-hexane, ethyl acetate and n-butanol. Which plants was this process applied to? Is the ethanol extract subjected to liquid extraction without dissolving it with any solvent? Parameters of the LC-QTOF-MS/MS system should be given. At section 2.2, more information about fragment patterns of fragmented ions is needed. Also base peak chromatograms of the extracts can be given as supplementary material.

Cholinesterase inhibitory effects of some Cassia species had been studied before. The discussion section should be enriched with reference to these studies. Also the contribution of anthraquinones, the major metabolites of Cassia species, to the pharmacological effect, should be discussed. Although the study is not a bioactivity-guided isolation, this term was used in the discussion section.

3. Reviewer Comments

The authors submitted a paper concerning the anticholinesterase activity of selected Cassia species. The publication is generally acceptable with minor changes.

Some minor issues are listed below:

The abstract should include only the purpose, the essence and the significant results of the study, No need for details such as calculation of inhibitory activity or the program used.

It should also be stated in the figure descriptions that the values given in Figure 1 and Figure 2 are the percent inhibition values. It should be clearly stated what the x and y axes represent.

As can be realized from figure 1, the percent inhibition values of each plant extract were first examined. Then, different fractions of the most active extract were prepared and their IC50 values were examined. As ethylacetate and n butanol are already very close to each other in terms of polarity, the results can be predicted to be similar. It was not understood why the authors had chosen these two nearby solvents.

If IC 50 values of each plant extract are not calculated separately, IC 50 values should not be given in abstract and text. Otherwise, it is not possible to make a homogeneous and objective comparison between the extracts. The recommendation for this is to give all IC50 values (together with the percent inhibition values) of different plant species in a table.

English grammar and language errors should be checked again in the whole article.

Editor : The herbarium number of the plant specimen as well as the name of the herbarium in which the plant material is kept should be provided.

Manuscript Information

Manuscript ID: MPJ-4095

Title in English: Evaluation of Cholinesterase Inhibitory Activity of Cassia Species

Small Title in English: No information entered

Authors: Suci Suciati¹, Erlinda Laali¹, Debora Poerwanto¹, Anita Hapsari¹, Lailatul Gifanda¹, Karma Rabgay², Wiwied Ekasari¹, Kornkanok Ingkaninan²

Institutions: ¹Faculty of Pharmacy, Universitas Airlangga, Pharmacognosy and Phytochemistry, Surabaya, Indonesia

²Faculty of Pharmaceutical Sciences, Naresuan University, Department of Pharmaceutical Chemistry and Pharmacognosy, Phitsanulok, Thailand

Keywords in English: cholinesterase inhibitor ; Alzheimer's disease ; cassia species ; Cassia spectabilis.

Manuscript Type: Research article

Processing Status: Major Revision

Score Sheet

1. Reviewer

Does the content and value of the work justify publication in Marmara Pharmaceutical Journal ?

After revision

Does the title of the manuscript reflect the contents of the study ?

Yes

Are the keywords sufficient and appropriate ?

Yes

Is the summary concise and informative?

Yes

Is the text divided appropriately according to the article type ?

Yes

Is the language adequate?

Yes

Are the nomenclature and scientific terminology correct?	Yes
Are the references complete and recent?	Yes
Are the figures tables and graphics necessary ?	Yes
Are the figures tables and graphics clear ?	Yes
Is the introduction part	sufficiently developed
Are the experimental procedures sound?	Yes
Is the results and discussion part	sufficiently developed
Is conclusion sufficient and correlated with the results ?	Yes

Is the information about the approval of ETHICAL COMMISSION presented ?	Not applicable
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2. Reviewer

Does the content and value of the work justify publication in Marmara Pharmaceutical Journal ?	Yes
Does the title of the manuscript reflect the contents of the study ?	No
Are the keywords sufficient and appropriate ?	No
Is the summary concise and informative?	No
Is the text divided appropriately according to the article type ?	No
Is the language adequate?	No

Are the nomenclature and scientific terminology correct?	No
Are the references complete and recent?	No
Are the figures tables and graphics necessary ?	Yes
Are the figures tables and graphics clear ?	No
Is the introduction part	NOT sufficiently developed
Are the experimental procedures sound?	Yes
Is the results and discussion part	sufficiently developed
Is conclusion sufficient and correlated with the results ?	No

Is the information about the approval of ETHICAL COMMISSION presented ?	Not applicable
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3. Reviewer

Does the content and value of the work justify publication in Marmara Pharmaceutical Journal ?	Yes
Does the title of the manuscript reflect the contents of the study ?	Yes
Are the keywords sufficient and appropriate ?	Yes
Is the summary concise and informative?	Yes
Is the text divided appropriately according to the article type ?	Yes
Is the language adequate?	Yes
Are the nomenclature and scientific terminology correct?	Yes

Are the references complete and recent?	Yes
Are the figures tables and graphics necessary ?	Yes
Are the figures tables and graphics clear ?	No
Is the introduction part	sufficiently developed
Are the experimental procedures sound?	Yes
Is the results and discussion part	sufficiently developed
Is conclusion sufficient and correlated with the results ?	Yes
Is the information about the approval of ETHICAL COMMISSION presented ?	Not applicable

Tanggapan Terhadap Komenta Reviewers

Response to Reviewers Comments 1

Manuscript Title: Evaluation of Cholinesterase Inhibitory Activity of Six Indonesian *Cassia* Species

REVIEWER 1

Reviewer Comments	Response
What was the yield for ethanol extraction? What about fractions obtained after the liquid-liquid partition?	The yields of extracts and fractions have been added in the revised manuscript section 4.2 plant collection, extraction, and fractionation
Which is the fraction where most of the alkaloids are concentrated?	Based on TLC and LC-MS/MS data both ethyl acetate and n-butanol fractions contain alkaloid, however, quantitation of the alkaloid were not undertaken in this study.
What do we know about the pharmacological activity of compounds other than alkaloids found in <i>Cassia</i> sp.?	The pharmacological activities of compounds from <i>Cassia</i> spp have been stated in the original manuscript (introduction section).
There are some grammatically incorrect and/or confusing sentences, which needs to be	Grammatical errors have been revised (yellow colour texts)
Page 2, line 12: „These plants not only used as ornamental trees but also have great economic importance as tanning material as	sentence has been revised to “ <i>Cassia</i> spp are usually grown as ornamental plants, but many of these plants have great economic

well as its utilization in traditional medicine.”	importance as well as its utilization in traditional medicine.”
Page 2, line 16: „Metabolites reported from Cassia species include anthraquinone, terpenoid, xanthone, flavonoid as well as alkaloid which posses promising biological activity...”	sentence has been revised to” Metabolites reported from <i>Cassia</i> species include anthraquinone, terpenoid, xanthone, flavonoid as well as alkaloid. Many of these compounds posses promising biological activities, such as antioxidant, anti-inflammatory, anticancer, antiplasmodial and hepatoprotective agents”
Page 5, Conclusion section: „Cassine and spectaline were identified in the active fractions by LC-MS/MS may responsible for the cholinesterase inhibitory activity”	sentence has been revised to ” Piperidine alkaloids, cassine and spectaline, which were identified in the active fractions, may contribute to the cholinesterase inhibitory activity of <i>C. spectabilis</i> .
Some minor points: Abstract, line 3: use „cholinesterase” instead of „Cholinesterase” Abstract, line 13: delete „nad” and insert „and” Section Plant collection, extraction and fractionation, penultimate line: delete „nad” and insert „and” Page 5, Line 2: delete „nad” and insert „and”	Words have been revised

REVIEWER 2

Reviewer Comments	Response
There are many spelling and grammatical errors that affect the intelligibility of the article. Errors on the abstract are marked. Others are recommended to be corrected by the authors.	Grammatical errors have been revised (yellow colour texts)
Near about hundreds of Cassia species are present so the title of the manuscript can be revised according to the number of species studied or where it grows.	Title of manuscript has been revised to “Evaluation of Cholinesterase Inhibitory Activity of Six Indonesian <i>Cassia</i> Species”
The source from which the last sentence of the first paragraph was taken in the introduction section, is incorrect. The allegation made in this sentence is not included in the stated source.	The reference was for the previous sentence “This disease commonly occurs in elderly ages, and the onset mostly appears in the mid 60’s population” therefore reference was placed after this sentences
The reason for investigating the cholinesterase inhibitory effects of Cassia species and the hypothesis of the study should be explained.	Background of the study has been incorporated in the revised mancript (introduction section, last paragraph)

<p>The names of the <i>Cassia</i> species should be checked and given with the author abbreviations. Also the epithets of the species should be checked. <i>Cassia spectabilis</i> DC. is a synonym of <i>Senna spectabilis</i> (DC.) H.S.Irwin & Barneby and <i>Cassia siamea</i> Lam. is a synonym of <i>Senna siamea</i> (Lam.) H.S.Irwin & Barneby. Therefore, two of the study materials should be considered and discussed as belonging to a different genus.</p>	<p>The name of <i>Cassia</i> species have been revised in the results and discussion. Since <i>Senna siamea</i> is the synonym of <i>Cassia siamea</i> and <i>Senna spectabilis</i> is synonym of <i>Cassia spectabilis</i>, therefore author prefer to keep those two plants in the name of <i>Cassia</i>.</p>
<p>In plants containing anthraquinones, the stoge conditions and storage period are important factors. Therefore, the statement "The leaves were air-dried for several days" in section 4.2 should be detailed.</p>	<p>Leaves of <i>Cassia</i> spp were air dried for 7 days at room temperature. This information has been added in the revised manuscript (section 4.2)</p>
<p>It was written that the ethanolic extract was separated by a liquid-liquid partition with n-hexane, ethyl acetate and n-butanol. Which plants was this process applied to?</p>	<p>This process was applied to <i>C. spectabilis</i> extract. This information has been added in the manucript (section 4.2)</p>
<p>Is the ethanol extract subjected to liquid extraction without dissolving it with any solvent?</p>	<p>Extract was dissolved in ethanol:water (1:1) before partition with n-hexane, ethyl acetate and n-butanol. This information has been added in the revised manuscript (section 4.2)</p>
<p>Parameters of the LC-QTOF-MS/MS system should be given</p>	<p>Parameters of the LC-MS/MS study have been revised</p>
<p>At section 2.2, more information about fragment patterns of fragmented ions is needed. Also base peak chromatograms of the extracts can be given as supplementary material.</p>	<ul style="list-style-type: none"> • Fragmentations have been added in Table 2 LC-MS/MS data for identified alkaloids (1-3) from <i>C. spectabilis</i> extract • ESI-MS chromatogram and ESI-MS/MS chromatograms have been added as supplementary material
<p>Cholinesterase inhibitory effects of some <i>Cassia</i> species had been studied before. The discussion section should be enriched with reference to these studies. Also the contribution of anthraquinones, the major metabolites of <i>Cassia</i> species, to the pharmacological effect, should be discussed.</p>	<ul style="list-style-type: none"> • Cholinesterase of several cassia species as well as metabolites derived from <i>Cassia</i> have been added in the revised manuscript (Introduction section). • Authors prefer to keep the discussion section focus on <i>C. spectabilis</i> since several alkaloid and derivatives from <i>C. spectabilis</i> have also been reported for AChE inhibitory activity.
<p>Although the study is not a bioactivity-guided isolation, this term was used in the discussion section.</p>	<p>The word "bioactivity-guided" was removed., and sentence was revise to "Further fractionation on <i>C. spectabilis</i> extract suggested the ethyl acetate and the <i>n</i>-butanol fractions gave better</p>

	inhibitory activity compared to the <i>n</i> -hexane fraction”
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REVIEWER 3

Reviewer Comments	Response
The abstract should include only the purpose, the essence and the significant results of the study, No need for details such as calculation of inhibitory activity or the program used.	Authors prefer to keep detail of the method used in the abstract
It should also be stated in the figure descriptions that the values given in Figure 1 and Figure 2 are the percent inhibition values. It should be clearly stated what the x and y axes represent.	<ul style="list-style-type: none"> • Title of Figures 1 and 2 have been revised “Figure 1. %Inhibition of <i>Cassia</i> spp extracts and control (galantamine) against AChE and” and “Figure 2. %Inhibition of <i>C. spectabilis</i> fractions and control (galantamine) against AChE and BchE • The x and y title have been added in the revised manucrypt (Figures 1 and 2)
As can be realized from figure 1, the percent inhibition values of each plant extract were first examined. Then, different fractions of the most active extract were prepared and their IC50 values were examined. As ethylacetate and n butanol are already very close to each other in terms of polarity, the results can be predicted to be similar. It was not understood why the authors had chosen these two nearby solvents.	The polarity of the two solvents are close to each other however their selectivity are different. This can be seen in the LC-MS/MS results, several compounds were not seen in the n-butanol fraction
If IC 50 values of each plant extract are not calculated separately, IC 50 values should not be given in abstract and text. Otherwise, it is not possible to make a homogeneous and objective comparison between the extracts. The recommendation for this is to give all IC50 values (together with the percent inhibition values) of different plant species in a table.	All samples were screened for its cholinesterase inhibitory activity at 100 µg/mL. Authors only selected active extract with %inbition >50% (at 100 µg/mL) for IC50 assay. Comparison of the inhibitory activity of the extracts was based on the %inhibition data.
English grammar and language errors should be checked again in the whole article	English grammar and languange errors have been revised

EDITOR COMMENTS

Editor Comments	Response
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The herbarium number of the plant specimen as well as the name of the herbarium in which the plant material is kept should be provided.

Samples specimen numbers have been added as well as the identification letter number.



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Thu, Feb 13, 2020, 2:58 PM ☆ ↶

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This is to acknowledge receipt of your revised manuscript entitled Evaluation of Cholinesterase Inhibitory Activity of Cassia Species. You can follow the stage of your manuscript in the review process through the author center. Thank you.

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Hasil Review tahap 2

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Suggestions

1. Reviewer Comments

Detailed information about the method should be removed from the abstract. Instead, the abstract should include the LC-MS/MS results and the names of the identified compounds which is one of the most important parts of the article.

While discussing the LC-MS/MS results, the differences of the two similar solvent fractions used should be adequately explained. It is stated that there are different minor substances in the ethyl acetate fraction, but only one of these minor substances is explained.

2. Reviewer Comments

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Manuscript Information

Manuscript ID: MPJ-4095.REV-1

Title in English: Evaluation of Cholinesterase Inhibitory Activity of Cassia Species

Small Title in English: No information entered

Authors: Suci Suciati¹, Erlinda Laili¹, Debora Poerwantoro¹, Anita Hapsari¹, Lailatul Gifanda¹, Kama Rabgay², Wiwied Ekasari¹, Kornkanok Ingkaninan²

Institutions: ¹Faculty of Pharmacy, Universitas Airlangga, Pharmacognosy and Phytochemistry, Surabaya, Indonesia

²Faculty of Pharmaceutical Sciences, Naresuan University, Department of Pharmaceutical Chemistry and Pharmacognosy, Phitsanulok, Thailand

Keywords in English: cholinesterase inhibitor ; Alzheimer's disease ; cassia species ; Cassia spectabilis.

Manuscript Type: Research article

Processing Status: Minor Revision

Tanggapan terhadap Komentar Reviewers

Response to Reviewers Comments 2

Manuscript Title: Evaluation of Cholinesterase Inhibitory Activity of Six Indonesian *Cassia* Species

REVIEWER

Reviewer Comments	Response
Detailed information about the method should be removed from the abstract. Instead, the abstract should include the LC-MS/MS results and the names of the identified compounds which is one of the most important parts of the article.	Detailed information of the method has been removed. Author added sentence “ The chemistry of the active fractions was studied by LC-MS/MS method. ” Information of the LC-MS/MS results of two fractions was added, the name of the compounds have been stated.
While discussing the LC-MS/MS results, the differences of the two similar solvent fractions used should be adequately explained. It is stated that there are different minor substances in the ethyl acetate fraction, but only one of these minor substances is explained.	Several minor peaks were present in the ethyl acetate fraction, however only one minor compound can be identified properly, based on LC-MS/MS data. Information was added “ In the ethyl acetate fraction, a peak at RT 6.95 was observed, which was identified as 3-O-acetylspectaline (3). Other minor peaks can not be identified unambiguously ”

Journal of Research in Pharmacy : Confirmation for revised manuscript > Inbox x



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Sun, Apr 5, 2020, 12:08 PM ☆ ↶

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Dear Suci Suciati,

This is to acknowledge receipt of your revised manuscript entitled Evaluation of Cholinesterase Inhibitory Activity of *Cassia* Species. You can follow the stage of your manuscript in the review process through the author center. Thank you.

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3. Acceptance Letter


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Suggestions

Accepted

Manuscript Information

Manuscript ID:	MPJ-4095.REV-7
Title in English:	Evaluation of Cholinesterase Inhibitory Activity of Cassia Species
Small Title in English:	No information entered
Authors:	Suci Suciati ¹ , Erlinda Laili ¹ , Debora Poerwantoro ¹ , Anita Hapsari ¹ , Lailatul Gifanda ¹ , Karma Rabgay ² , Wiwied Ekasari ¹ , Kornkanok Ingkaninan ²
Institutions:	¹ Faculty of Pharmacy, Universitas Airlangga, Pharmacognosy and Phytochemistry, Surabaya, Indonesia ² Faculty of Pharmaceutical Sciences, Naresuan University, Department of Pharmaceutical Chemistry and Pharmacognosy, Phitsanulok, Thailand
Keywords in English:	cholinesterase inhibitor ; Alzheimer's disease ; cassia species ; Cassia spectabilis.
Manuscript Type:	Research article
Processing Status:	Accepted

4. Artikel Diterbitkan


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Evaluation of cholinesterase inhibitory activity of six Indonesian Cassia species

Suciati SUCIATI¹, Erlinda Rhoematul LAILI¹, Debora POERWANTORO¹, Anita Probo HAPSARI¹, Lailatul Zakiyah GIFANDA¹, Karma RABGAY², Wiwied EKASARI¹, Kornkanok INGGANINAN¹

¹Department of Pharmacognosy and Phytochemistry, Faculty of Pharmacy, Universitas Airlangga, Surabaya 60115, East Java, Indonesia
²Center for Natural Product Medicine Research and Development, Institute of Tropical Diseases, Universitas Airlangga, Surabaya 60115, East Java, Indonesia
³Bioscreening Unit, Department of Pharmaceutical Chemistry and Pharmacognosy, Faculty of Pharmaceutical Sciences and Center of Excellence for Innovation in Chemistry, Naresuan University, Phitsanulok, Thailand

DOI : 10.35333/jrp.2020.195

Alzheimer's disease (AD) is a neurodegenerative disorder, which is the most common cause of dementia. The aging population means that the number of people suffering from AD is expected to increase each year if there is no effective