Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Bukti Korespondensi

Judul Artikel: Isolation and Evaluation of Pharmacological Activities of a Bioactive Hydroxylated C28 Steroid from the Leaf of Laportea decumana (Roxb.) Wedd.

No	Proses	Tanggal
1	Artikel di ajukan ke jurnal *Tropical Journal of Natural Product Research*	22-4-2022
2	Artikel dalam proses review	23-4-2022
3	Revisi pertama: *accepted with moderate corrections*	16-5-2022
4	Editor mengirim hasil review artikel • Review form	28-6-2022
5	Author mengirimkan hasil revisi artikel • Response to reviewer • Plagiarism check • Revised article	29-6-2022
6	Revisi kedua: Minor revisions dan permohonan *English editing and proper formatting*	17-7-2022
7	Author mengirimkan hasil revisi artikel • Response to reviewer • Plagiarism check • Revised article	22-7-2022
8	Manajer editor mengirimkan *galley proof*	31-7-2022
9	Author mengirimkan kembali kepada manajer editor hasil revisi *galley proof*	1-8-2022
10	Artikel dipublikasi pada *Tropical Journal of Natural Product Research*. Vol. 6. Issue 7. Agustus 2022	3-8-2022

1. Artikel di ajukan ke jurnal *Tropical Journal of Natural Product Research*

(22-4-2022)

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Submited Article

apt. Rollando , S.Farm, M.Sc. <ro.llando@machung.ac.id>
Fri 4/22/2022 11:11 AM

To:Editor-in-Chief Tjnpr <editor.tjnpr@gmail.com>;Managing Editor TJNPR <p.editor.tjnpr@gmail.com>
Cc:apt. Rehmadanta Sitepu, S.Farm., M.Si. <rehmadanta.sitepu@machung.ac.id>

6 attachments (1 MB)
Abstract.docx; Authors Contributor.docx; Cover latter.doc; DECLARATION AND COPYRIGHT TRANSFER FORM.docx; Potential Reviewer.docx; Submited Manuscript_Rollando.docx;

Dear Prof. Abiodun Falodun, PhD

I am herewith submitting the manuscript entitled "BIOACTIVE HYDROXYLATED C28 STEROID FROM THE LEAF OF LAPORTEA DECUMANA (ROXB.) WEDD." for publication in Tropical Journal of Natural Product Research.
The manuscript discusses about the finding of steroid compound from laportea decumana leaves. We first found the steroid C28 compound from this plant. The compound have high antioxidant, anticancer dan antibacterial activity.

The manuscript has not been currently submitted for review to any other journal and will not be submitted elsewhere before a decision is made by this journal. We look forward for your positive response.

Kind regards

Rollando, M.Sc.
Program of Pharmacy
Ma Chung University
65151 Malang
Indonesia

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Re: Submited Article

Editor-in-Chief Tjnpr <editor.tjnpr@gmail.com>
Sat 4/23/2022 6:27 AM
To:apt. Rollando , S.Farm, M.Sc. <ro.llando@machung.ac.id>

Thank you for your submission to the Tropical Journal of Natural Product Research (www.tjnpr) https://www.scopus.com/sourceid/21100933230 SCOPUS , published by the University of Benin and Natural Product Research Group.

The peer-review process will commence immediately, as the manuscript will be passed to an editor for initial assessment as soon as possible.

Title: ***BIOACTIVE HYDROXYLATED C28 STEROID FROM THE LEAF OF LAPORTEA DECUMANA (ROXB.) WEDD.***

Best regards

Abiodun

Professor Abiodun Falodun, PhD

Editor-in-Chief:
Tropical Journal of Natural Product Research (TJNPR)
Head, Natural Product Research Group, University of Benin
Email:editor.tjnpr@uniben.edu; editor.tjnpr@gmail.com
www.tjnpr.org **SCOPUS, SCImago SJR Q4 0.13**
https://www.scopus.com/sources.uri

Professor of Pharmaceutical Chemistry
Fellow, Fulbright (USA)
Deputy Vice-Chancellor (Academic) 2014-2016
Faculty of Pharmacy
University of Benin
Phone: +234-807-318-4488;
email: faloabi@uniben.edu; abiodun.falodun@fulbrightmail.org
Google Scholar Citations
SCOPUS https://www.scopus.com/authid/detail.uri?

https://orcid.org/0000-0003-2929-3305authorId=12794326500#top

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2. Artikel dalam proses review

(23-4-2022)

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Manuscript Under Peer-Review Process

Editor-in-Chief Tjnpr <editor.tjnpr@gmail.com>
Sat 4/23/2022 1:55 PM
To:apt. Rollando , S.Farm, M.Sc. <ro.llando@machung.ac.id>
Cc:cd.kurniawan@gmail.com <cd.kurniawan@gmail.com>;apt. Rehmadanta Sitepu, S.Farm., M.Si. <rehmadanta.sitepu@machung.ac.id>

The manuscript submitted to the Tropical Journal of Natural Product Research https://www.scopus.com/sourceid/21100933230 SCOPUS by the corresponding author is undergoing the peer-review process.

Title: BIOACTIVE HYDROXYLATED C28 STEROID FROM THE LEAF OF LAPORTEA DECUMANA (ROXB.) WEDD.

Journal: Tropical Journal of Natural Product Research www.tjnpr.org

Corresponding Author:Rollando Rollando

Co-authors: Cristopher Daniel Kurniawan, Rehmadanta Sitepu

Manuscript No: TJNPR FB282ARN

If you have any objections, please contact the editorial office as soon as possible. If we do not hear from you, we will assume you agree with your co-authorship.

If you did not co-author this submission, please contact the corresponding author directly

Thank you very much.

Best regards

Abiodun

Professor Abiodun Falodun, PhD

Editor-in-Chief:
Tropical Journal of Natural Product Research (TJNPR)
Head, Natural Product Research Group, University of Benin
Email:editor.tjnpr@uniben.edu; editor.tjnpr@gmail.com
www.tjnpr.org **SCOPUS, SCImago SJR Q4 0.13**
https://www.scopus.com/sources.uri

Professor of Pharmaceutical Chemistry
Fellow, Fulbright (USA)
Deputy Vice-Chancellor (Academic) 2014-2016
Faculty of Pharmacy
University of Benin
Phone: +234-807-318-4488;
email: faloabi@uniben.edu; abiodun.falodun@fulbrightmail.org
Google Scholar Citations
SCOPUS https://www.scopus.com/authid/detail.uri?

https://orcid.org/0000-0003-2929-3305authorId=12794326500#top

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3. Revisi pertama: accepted with moderate corrections

(16-5-2022)

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Editorial decision on manuscript submitted for publication in TJNPR

Editor-in-Chief Tjnpr <editor.tjnpr@gmail.com>
Mon 5/16/2022 1:15 AM
To:apt. Rollando , S.Farm, M.Sc. <ro.llando@machung.ac.id>

1 attachments (182 KB)
Provisional acceptance 282.pdf;

Dear Dr. Rollando,

The manuscript submitted to the Tropical Journal of Natural Product Research www.tjnpr.org Q4 https://www.scopus.com/sourceid/21100933230 has been carefully reviewed by competent experts.

Find attached the details of the decision.

Please send your response urgently to the editor-in-Chief, to enable us to process your manuscript for the next issue **Vol 6 issue 5, 2022**.
Kindly acknowledge the receipt of the mail.

Title: BIOACTIVE HYDROXYLATED C28 STEROID FROM THE LEAF OF Laportea Decumana (ROXB.) WEDD
Authors: Rollando Rollando*, Cristopher Daniel Kurniawan, Rehmadanta Sitepu

TJNPR Editorial Decision: accepts with moderate corrections

Congratulations

Best regards

Abiodun

Professor Abiodun Falodun, PhD

Editor-in-Chief:
Tropical Journal of Natural Product Research (TJNPR)
Head, Natural Product Research Group, University of Benin
Email:editor.tjnpr@uniben.edu; editor.tjnpr@gmail.com
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Professor of Pharmaceutical Chemistry
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Faculty of Pharmacy
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Phone: +234-807-318-4488;
email: faloabi@uniben.edu; abiodun.falodun@fulbrightmail.org
Google Scholar Citations
SCOPUS https://www.scopus.com/authid/detail.uri?

https://orcid.org/0000-0003-2929-3305authorId=12794326500#top

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Tropical Journal of Natural Product Research
Official Journal of the Natural Product Research Group
Faculty of Pharmacy, University of Benin 300001, Benin City, Nigeria
Phone: +2348073184488, Email: editor.tjnpr@gmail.com; editor.tjnpr@uniben.edu; Website: www.tjnpr.org

ISSN: 2616-0684 (Print); 2616-0692 (Online), **DOI**: 10.26538/tjnpr, ISI IF: 0.562 (2017)
Abstracted/Indexed: Index Copernicus **ICV (2017): 59.83**, Open-J-Gate, EMBASE, EBSCO Host, WorldCat, AJOL, CrossRef (USA), JIF, NCBI (PubMed), CAS

SCOPUS indexed, SCImago SjR 0.13

Ref. No. 406801802214 DATE: 15th May, 2022

Pharmacy Department, Faculty of Science and Technology, Ma Chung University, Malang 65151, Indonesia

Dear Dr. Rollando,

Provisional Acceptance letter for Article Manuscript Number TJNPR FB282ARN

Title: BIOACTIVE HYDROXYLATED C28 STEROID FROM THE LEAF OF Laportea Decumana (ROXB.) WEDD

Authors: Rollando Rollando*, Cristopher Daniel Kurniawan, Rehmadanta Sitepu

I am pleased to inform you that your paper sent to the Tropical Journal of Natural Product Research has been reviewed and recommended for publication as a research article.

However, before the issues raised by the Reviewers are forwarded, to enable you revise your manuscript accordingly, please pay a publication charge of **$ USD270**. The actual publication of the paper will be in the upcoming issue (**Vol 6 issue 5, 2022**).

Please, the manuscript number (TJNPR ----) should be included in the bank transfer.

Congratulations.

The money should be remitted in favour of

Name of account:	Teamee Natural Product
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Account Number:	0248808386.
Sort Code:	058044128
Swift code:	GTBINGLA
Address of Bank:	Uselu Lagos Road, Benin City, Edo State, Nigeria

Sincerely,

Professor Abiodun Falodun
Editor-in-Chief

4. Editor mengirim hasil review artikel

(21-10-2021)

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Review Comments

Editor-in-Chief Tjnpr <editor.tjnpr@gmail.com>
Tue 6/28/2022 11:03 PM
To:apt. Rollando , S.Farm, M.Sc. <ro.llando@machung.ac.id>

2 attachments (138 KB)
BH1-TJNPR-2022-M121 Reviewer 1.pdf; BH2-TJNPR-2022-M121 Reviewer 2.pdf;

Review Comments (BIOACTIVE HYDROXYLATED C28 STEROID FROM THE LEAF OF LAPORTEA DECUMANA (ROXB.) WEDD)

Editorial comments to authors

Abstract: begin with a brief background and include a well stated aim in this section.

For in-text referencing, the superscript numerals should come after a comma or full-stop.

List of references should follow journal adopted format. See journal website at http://www.tjnpr.org/guideforauthors.aspx# **for details. Abbreviate all Journal names.**

All comments/corrections made by reviewers should be completely addressed, point by point, and make appropriate changes in the manuscript, or provide a suitable rebuttal to any specific request for change that has not been made.
All corrections/changes made in the manuscript should be **highlighted in yellow colour** when submitting the manuscript in the revised form on or before 30th June 2022
The authors should carefully revise and correct the manuscript taking into consideration the comments of all the reviewers. 50% of the references cited should be between 2016-2020. The revised and corrected manuscript should be subjected to plagiarism checker (17% allowed in TJNPR) and English language editing. Evidence of the checks should be attached when submitting the revised/corrected manuscript.

During submission of the revised manuscript include another file labelled "**Responses to reviewers' comments**" (a matrix) clearly showing your responses to each of the issues raised by the reviewers; mention the section, page and paragraph/lines where and how the changes/corrections have been made.

Strictly adhere to the author guidelines. Make sure that all the facts and information provided in the manuscript are correct. Check grammar, spelling, spacing, other information and facts including scientific names, formulae, symbols, equations, etc.

Ensure that all the references are correctly cited in the text and list. Verify all the references from their original sources. Confirm correctness of the citation info such as authors' names (surnames, initials, spelling, arrangements, etc), year, title, journal, volume, pages, punctuation, etc. The numbers and units must be presented according to the journal style. Use clearly distinguishable patterns for the illustrations/figures (e.g., graphs and charts) such that they should be legible even for black and white printing or when reduced in size.

Proofread the whole document after effecting all the corrections. The revised version should be approved by all the co-authors before submitting it.

A manuscript not complying with these and other instructions will not be processed and may be rejected.

Please find the attached review comments for your revisions.

Best regards

Abiodun

Professor Abiodun Falodun, PhD

Editor-in-Chief:
Tropical Journal of Natural Product Research (TJNPR)
Head, Natural Product Research Group, University of Benin
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Professor of Pharmaceutical Chemistry
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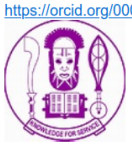

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REVIEW FORM

The Editorial Team of the Tropical Journal of Natural Product Research kindly request you to review the enclosed article. Please complete the form and return to the Editor-in-Chief, editor.tjnpr@gmail.com; editor.tjnpr@uniben.edu

A. MANUSCRIPT

Journal	**Tropical Journal of Natural Product Research**
Manuscript Number	TJNPR FB282AR
Type of paper	Original article
Title of paper	Bioactive hydroxylated c28 steroid from the leaf of laportea decumana (roxb.) wedd.
Name of Authors	Rollando Rollando

B. REVIEWER'S SPECIFIC COMMENTS PER SECTION OF MANUSCRIPT

Abstract	The abstract is written clearly, and ultimately, the research objectives have been reported.
Introduction	The introduction is well written and comprehensive.
Methodology	The entire research process is written in total, and the research steps are clear
Results	The research results follow the research methodology.
Discussion	The discussion is written in full. But English needs to be improved.
Conclusion	The conclusion has been written in full.
References	All references have been cited in the article.
Figures, Tables	Tables and figures are written correctly and have been cited in the results and discussion.

D. REVIEWER'S RECOMMENDATION
Please mark with "**X**" one of the options.

You state the article should:

Publish as it is	√
Accept with minor revisions (editor will check), specific comments to the editor below	√
Accept with moderate revisions as recommended by reviewer	
Accept with major corrections (the article should be thoroughly changed)	
Full article	√
Short communication	
Reject for reasons noted by the reviewer (please be specific)	

E. REVIEWER'S INFORMATION

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

REVIEW FORM

The Editorial Team of the Tropical Journal of Natural Product Research kindly request you to review the enclosed article. Please complete the form and return to the Editor-in-Chief, editor.tjnpr@gmail.com; editor.tjnpr@uniben.edu

A. MANUSCRIPT

Journal	**Tropical Journal of Natural Product Research**
Manuscript Number	tjnpr fb282ar
Type of paper	research article
Title of paper	Bioactive hydroxylated c28 steroid from the leaf of laportea decumana (roxb.) wedd.
Name of Authors	

B. REVIEWER'S SPECIFIC COMMENTS PER SECTION OF MANUSCRIPT

Abstract	Abstract written completely and structured.
Introduction	The introduction is written in full.
Methodology	All methods used to obtain data have been written in full.
Results	The research results are written in full. discussed properly.
Discussion	The discussion is well written and correct. The data obtained have been discussed well. The theoretical discussion is already related to the data.
Conclusion	Conclusion is written correctly
References	Complete bibliography
Figures, Tables	All tables and figures have been cited

D. REVIEWER'S RECOMMENDATION
Please mark with "**X**" one of the options.

You state the article should:

Publish as it is	X
Accept with minor revisions (editor will check), specific comments to the editor below	x
Accept with moderate revisions as recommended by reviewer	
Accept with major corrections (the article should be thoroughly changed)	
Full article	x
Short communication	
Reject for reasons noted by the reviewer (please be specific)	

E. REVIEWER'S INFORMATION

5. Author mengirimkan hasil revisi artikel

(29-6-2022)

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Re: Review Comments

apt. Rollando , S.Farm, M.Sc. <ro.llando@machung.ac.id>
Wed 6/29/2022 9:56 PM
To:Editor-in-Chief Tjnpr <editor.tjnpr@gmail.com>

4 attachments (2 MB)
Plagiarism Check 1.pdf; Responses to reviewers' comments.docx; Revised Manuscript_Rollando.docx; Proofread Certificate.jpg;

Dear Editor in Chief TJNPR

Here i send my revised article. I also attach the proof read certification, responses to reviewer comment, and plagiarism check.

Regards
Rollando

Prof. Abiodun Falodun

Editor in Chief

Tropical Journal of Natural Product Research

Dear Abiodun,

I would like to submit our revised manuscript entitles" **Bioactive Hydroxylated C28 Steroid from the Leaf of Laportea decumana (roxb.) Wedd."** for the consideration of publication in the tropical journal of natural product research.

I also have answered a point by point questions, comments and suggestions from the reviewer to improve our article quality as written along with this letter. I really appreciate for all these valuable comments and suggestions..

I hope very much that you would consider this manuscript for the publication in your esteemed journal. Your kind consideration would be gratefully acknowledged.

Thank you
Your sincerely

Rollando Rollando

Responses to reviewers' comments

The weakness is in writing in English. English must be improved (Reviewer 1 and 2).	We have fixed the grammatical error in the article. We have improved the English language. Proofread certificate already exists.
Begin with a brief background and include a well stated aim in this section (Editor).	The abstract section has added words and research objectives. Please check the changes on page 1 line 8-9 (Abstract). In the introduction, it has been fixed in the research objectives section on page 2 line 36-40.
For in-text referencing, the superscript numerals should come after a comma or full-stop.	We have fixed text referencing.
List of references should follow journal adopted format. See journal website at http://www.tjnpr.org/guideforauthors.aspx# for details. Abbreviate all Journal names.	We have fixed reference writing.
Ensure that all the references are correctly cited in the text and list.	We have fixed reference writing. All citations have been confirmed to be in the bibliography.
50% of the references cited should be between 2016-2020.	We've fixed the reference. In this article 96% references between 2016-2020

Plagiarism Checker X Originality Report

Similarity Found: 11%

Date: Wednesday, June 29, 2022
Statistics: 775 words Plagiarized / 4565 Total words
Remarks: Low Plagiarism Detected - Your Document needs Optional Improvement.

Bioactive Hydroxylated C28 Steroid from the Leaf of Laportea decumana (roxb.) Wedd. Rollando Rollando1*, Eva Monica1, Muhammad Hilmi Aftoni1, Cristopher Daniel Kurniawan1, Rehmadanta Sitepu1 1Pharmacy Department, Faculty of Science and Technology, Ma Chung University, Malang 65151, Indonesia ABSTRACT Cancer and infection have a high prevalence at this time, so they need to be treated.

The purpose of this study was to isolate the bioactive compounds in the leaves of Laportea decumana (roxb.). One new hydroxylated C28 steroid has been isolated from the leaf of L. decumana (roxb.) wedd., collected at Sorong, West Papua, Indonesia. The compound possess 9-(buta-1,3-dien-2-yl)-11-hydroxy-2,2a,2b,3,4,5,6,6a,7,8,8a1,9,9a,12,12a,14b-hexadecahydro-1H,11H,14H-13 oxacyclobuta[1,5]cyclopenta[1,2-a]cyclobuta[de]cyclopropa[b]chrysen-14-one.

The structure of hydroxylated C28 steroid were determined using UV/Vis, IR, MS, 1H-NMR, 13C-NMR, HMQC and HMBC. Molecular docking was conducted by using PyRx program. The compound was tested for the inhibitory effect on 4T1 cancer cells, 2,2-diphenyl-1-picrylhydrazyl radical, and two bacterias. The compound was active against 4T1 cancer cells and had the potential to develop as a cancer cell inhibitor.

Keywords: Laportea decumana, antioxidant, antibacterial, cytotoxic, molecular docking *Corresponding author. E mail: ro.llando@machung.ac.id Tel: +6282220379864 Introduction Laportea decumana (Roxb.) Wedd is Indonesia's native plant that has medicinal use. Growing vastly in Papua and Papua New Guinea, it has been used as an herbal remedy to cure pain and headache.1

MPWS
Rich Proofreading

CERTIFICATE OF PROOFREADING

THIS IS TO ACKNOWLEDGE THAT THE ARTICLE ENTITLED

Bioactive hydroxylated c28 steroid from the leaf of laportea decumana (roxb.) wedd

WRITTEN BY

ROLLANDO ROLLANDO

HAS BEEN PROOFREAD AND RETURNED TO THE CUSTOMER ON

29 JUNE 2022

.....

INTAN RAIHANA HUSIN
Chief Executive Officer
MPWS RICH RESOURCES SDN BHD

Bioactive Hydroxylated C28 Steroid from the Leaf of Laportea decumana (roxb.) Wedd.

Rollando Rollando[1*], Eva Monica[1], Muhammad Hilmi Aftoni[1], Cristopher Daniel Kurniawan[1], Rehmadanta Sitepu[1]

[1]Pharmacy Department, Faculty of Science and Technology, Ma Chung University, Malang 65151, Indonesia

ABSTRACT

Cancer and infection have a high prevalence at this time, so they need to be treated. The purpose of this study was to isolate the bioactive compounds in the leaves of *Laportea decumana* (roxb.). One new hydroxylated C28 steroid has been isolated from the leaf of *L. decumana* (roxb.) wedd., collected at Sorong, West Papua, Indonesia. The compound possess 9-(buta-1,3-dien-2-yl)-11-hydroxy-2,2a,2b,3,4,5,6,6a,7,8,8a1,9,9a,12,12a,14b-hexadecahydro-1H,11H,14H-13

HPLC. Both solvent optimization and preparative TLC were performed using silica gel 60 GF_{254}. Before using the plate, an activation procedure was conducted by heating the TLC plate at 115°C for 1 hour. Then, 20 mg/mL extract solution was applied to the activated preparative TLC plate. Next, the elution was done with n-hexane and ethyl acetate (4:1) to separate the triterpenoid compound. Finally, preparative HPLC with methanol mobile phase was used to purify the fraction. The isolate weight is 28,56 mg.

The antibacterial compound screening was done by placing an eluted plate on an agar medium containing bacterial culture. The contact between the plate and agar surface was kept for 3 hours before removing the plate. Then, the agar media was incubated at 37°C for 24 hours. A clear zone on the medium agar shows the position of an antibacterial compound on the TLC

Chemical structure elucidation

The UV/Vis spectrum of fraction 7 showed a single peak at 266 nm, indicating that the compound contains short chromophore chain. An absorbance on that wavelength gave information that the compound may contain chromophore having 2 or 3 π-bonds. Meanwhile, the IR spectrum showed that the compound has a hydroxyl group (3454.4 cm-1), carbonyl or olefinic group (1658.87 cm-1), and lactone group (1262.66; 1232.60; and 1115.10 cm-1). Other peaks on the IR spectrum showed that the compound is of alkane groups.[13]

The ^{13}C-NMR spectrum showed that the compound contains 28 carbons. It showed six olefinic atoms (C12, C22, C24, C25, C26, C27) detected on the chemical shift between 109.14 to 174.45 ppm. The carbon in 174.45 is responsible for the lactone group. Several carbon atoms were identified as neighbor of the olefinic carbon or electronegative group for having chemical shift of 63.61 (C11); 63.70 (C15); and 71.89 (C17) ppm. The DEPT spectrum showed no methyl

Acknowledgement

The authors would like to thank the The Ministry of Education, Culture, Research, and Technology of the Republic of Indonesia for support through "Penelitian Dasar Unggulan Perguruan Tinggi 2022" No. 045/SP2H/PT/LL7/2022.

References

1. Simaremare ES, Pratiwi RD, Rusnaeni R, Gunawan E, Dirgantara S. Utilization of itchy leaf plants (*Laportea decumana*) as anti fatigue drugs. Comm s jour. 2019; 3(4):97–103.
2. Simaremare ES, Souisa WV. Antihyperuricemic activity test of itchy leaf extract (*Laportea decumana* (roxb.) wedd) from papua. Pharm uho. 2021; 6(2):21–25.
3. Simaremare ES, Pratiwi RD, Gunawan E. Utilization of itchy leaf plants in arso XIV. Sem

6. Revisi kedua: Revisi kedua: Minor revisions dan permohonan *English editing and proper formatting*

(17-7-2022)

Dr. apt. Rollando, S.Farm., M.Sc.
Pengajuan ke Lektor Kepala

Revision to Your Manuscript

Managing Editor TJNPR <p.editor.tjnpr@gmail.com>
Sun 7/17/2022 10:23 AM
To:apt. Rollando , S.Farm, M.Sc. <ro.llando@machung.ac.id>
Cc:Editor-in-Chief Tjnpr <editor.tjnpr@gmail.com>

1 attachments (1 MB)
TJNPR-2022-M121 Revised.docx;

Abstract: include a summary of the methods before presenting the results.

State the meaning of all abbreviations at first mention.

Page 1, line 2; page 3, line 18; rephrase.

Page 5; include reference for equation and clarify; %IC50 or %inhibition?

Submit the Manuscript for English language Editing.

All corrections/changes made in the manuscript should be **highlighted in yellow colour** when submitting the manuscript in the revised form.

7. Author mengirimkan hasil revisi artikel

(22-7-2022)

MPWS
Rich Proofreading

CERTIFICATE OF PROOFREADING

THIS IS TO ACKNOWLEDGE THAT THE ARTICLE ENTITLED

Bioactive hydroxylated c28 steroid from the leaf of laportea decumana (roxb.) wedd

WRITTEN BY

ROLLANDO ROLLANDO

HAS BEEN PROOFREAD AND RETURNED TO THE CUSTOMER ON

29 JUNE 2022

.....

INTAN RAIHANA HUSIN
Chief Executive Officer
MPWS RICH RESOURCES SDN BHD

Bioactive Hydroxylated C28 Steroid from the Leaf of Laportea decumana (roxb.) Wedd.

Rollando Rollando[1*], Eva Monica[1], Muhammad Hilmi Aftoni[1], Cristopher Daniel Kurniawan[1], Rehmadanta Sitepu[1]

[1]Pharmacy Department, Faculty of Science and Technology, Ma Chung University, Malang 65151, Indonesia

ABSTRACT

Cancer and infections have increased very rapidly in the last decade. One way to treat cancer and infections can use medicinal plants. For example, Laportea decumana (roxb.) has long been used as a medicinal plant, but the scientific evidence on its pharmacological effects is minimal. The purpose of this study was to isolate the bioactive compounds in the leaves of *Laportea decumana* (roxb.). Isolation using preparative thin layer chromatography (TLC) and purification with preparative high-performance liquid chromatography (HPLC). The structure of compound determined using UV/Vis, IR, MS, ^{1}H-NMR, ^{13}C-NMR, HMQC and HMBC. Molecular docking was conducted by using PyRx program. One new hydroxylated C28 steroid has been isolated from the leaf of *L. decumana* (roxb.) wedd. The compound possess 9-(buta-1,3-dien-2-yl)-11-hydroxy-2,2a,2b,3,4,5,6,6a,7,8,8a1,9,9a,12,12a,14b-hexadecahydro-1H,11H,14H-13 oxacyclobuta[1,5]cyclopenta[1,2-a]cyclobuta[de]cyclopropa[b]chrysen-14-one. The compound was tested for the inhibitory effect on 4T1 cancer cells, 2,2-diphenyl-1-picrylhydrazyl radical, and two bacteria. The compound was active against 4T1 cancer cells and had the potential to develop as a cancer cell inhibitor.

Keywords: *Laportea decumana*, antioxidant, antibacterial, cytotoxic, molecular docking

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Original Research Article

Isolation and Evaluation of Pharmacological Activities of a Bioactive Hydroxylated C28 Steroid from the Leaf of *Laportea decumana* (Roxb.) Wedd

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ABSTRACT

Cancer and infection rates have risen significantly in the last decade. Medicinal plants can be used to treat cancer and infections. *Laportea decumana* (Roxb.) Wedd for example, has long been used as a medicinal plant, but scientific evidence on its pharmacological effects is limited. The present study was therefore aimed at isolating a bioactive compound from the leaves of *Laportea decumana* and evaluating its pharmacological activities. Ethanol was used to extract the leaves of *L. decu*mana using the maceration method. Preparative thin layer chromatography (TLC) was used for the isolation of an active compound, and preparative high-performance liquid chromatography (HPLC) was employed for purification. The structure of the active compound was determined by UV/Vis, IR, MS, ^{1}H-NMR, ^{13}C-NMR, HMQC, and HMBC. *Staphylococcus aureus* and *Escherichia coli* were used to examine the antibacterial activity of the compound. Furthermore, the antioxidant potential of the new compound was determined by the DPPH (2,2-diphenyl-1-picrylhydrazyl) assay. Molecular docking was performed with the PyRx program. Also, the compound was tested for its ability to inhibit 4T1 cancer cells. The results showed that the extraction process yielded an oily green, thick extract with a yield value of 5.19%. From the leaves of *L. decumana*, a new hydroxylated C28 steroid was isolated (MCI). The bioactive compound possessed antibacterial properties against the test organisms, as well as antioxidant activity by DPPH assay. Furthermore, MCI was active against 4T1 cancer cells. The findings of this study suggest that MCI has the potential as a cancer cell inhibitor.

Keywords: Antibacterial, Antioxidant, Cytotoxic, *Laportea decumana*, Molecular docking.

Introduction

Laportea decumana (Roxb.) Wedd is a medicinal plant native to Indonesia. It is widely grown in Papua and Papua New Guinea and has been used as a herbal remedy to treat pain and headaches.[1] It has potential pharmacological activity due to the presence of alkaloid, glycoside, and triterpenoid contents.[2] Previous studies found that *L. decumana* extract has analgesic and antibacterial properties.[3] The plant extract is a complex mixture of soluble compounds in the extractive solvent, including primary metabolites (molecular mass ≥ 2000 Dalton) and secondary metabolites (molecular mass < 2000 Dalton). Although secondary metabolites like alkaloid, flavonoid, coumarin, quinone, and terpenoid have important biological activities,[4] the presence of primary metabolites in extracts make a quantitative determination of the active compound dose impossible. The research on *L. decumana* pharmacological activity was only reported as an extract form in the literature study, so the active compound was still unknown.

Therefore, the present study was conducted to isolate a bioactive compound from the leaves of *Laportea decumana* and assess its pharmacological activities.

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Materials and Methods

Sources of chemicals

Silica gel F_{254}, Silica gel 60 PF_{254} containing gypsum, dimethyl sulfoxide (DMSO), methanol, chloroform, n-hexane, and ethyl acetate were obtained from Merck, United States. Dextrose, nutrient agar (NA), and Mueller Hinton were purchased from Oxoid, United Kingdom. RPMI 1640, fetal bovine serum, penicillin, streptomycin, fungizone, sodium bicarbonate, and L-glutamine were supplied by Gibco, United States. HEPES (4-[2-hydroxyethyl]-1-piperazineethanesulfonic acid) were purchased from Invitrogen, United States. DPPH (2,2-diphenyl-1-picrylhydrazyl) and MTT (3-[4,5-dimethylthiazol-2-yl]-2,5-diphenyltetrazolium bromide) were obtained from Sigma-Aldrich, United Statesμ.

Source of plant materials

The leaves of *Laportea decumana* were collected in January 2022 from Sorong, West Papua, Indonesia, and authenticated in the Department of Botany, University of Ma Chung, Indonesia. A voucher specimen number (FA:02-MACHUNG-2022) was assigned and the plant material was deposited in the herbarium unit of the department. Fresh leaves were washed and impurities were removed. The leaves were dried in an oven at 50°C for two days and then pulverized.[5]

*Extraction of Laportea decu*mana *leaves*

The extraction was accomplished through maceration at room temperature by soaking 150 g dried leaf sample in 1 L 96% ethanol for 24 hours. The filtered mixture was collected. To obtain a thicker filtrate, the obtained filtrate was evaporated in a rotary evaporator, which was followed by advanced evaporation in a water bath.[6]

*Isolation of active compounds from Laportea decu*mana *leaf extract*

Isolation of active compounds was divided into four stages: solvent optimization, bioautography, preparative thin layer chromatography (TLC), and purification with preparative high-performance liquid