# PEER REVIEW

**LEMBAR**
**HASIL PENILAIAN SEJAWAT SEBIDANG ATAU PEER REVIEW**
**KARYA ILMIAH : JURNAL ILMIAH**

Judul Jurnal Ilmiah (Artikel) : Short Commmmunication: Diversity of cellulolytic bacteria isolatet from coastal mangrove sediment in Logending Beach, Kemumen Indonesia.

Penulis Jurnal Ilmiah *) : 1 Hendro Pramono (*nama pengusul dicetak tebal)
**2** Afifah Mariana
3 Dini Ryandini
**4 Eming Sudiana**

Jumlah Penulis : 4

Status Penulis : Penulis Ke-4

Identitas Jurnal Ilmiah :

a. Nama Jurnal : BIODIVERSITAS Journal of Biological Diversity
b. Nomor ISSN : ISSN: P-1412-033x E-ISSN: 2085-4722
c. Edisi/Volume, Nomor : Edisi April 2021/Vol. 22 Nomor : 4
d. Penerbit : Fakultas MIPA Universitas Sebelas Maret
e. DOI artikel : https://doi.org/10.13057/biodiv/d220433
f. Alamat Web : https://smujo.id/biodiv/article/view/7036/4732
g. Terindeks di : SCOPUS, Q3, SJR : 0,27

Kategori Publikasi Jurnal Ilmiah : [√] Jurnal Ilmiah Internasional /Internasional Bereputasi
(beri v pada kategori yang tepat) [ ] Jurnal Ilmiah Nasional Terakreditasi
[ ] Jurnal Nasional/Nasional terindeks di..... *

Hasil Penilaian Peer Review :

| Komponen Yang Dinilai | Nilai Maksimal Jurnal Ilmiah | | | | | | Nilai Akhir Yang Diperoleh |
|---|---|---|---|---|---|---|---|
| | Internasional/ Internasional bereputasi | | Nasional Terakreditasi | | Nasional Terindeks *) | | |
| | 30 | | | | | | |
| a Kelengkapan unsur isi artikel (10%) | 10 % X 30= | 3 | | | | | 3,0 |
| b Ruang lingkup dan kedalaman pembahasan (30%) | 30 % X30= | 9 | | | | | 9,0 |
| c Kecukupan dan kemutahiran data/informasi dan metodologi (30%) | 30 % X30= | 9 | | | | | 9,0 |
| d Kelengkapan unsur dan kualitas terbitan/jurnal (30%) | 30 % X30= | 9 | | | | | 9,0 |
| **Total = (100%)** | **30** | | | | | | 29,8 |
| **Nilai Pengusul (40 % x Total)/3** | **4,00** | | | | | | 3,97 |

Catatan Penilaian artikel oleh Reviewer:

1. Tentang kelengkapan dan kesesuaian unsur : Unsur lengkap dan sesuai
2. Tentang ruang lingkup dan kedalaman pembahasan : Ruang Lingkup Sangat baik, spesifik, pembahasan sedang, cukup mendalam
3. Kecukupan dan kemutahiran data serta metodologi : Data cukup mutahir, metodologi sesuai dan mutahir.
4. Kelengkapan unsur kualitas penerbit : Lengkap, berkualitas (Scopus Q3), penerbit sangat lengkap
5. Indikasi plagiasi : Tidak ada indikasi plagiasi
6. Kesesuaian bidang ilmu : Sesuai dengan bidang ilmu

Purwokerto, 30 Maret 2021 *) Wajib diisi

Reviewer 1

Prof. Dr.rer.nat. Imam Widhiono M.Z., M.S.
NIP. 195904201985031002
Jabatan/Gol. : Guru Besar/(Gol. IV/c)
Bidang Ilmu : Entomologi
Unit Kerja : Fakultas Biologi Unsoed

Reviewer 2

Dr. Elly Proklamasiningsih, M.P.
NIP. 196108171986032001
Jabatan/Gol. : Guru Besar/(Gol. IV/c)
Bidang Ilmu : Fisiologi Tumbuhan
Unit Kerja : Fakultas Biologi Unsoed

**LEMBAR**
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[ ] Jurnal Nasional/Nasional terindeks di..... *

Hasil Penilaian Peer Review :

| Komponen Yang Dinilai | Nilai Maksimal Jurnal Ilmiah: Internasional/ Internasional bereputasi (30) | Nasional Terakreditasi | Nasional *) | Nilai Akhir Yang Diperoleh |
|---|---|---|---|---|
| a Kelengkapan unsur isi artikel (10%) | 10 % X 30= 3 | | | 2,9 |
| b Ruang lingkup dan kedalaman pembahasan (30%) | 30 % X30= 9 | | | 8,9 |
| c Kecukupan dan kemutahiran data/informasi dan metodologi (30%) | 30 % X30= 9 | | | 8,9 |
| d Kelengkapan unsur dan kualitas terbitan/jurnal (30%) | 30 % X30= 9 | | | 8,9 |
| **Total = (100%)** | **30** | | | 29,6 |
| **Nilai Pengusul (40 % x Total)/3** | **4,00** | | | 3,95 |

Catatan Penilaian artikel oleh Reviewer:

1. Tentang kelengkapan dan kesesuaian unsur : kelengkapan unsur isi artikel lengkap.
2. Tentang ruang lingkup dan kedalaman pembahasan : Ruang lingkup dan kedalaman pembahasan mendalam.
3. Kecukupan dan kemutahiran data serta metodologi : Data dan metodologi mutakhir.
4. Kelengkapan unsur kualitas penerbit : unsur kualitas penerbit sangat lengkap.
5. Indikasi plagiasi : tidak ada indikasi plagiasi.
6. Kesesuaian bidang ilmu : sesuai dengan bidang ilmu.

Purwokerto, 29/3-2021 *) wajib diisi

Reviewer 2

Dr. Elly Proklamasiningsih, M.P.
NIP. 196108171986032001
Jabatan/Gol. : Lektor Kepala/(Gol. IV/b)
Bidang Ilmu : Fisiologi Tumbuhan
Unit Kerja : Fakultas Biologi Unsoed

Mengetahui,
Dekan,

Prof. Dr.rer.nat. Imam Widhiono M.Z, M.S.
NIP. 195904201985031002
Unit Kerja : Fakultas Biologi Unsoed

**LEMBAR**
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| Komponen Yang Dinilai | Nilai Maksimal Jurnal Ilmiah: Internasional/ Internasional bereputasi (30) | | Nasional Terakreditasi ( ) | | Nasional *) ( ) | | Nilai Akhir Yang Diperoleh |
|---|---|---|---|---|---|---|---|
| a Kelengkapan unsur isi artikel (10%) | 10 % X 30= | 3 | | | | | 3 |
| b Ruang lingkup dan kedalaman pembahasan (30%) | 30 % X30= | 9 | | | | | 9 |
| c Kecukupan dan kemutahiran data/informasi dan metodologi (30%) | 30 % X30= | 9 | | | | | 9 |
| d Kelengkapan unsur dan kualitas terbitan/jurnal (30%) | 30 % X30= | 9 | | | | | 9 |
| **Total = (100%)** | **30** | | | | | | 30 |
| **Nilai Pengusul (40 % x Total)/3** | **4,00** | | | | | | 4,00 |

Catatan Penilaian artikel oleh Reviewer:

1. Tentang kelengkapan dan kesesuaian unsur : lengkap dan sesuai unsur.
2. Tentang ruang lingkup dan kedalaman pembahasan : spesifik dgn kedalaman sesuai
3. Kecukupan dan kemutahiran data serta metodologi : data cukup mutakhir metodologi sesuai terkini lengkap dan terkini.
4. Kelengkapan unsur kualitas penerbit : lengkap dan terkini
5. Indikasi plagiasi : tidak ada
6. Kesesuaian bidang ilmu : sesuai.

Purwokerto, *) wajib diisi

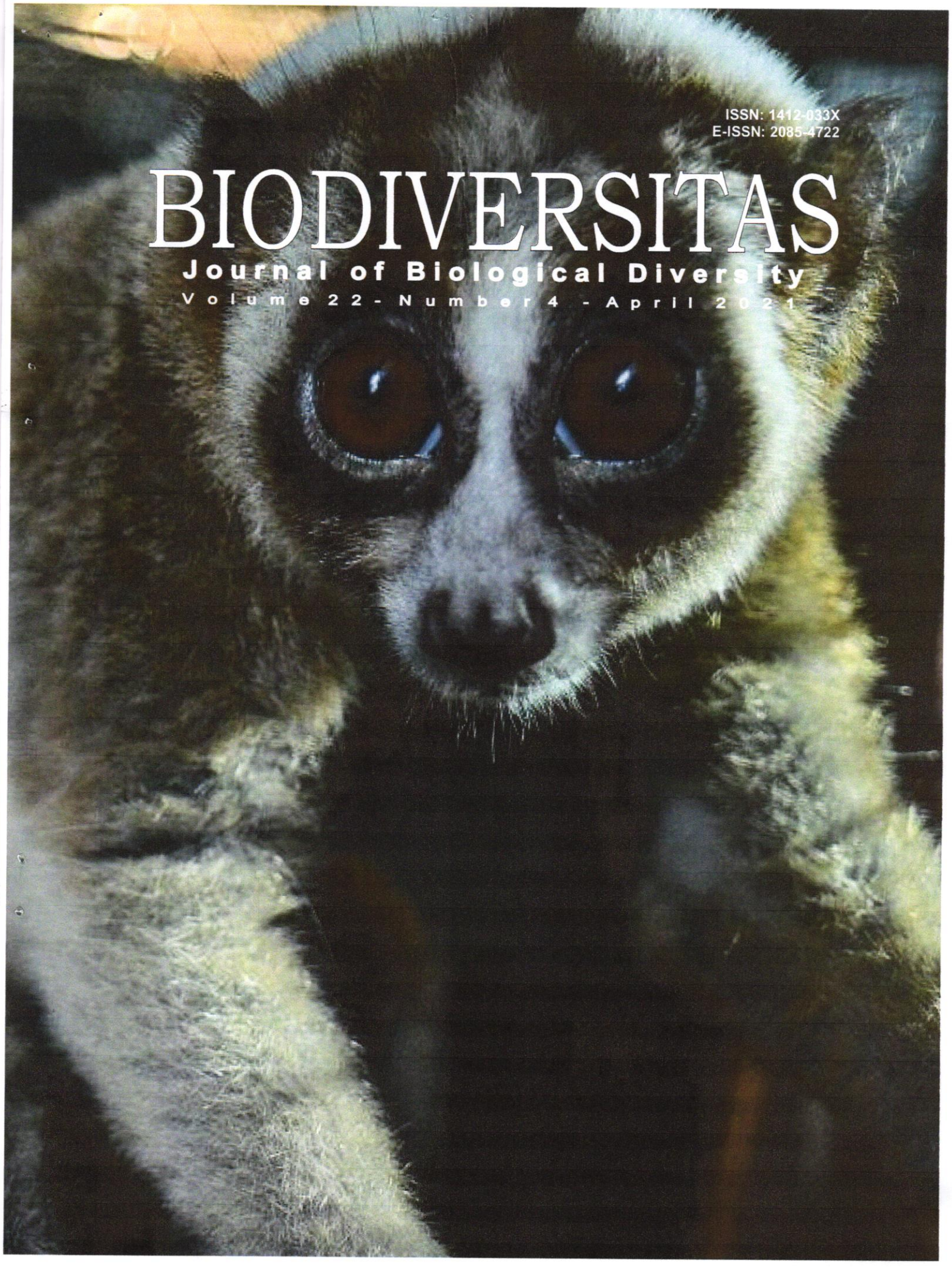
Reviewer 1

Prof. Dr.rer.nat. Imam Widhiono M.Z., M.S.
NIP. 195904201985031002
Jabatan/Gol. : Guru Besar/(Gol. IV/c)
Bidang Ilmu : Entomologi
Unit Kerja : Fakultas Biologi Unsoed

Mengetahui,
Dekan

Prof. Dr.rer.nat. Imam Widhiono M.Z, M.S.
NIP. 195904201985031002
Unit Kerja : Fakultas Biologi Unsoed

# ARTIKEL



ISSN: 1412-033X
E-ISSN: 2085-4722

# BIODIVERSITAS

## Journal of Biological Diversity

Volume 22 - Number 4 - April 2021

# BIODIVERSITAS

Journal of Biological Diversity

Volume 22 – Number 4 – April 2021

**ISSN/E-ISSN:**
1412-033X (printed edition), 2085-4722 (electronic)



**PUBLISHER:**
The Society for Indonesian Biodiversity

**CO-PUBLISHER:**
Department of Biology, Faculty of Mathematics and Natural Sciences, Sebelas Maret University, Surakarta

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biodiversitas.mipa.uns.ac.id; smujo.id/biodiv

Society for Indonesia Biodiversity

Sebelas Maret University Surakarta

Published by Smujo International for The Society for Indonesian Biodiversity and Sebelas Maret University Surakarta

# GUIDANCE FOR AUTHORS

**Aims and Scope** *Biodiversitas, Journal of Biological Diversity* or abbreviated as *Biodiversitas* encourages submission of manuscripts dealing with all biodiversity aspects of plants, animals and microbes at the level of the gene, species, and ecosystem as well as ethnobiology.

**Article types** The journal seeks original full-length research papers, reviews, and short communication. Manuscript of original research should be written in no more than 8,000 words (including tables and picture), or proportional with articles in this publication number. Review articles will be accommodated, while, short communication should be written at least 2,000 words, except for pre-study.

**Submission** The journal only accepts online submission, through open journal system (**https://smujo.id/biodiv/about/submissions**) or email to the editors at **unsjournals@gmail.com**. Submitted manuscripts should be the original works of the author(s). The manuscript must be accompanied by a cover letter containing the article title, the first name and last name of all the authors, a paragraph describing the claimed novelty of the findings versus current knowledge. Submission of a manuscript implies that the submitted work has not been published before (except as part of a thesis or report, or abstract); and is not being considered for publication elsewhere. When a manuscript written by a group, all authors should read and approve the final version of the submitted manuscript and its revision; and agree the submission of manuscripts for this journal. All authors should have made substantial contributions to the concept and design of the research, acquisition of the data and its analysis; drafting of the manuscript and correcting of the revision. All authors must be responsible for the quality, accuracy, and ethics of the work.

**Ethics** Author(s) must obedient to the law and/or ethics in treating the object of research and pay attention to the legality of material sources and intellectual property rights.

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**A charge** Starting on January 1, 2019, publishing costs waiver is granted to authors of graduate students from **Least Developed Countries**, who first publish the manuscript in this journal. However, other authors are charged USD 250 (IDR 3,500,000). Additional charges may be billed for language editing, USD 75-150 (IDR 1,000,000-2,000,000).

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**Manuscript preparation** Manuscript is typed on A4 (210x297 $mm^2$) paper size, **in a single column**, single space, 10-point (10 pt) Times New Roman font. The margin text is 3 cm from the top, 2 cm from the bottom, and 1.8 cm from the left and right. Smaller lettering size can be applied in presenting table and figure (9 pt). Word processing program or additional software can be used, however, it must be PC compatible and Microsoft Word based (*.doc* or *.rtf*; **not *.docx***). **Scientific names** of species (incl. subspecies, variety, etc.) should be written in italic, except for italic sentence. Scientific name (genera, species, author), and cultivar or strain should be mentioned completely for the first time mentioning it in the body text, especially for taxonomic manuscripts. Name of genera can be shortened after first mentioning, except generating confusion. Name of the author can be eliminated after first mentioning. For example, *Rhizopus oryzae* L. UICC 524, hereinafter can be written as *R. oryzae* UICC 524. Using trivial name should be avoided, otherwise generating confusion. **Biochemical and chemical nomenclature** should follow the order of the IUPAC - IUB. For DNA sequence, it is better used Courier New font. Symbols of standard chemical and abbreviation of chemistry name can be applied for common and clear used, for example, completely written butilic hydroxyl toluene (BHT) to be BHT hereinafter. **Metric measurement** use IS denomination, usage other system should follow the value of equivalent with the denomination of IS first mentioning. Abbreviations set of, like g, mg, mL, etc. do not follow by dot. Minus index ($m^{-2}$, $L^{-1}$, $h^{-1}$) suggested to be used, except in things like "per-down in one column with text, in that case can be written separately. **Number** one to ten are expressed with words, except if it relates to measurement, while values above them written in number, except in early sentence. The fraction should be expressed in decimal. In the text, it should be used "%" rather than "percent". Avoid expressing ideas with complicated sentence and verbiage, and used efficient and effective sentence.

**Title** of the article should be written in compact, clear, and informative sentence, preferably not more than 20 words. Name of author(s) should be completely written. **Name and institution** address should also be completely written with street name and number (location), postal code, telephone number, facsimile number, and email address. Manuscript written by a group, author for correspondence along with address is required. First page of the manuscript is used for writing above information.

**Abstract** should not be more than 200 words. **Keywords** is about five words, covering scientific and local name (if any), research theme, and special methods which used; and sorted from A to Z. All important **abbreviations** must be defined at their first mention. **Running title** is about five words. **Introduction** is about 400-600 words, covering the background and aims of the research. **Materials and Methods** should emphasize on the procedures and data analysis. **Results and Discussion** should be written as a series of connecting sentences, however, for manuscript with long discussion should be divided into subtitles. Thorough discussion represents the causal effect mainly explains for why and how the results of the research were taken place, and do not only re-express the mentioned results in the form of sentences. **Concluding** sentence should be given at the end of the discussion. **Acknowledgments** are expressed in a brief; all sources of institutional, private and corporate financial support for the work must be fully acknowledged, and any potential conflicts of interest are noted.

**Figures and Tables** of maximum of three pages should be clearly presented. Title of a picture is written down below the picture, while title of a table is written above the table. Colored figures can only be accepted if the information in the manuscript can lose without those images; chart is preferred to use black and white images. Author could consign any picture or photo for the front cover, although it does not print in the manuscript. All images property of others should be mentioned source. **There is no appendix**, all data or data analysis are incorporated into Results and Discussions. For broad data, it can be displayed on the website as a supplement.

**References** Author-year citations are required. In the text give the authors name followed by the year of publication and arrange from oldest to newest and from A to Z. In citing an article written by two authors, both of them should be mentioned, however, for three and more authors only the first author is mentioned followed by et al., for example: Saharjo and Nurhayati (2006) or (Boonkerd 2003a, b, c; Sugiyarto 2004; El-Bana and Nijs 2005; Balagadde et al. 2008; Webb et al. 2008). Extent citation as shown with word "*cit*" should be avoided. Reference to unpublished data and personal communication should not appear in the list but should be cited in the text only (e.g., Rifai MA 2007, pers. com. (personal communication); Setyawan AD 2007, unpublished data). In the reference list, the references should be listed in an alphabetical order (better, if only 20 for research papers). Names of journals should be abbreviated. Always use the standard abbreviation of a journal's name according to the **ISSN List of Title Word Abbreviations** (www.issn.org/2-22661-LTWA-online.php). The following examples are for guidance.

**Journal:**

Saharjo BH, Nurhayati AD. 2006. Domination and composition structure change at hemic peat natural regeneration following burning; a case study in Pelalawan, Riau Province. Biodiversitas 7: 154-158.

**Book:**

Rai MK, Carpinella C. 2006. Naturally Occurring Bioactive Compounds. Elsevier, Amsterdam.

**Chapter in book:**

Webb CO, Cannon CH, Davies SJ. 2008. Ecological organization, biogeography, and the phylogenetic structure of rainforest tree communities. In: Carson W, Schnitzer S (eds) Tropical Forest Community Ecology. Wiley-Blackwell, New York.

**Abstract:**

Assaeed AM. 2007. Seed production and dispersal of *Rhazya stricta*. 50th annual symposium of the International Association for Vegetation Science, Swansea, UK, 23-27 July 2007.

**Proceeding:**

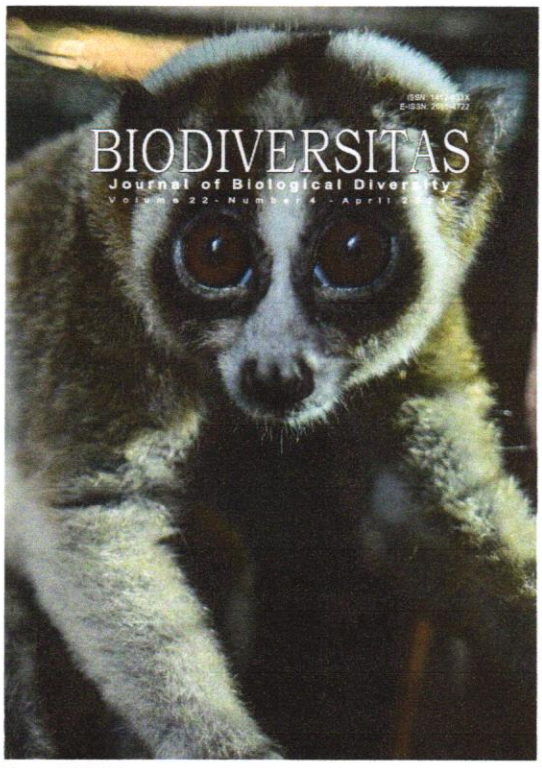
Alikodra HS. 2000. Biodiversity for development of local autonomous government. In: Setyawan AD, Sutarno (eds.) Toward Mount Lawu National Park; Proceeding of National Seminary and Workshop on Biodiversity Conservation to Protect and Save Germplasm in Java Island. Universitas Sebelas Maret, Surakarta, 17-20 July 2000. [Indonesian]

**Thesis, Dissertation:**

Sugiyarto. 2004. Soil Macro-invertebrates Diversity and Inter-Cropping Plants Productivity in Agroforestry System based on Sengon. [Dissertation]. Universitas Brawijaya, Malang. [Indonesian]

**Information from internet:**

Balagadde FK, Song H, Ozaki J, Collins CH, Barnet M, Arnold FH, Quake SR, You L. 2008. A synthetic *Escherichia coli* predator-prey ecosystem.




(https://smujo.id/biodiv/issue/view/299)

Vol. 22 No. 4 (2021)

## Full Issue

Front Cover (https://smujo.id/biodiv/issue/view/299/151)

## Articles

### DNA barcoding of the tidal swamp rice (Oryza sativa) landraces from South Kalimantan, Indonesia (https://smujo.id/biodiv/article/view/7780)

DINDIN HIDAYATUL MURSYIDIN, YUDHI AHMAD NAZARI, BADRUZSAUFARI, MUHAMMAD RIDHO DINTA MASMITRA

PDF (https://smujo.id/biodiv/article/view/7780/4693)

## Genetic variation of longtail tuna Thunnus tonggol landed in four fish markets in Indonesia based on mitochondrial DNA (https://smujo.id/biodiv/article/view/7304)

IDA AYU ASTARINI, M. DANIE AL MALIK, NI LUH ASTRIA YUSMALINDA, ANDRIANUS SEMBIRING, NI PUTU DIAN PERTIWI, NI KADEK DITA CAHYANI

PDF (https://smujo.id/biodiv/article/view/7304/4700)

## The effect of biological agent and botanical fungicides on maize downy mildew (https://smujo.id/biodiv/article/view/7908)

JOKO PRASETYO, CIPTA GINTING, HASRIADI MAT AKIN, RADIX SUHARJO, AININ NISWATI, AULIANA AFANDI, REZA ADIWIJAYA, SUDIONO, MUHAMMAD NURDIN

PDF (https://smujo.id/biodiv/article/view/7908/4701)

## Six new species and a new record of Curcuma L. (Zingiberaceae) from Thailand (https://smujo.id/biodiv/article/view/8062)

SURAPON SAENSOUK, THAWATPHONG BOONMA, PIYAPORN SAENSOUK

PDF (https://smujo.id/biodiv/article/view/8062/4702)

## Combination of plant growth-promoting bacteria and botanical pesticide increases organic red rice yield and reduces the Leptocorisa acuta population (https://smujo.id/biodiv/article/view/8079)

MOHAMMAD HOESAIN, SIGIT PRASTOWO, SUHARTO, ANKARDIANSYAH PANDU PRADANA, IIS NUR ASYIAH, FARIZ KUSTIAWAN ALFARIZY, MUH. ADIWENA

PDF (https://smujo.id/biodiv/article/view/8079/4710)

## Agronomic characteristics of 30 promising lines of aromatic, red, and black rice and their antioxidant and cytotoxic effects in some cancer cells (https://smujo.id/biodiv/article/view/7193)

YUSUF LIMBONGAN, RICO RAMADHAN , KUNIYOSHI SHIMIZU, ENOS TANGKE ARUNG

PDF (https://smujo.id/biodiv/article/view/7193/4703)

## The potential of bird diversity in the urban landscape for birdwatching in Java, Indonesia (https://smujo.id/biodiv/article/view/7448)

INSAN KURNIA, HARNIOS ARIEF, ANI MARDIASTUTI, RACHMAD HERMAWAN

PDF (https://smujo.id/biodiv/article/view/7448/4704)

Comparison of the effectiveness of pregnancy diagnosis in Aceh cattle through measurement of interferon-tau and progesterone concentrations (https://smujo.id/biodiv/article/view/7653)

BUDIANTO PANJAITAN, TONGKU NIZWAN SIREGAR, HAFIZUDDIN, ARMAN SAYUTI, MULYADI ADAM, TEUKU ARMANSYAH, SYAFRUDDIN

PDF (https://smujo.id/biodiv/article/view/7653/4705)

Potential of fungi isolated from a mangrove ecosystem in Northern Sulawesi, Indonesia: Protease, cellulase and anti-microbial capabilities (https://smujo.id/biodiv/article/view/7791)

AGUS TRIANTO, OCKY KARNA RADJASA, SUBAGIYO, HARTUTI PURNAWENI, MUHAMMAD SYAIFUDIEN BAHRY, RIGNOLDA DJAMALUDIN, AIYEN TJOA, IAN SINGLETON, KAREN DIELE, DARREN EVAN

PDF (https://smujo.id/biodiv/article/view/7791/4706)

Isolation, characterization, activity test and molecular identification of thermophilic bacteria producing proteases from Dolok Tinggi Raja Natural Hot Springs, North Sumatra, Indonesia (https://smujo.id/biodiv/article/view/7832)

EDY FACHRIAL, VISENSIUS KRISDIANILO, HARMILENI, I NYOMAN EHRICH LISTER, TITANIA T. NUGROHO, SARYONO

PDF (https://smujo.id/biodiv/article/view/7832/4707)

Implementation of species protection act for the conservation of Tanimbar corella, Cacatua goffiniana (Finsch, 1863) (https://smujo.id/biodiv/article/view/7005)

TRI HARYOKO , MARK O'HARA, BERENIKA MIODUSZEWSKA, HARI SUTRISNO, LILIK BUDI PRASETYO, ANI MARDIASTUTI

PDF (https://smujo.id/biodiv/article/view/7005/4709)

Chemical composition, antibacterial and antioxidant activities of essential oils extracted from dry and fresh Brocchia cinerea (Vis.) (https://smujo.id/biodiv/article/view/7840)

NISRINE CHLIF; ABDELAZIZ ED-DRA; MOHAMMED DIOURI, NOUREDDINE EL MESSAOUDI, BADR ZEKKORI, FOUZIA RHAZI FILALI, AMAR BENTAYEB

PDF (https://smujo.id/biodiv/article/view/7840/4708)

Application of selected teak clone and organic fertilizer to accelerate

rehabilitation of lowland forest in Java, Indonesia (https://smujo.id/biodiv/article/view/7436)

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**BIODIVERSITAS**
Volume 22, Number 4, April 2021
Pages: 1869-1878

ISSN: 1412-033X
E-ISSN: 2085-4722
DOI: 10.13057/biodiv/d220433

# Short Communication: Diversity of cellulolytic bacteria isolated from coastal mangrove sediment in Logending Beach, Kebumen, Indonesia

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Manuscript received: 28 October 2020. Revision accepted: 22 March 2021.

**Abstract.** *Pramono H, Mariana A, Ryandini D, Sudiana E. 2021. Short Communication: Diversity of cellulolytic bacteria isolated from coastal mangrove sediment in Logending Beach, Kebumen, Indonesia. Biodiversitas 22: 1869-1878.* Bacteria from mangrove sediments have the potential to produce active enzymes and metabolites needed in the industry. Mangrove ecosystems in various parts of Indonesia have been explored as sources of bacteria. However, the mangrove ecosystems in Logending Beach, Kebumen District, Indonesia are still unexplored. This study aims to determine the diversity of cellulolytic bacteria using the 16S rRNA gene. Ninety-nine bacteria were isolated from Logending coastal mangrove sediments. Most of them (87.87%) had a cellulolytic activity, with the cellulolytic index values ranged from 0.25–13.96. The LG2 isolate had the highest cellulolytic activity (13.96). Molecular characterization of the LG2 isolate based on the 16S rRNA gene showed that it was similar to the *Fictibacillus nanhaiensis* strain JSM 082006. This result is new information on cellulolytic bacteria and might be used as a future source of cellulolytic enzymes.

**Keywords**: 16S rRNA gene cellulolytic, mangrove, molecular, sediments

## INTRODUCTION

Mangrove forests are very productive ecosystems and a potential source of superior bacteria that produce active metabolites and industrial enzymes. In tropical mangroves, 91% of the microbial biomass is bacteria and fungi (Naresh et al. 2019). The existence of microorganisms in mangrove ecosystems is closely related to the stability of the ecosystem. Extracellular enzymes produced by microbes are used to break down complex organic material used by cells as a nutrition source (Thompson et al. 2013). The complexity of the mangrove ecosystems and anthropogenic pressure require microbes to produce extracellular enzymes that function under these complex conditions. Enzymes are susceptible to environmental factors; however, the enzymes produced by microbes in complex ecosystems have a high tolerance to environmental factors (Pal et al. 2018). Mangroves harbor potential bacteria that produce active metabolites and enzymes (Bibi et al. 2017).

The study of microbial interactions with other ecosystem components, such as plant roots, is important for understanding the function and remediation of mangrove ecosystems (Chantarasiri 2015). The bacteria in the mangrove ecosystems break down mangrove leaf litter into organic material used as a source of nutrition by the organisms living in mangrove forests (Glaeser et al. 2013). Environmental factors strongly influence the diversity of bacteria in mangrove ecosystems. Sakhia et al. (2016) showed that bacterial species are affected by mangrove vegetation in the water, especially towards the sea (Liu et al. 2019). Although many microorganisms can degrade cellulose, only a few produce significant quantities of cell-free bioactive compounds capable of completely hydrolyzing crystalline cellulose in vitro (Rajagopal and Kannan 2015).

Mangrove bacteria can degrade cellulose (Behera et al. 2013; Basak et al. 2016). In a study of mangrove sediments in Dhulibhashani, Sundarbands, India, Basak et al. (2016) found that Proteobacteria, including Bacteroidetes, Acidobacteria, Firmicutes, Actinobacteria, Nitrospirae, Cyanobacteria, Planctomycetes, and Fusobacteria, were dominant using 16S rRNA gene tag sequencing. *Micrococcus*, *Bacillus*, *Pseudomonas*, *Xanthomonas*, and *Brucella* spp. were collected from the Mahanadi River Delta in Odisha, India, which has a high capacity carboxymethyl cellulose hydrolysis (Behera et al. 2013). In Andaman Island mangroves, Rajagopal and Kannan (2017) isolated 19 morphologically distinct Actinobacteria strains from Andaman Island mangroves and 4 strains possessed good cellulose degradation activity, of which strain MHA15 produced the highest cellulase (14,379 IU/mL). Taxonomically, the four strains are members of the genus *Actinoalloteichus* differing from *Actinoalloteichus cyanogriseus* (Behera et al. 2013). Pal et al. (2018) isolated bacteria strains from the Indian Ganges Delta that belongs to the family Bacillaceae and related to the genus *Fictibacillus* based on 16S rRNA sequencing.

Several studies have been carried out on the cellulolytic bacterial diversity of several mangrove ecosystems in Indonesia. Hastuti et al. (2015) found four cellulolytic bacteria species (*Micrococcus luteus*, *Bacillus pumilus*, *Planococcus citreus*, and *Bacillus cereus*) that originated

from mangrove bark on Waai Beach, Ambon Island. Ambeng et al. (2019) isolated 35 cellulolytic bacteria from mangrove sediments in Pangkep, South Sulawesi, belonging to seven genera, i.e., *Bacillus*, *Staphylococcus*, *Vibrio*, *Micrococcus*, *Alteromonas*, *Escherichia*, and *Listeria*. Kurniawan et al. (2017) isolated *Bacillus pumilus*, *Pseudomonas* sp., *Bacillus amyloliquefaciens*, *Bacillus alvei*, and *Bacillus coagulant* from mangrove sediment on Bangka Island, and *Pseudomonas aeruginosa,* which produced the highest cellulase. Nursyiwani et al. (2020) examined 24 isolates of cellulolytic antimicrobial pathogens from mangrove sediment samples from Dumai, Riau and found that 9 isolates could inhibit the development of pathogenic microbes; and 3 of these isolates were similar to *Bacillus toyonensis*.

Maharsiwi et al. (2020) has identified cellulolytic bacteria on mangrove sponges from the Seribu Islands similar to *Pseudomonas luteola* strain NBRC 103146, *Bacillus cereus* strain 24k, *Pseudomonas aeruginosa* strain DSM 50071, *Microbacterium maritypicum* strain DSM 20578, and *Brachybacterium conglomeratum* strain J 1015. Kurniawan et al. (2019) isolated bacteria from the tin-mining region in West Bangka and identified them as *Bacillus amyloliquefaciens*.

Since there has been no study on bacterial diversity in the mangrove area of the southern coast of Java, especially at Logending Beach, Kebumen, we conducted the study to determine the diversity of cellulolytic bacteria mangrove sediments and identified using 16S rRNA sequencing.

## MATERIALS AND METHODS

### Study site

Sediments were collected at six locations in the mangrove forest in Logending Beach, Ayah Subdistrict, Kebumen District, Central Java Province, Indonesia (7° 42' 28.0'' S and 109° 23' 20.1" E). Two locations are at the estuary and three locations along the coast (Figure 1). The samples were processed in the Microbiology Laboratory of the Faculty of Biology, Jenderal Soedirman University, Purwokerto, Banyumas, Indonesia.

### Isolation of mangrove sediment bacteria

Bacteria were isolated using the pour plate technique on nutrient agar (NA) medium. Ten g of sediment samples were homogenized in 90 mL physiological saline and then diluted up to $10^{-5}$ fold. The last three dilutions were cultured in NA medium and incubated for 24 hours at room temperature. Bacterial colonies grow on the diluted medium of $10^{-3}$, $10^{-4}$, and $10^{-5}$ were counted, and the bacterial density was reported as the total number of bacteria per gram of mangrove sediment. Bacterial diversity was observed from colony morphology. The colonies were further purified by the quadrant streak technique to isolate and characterize bacterial isolates.


**Figure 1.** Sampling locations (white circles) in the mangrove forest of Logending Beach, Ayah Subdistrict, Kebumen District, Central Java Province, Indonesia

**Cellulolytic activity test**

Bacterial isolates were inoculated on CMC agar and incubated for 24 hours. The cellulolytic activity was observed by dripping Congo Red solution on the culture surface. The formation of clear zones around a colony indicates cellulolytic activity. The clear zone and colony diameters were measured, and enzymatic activity was calculated by dividing the diameter of the clear zone by the colony diameter.

**Isolation of bacterial genomic DNA**

Bacterial chromosomal DNA was isolated using the method of Klijn et al. (1991) with some modifications. A single bacterial colony was suspended in 50 µL 10 mM Tris-HCl (pH 8) buffer solution containing 400 µg lysozyme and incubated for 1 hour at 37°C. Cell lysis was improved by adding 50 µL 10% SDS and 250 µL buffer. The mixture was centrifuged at 5,000 × *g* for 10 minutes. The supernatant was transferred to a new tube, and 60 mL 3 M sodium acetate and 1 mL 96% chilled ethanol were added. After centrifuging at 12,000 × *g* for 15 minutes, the DNA pellet was placed in 300 µL 70% ethanol and centrifuged at 8,000 × *g* for 5 minutes. The pellet was dried at room temperature until all the ethanol evaporated, and then it was dissolved in 50 µL TE buffer. The chromosomal DNA was analyzed by agarose gel electrophoresis.

**16S rRNA gene amplification and phylogenetic analysis**

The 16S rRNA gene was amplified by polymerase chain reaction (PCR) using a DNA chromosome template. The PCR mixture consisted of 8.55 µL $ddH_2O$, 3 µL 10× PCR buffer, 0.45 µL 5 mM dNTP, 1.05 µL 0.6 mM $MgCl_2$, 2 µL primers, 0.15 µL *Taq* polymerase, and 0.6 µL chromosomal DNA. PCR analysis was performed on a thermocycler, with an initial denaturation step at 94°C for 4 min, followed by 34 cycles of 94°C for 30 s, 50°C for 1 min, and 72°C for 2 min. The primers used to amplify the 16S rRNA gene were Bact27F (5'-AGAGTTTGATCATG GCTCAG-3') and Uni1492R (5'-GGTTACCTTGT TACGACTT-3'). The PCR products were verified by agarose gel electrophoresis and then sequenced.

**Data analysis**

Using a BLAST search, selected 16S rRNA sequences were compared to other sequences in GenBank to determine the level of similarity. Then, a phylogenetic tree of the 16S rRNA sequences was constructed using MEGA ver. 6. (Tamura et al. 2013)

## RESULTS AND DISCUSSION

**Diversity of cellulolytic bacteria in coastal mangrove sediment from Logending**

In total, there were 99 bacteria strains isolated from the Logending mangrove sediments (Table 1). The highest number of isolates was obtained from the first sampling location (58 isolates), followed by the 3rd and 5th sampling locations (53 isolates from each location), and the lowest number was obtained from the 6th sampling location (14 isolates) (Figure 2). The first sampling point is directly opposite the river flow direction, while the 6th sampling location faces the sea. The difference in the number of isolates among sampling points is probably due to the sediment type. The result showed that 87 bacterial isolates have cellulolytic activity, as indicated by a clear zone around the colony on CMC agar containing Congo Red (Figure 3).

The cellulolytic index (CI) reflects the ability of the cellulase produced by bacterial isolates to degrade cellulose on CMC agar. The CI values ranged from 0.25 to 13.96 (Table 1), and 12 isolates had a CI > 3.0. The highest CI (13.96) was produced by the LG2 isolate (Table 1).

Mangrove sediments in Logending Beach have a high potential for cellulolytic bacteria, and only a few isolates have lower CI values than in other mangrove areas in Indonesia. Kurniawan et al. (2017) only found five cellulolytic bacteria from mangrove sediments at Sungailiat and Tukak Sadai on Bangka Island. Nursyirwani et al. (2020) obtained 24 cellulolytic bacterial isolates with CI values ranging from 1.00 to 2.86.

Many cellulolytic bacteria have been isolated from sediments and tropical mangrove soils. Functional metagenomic analyses have shown that mangrove land has high cellulolytic enzyme activity. Specific sequences involved in cellulose degradation were identified in mangroves from Brazil (Thompson et al. 2013). High cellulolytic activity can be found in sediments with high mangrove litter levels, which creates a cellulose-rich environment for bacterial populations.

**Identification of 16S rRNA genes**

Based on 16S rRNA sequences and using BLAST from NCBI, it showed the LG2 isolate had the highest similarity (99.86%) to *Fictibacillus nanhaiensis* strain JSM 082006 in GenBank (Table 2). Other closely related strains were *F. halophilus* (99.79%), *F. phosphorivorans* (99.86%), *F. barbaricus* (98.21%), *F. aquaticus* (98.21%), *F. arsenicus* (97.43.0%), *F. rigui* (97%), *F. enclensis* (96.64%), *F. solisalsi* (96.43%), and *Bacillus tianmuensis* (96.22%) (Table 2).

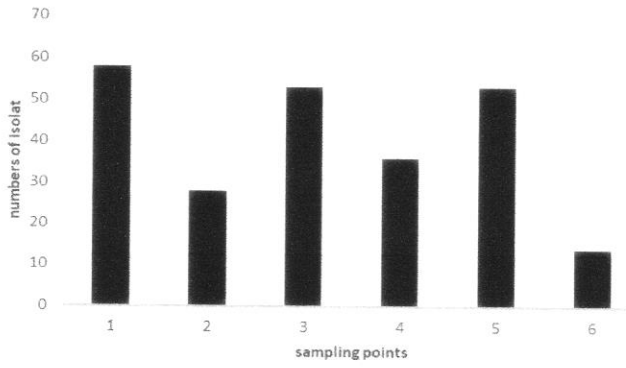


**Figure 2.** Number of isolates in each sampling locations

**Table 1.** Isolat richness and densities (means + stdev)

| Isolate name | Sampling points (n = 6) | | | | | |
|---|---|---|---|---|---|---|
| | **1** | **2** | **3** | **4** | **5** | **6** |
| LG 1 | 0 | 0 | 0 | 0 | 2500,00 ± 577,35 | 0 |
| LG 2 | 633,33 ± 115,47 | 25125,00 ± 3544,36 | 3000,00 ± 433,01 | 338333,30 ± 17559,42 | 17833,33 ± 2565,80 | 16833,33 ± 288,68 |
| LG 3 | 0 | 0 | 0 | 2166,67 ± 577,35 | 5000,00 ± 500,00 | 0 |
| LG 4 | 350000,00 ± 28867,51 | 1250,00 ± 204,12 | 16666,67 ± 1443,38 | 0 | 453333,30 ± 45092,50 | 500,00 ± 86,60 |
| LG 5 | 0 | 0 | 0 | 0 | 5000,00 ± 500,00 | 0 |
| LG 6 | 0 | 250000,00 ± 40824,83 | 33333,33 ± 5773,50 | 4166,67 ± 577,35 | 18333,33 ± 2886,75 | 16833,33 ± 288,68 |
| LG 7 | 40000,00 ± 8231,04 | 19969125,00 ± 1353518,77 | 58333,33 ± 5773,50 | 183333,30 ± 28867,51 | 54000,00 ± 3605,55 | 16666,67 ± 2886,75 |
| LG 8 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 9 | 0 | 0 | 166,67 ± 14,43 | 0 | 0 | 0 |
| LG 10 | 0 | 0 | 333,33 ± 43,30 | 0 | 16666,67 ± 2886,75 | 0 |
| LG 11 | 0 | 0 | 2333,33 ± 144,34 | 0 | 35000,00 ± 5000,00 | 0 |
| LG 12 | 0 | 0 | 17500,00 ± 1322,88 | 0 | 0 | 0 |
| LG 13 | 13333,33 ± 2886,75 | 62500,00 ± 5000,00 | 8333,33 ± 2886,75 | 5000,00 ± 288,68 | 18333,33 ± 2886,75 | 0 |
| LG 14 | 1666,67 ± 288,68 | 50375,00 ± 3685,56 | 500,00 ± 43,30 | 500,00 ± 50,00 | 17833,33 ± 2565,80 | 0 |
| LG 15 | 0 | 0 | 0 | 0 | 19000,00 ± 1732,05 | 0 |
| LG 16 | 0 | 125000,00 ± 28867,51 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 17 | 46166,67 ± 5392,90 | 3000,00 ± 408,25 | 103333,30 ± 2886,75 | 15000,00 ± 2500,00 | 8500,00 ± 866,03 | 334000,00 ± 27712,81 |
| LG 18 | 0 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 |
| LG 19 | 0 | 125000,00 ± 41231,06 | 0 | 0 | 173333,30 ± 46188,02 | 0 |
| LG 20 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 21 | 0 | 0 | 666,67 ± 57,74 | 1666,67 ± 288,68 | 0 | 0 |
| LG 22 | 0 | 0 | 0 | 11666,67 ± 2886,75 | 16666,67 ± 2886,75 | 0 |
| LG 23 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 |
| LG 24 | 0 | 0 | 0 | 7666,67 ± 577,35 | 0 | 0 |
| LG 25 | 1328333,00 ± 183462,08 | 149625,00 ± 33623,84 | 38333,33 ± 7637,63 | 1666,67 ± 288,68 | 9333,33 ± 2081,67 | 17666,67 ± 3511,88 |
| LG 26 | 0 | 0 | 1666,67 ± 152,75 | 166666,70 ± 28867,51 | 0 | 0 |
| LG 27 | 1666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 28 | 1666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 29 | 5000,00 ± 0,00 | 0 | 0 | 15000,00 ± 2000,00 | 0 | 0 |
| LG 30 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 31 | 0 | 0 | 0 | 3333,33 ± 288,68 | 3500,00 ± 500,00 | 0 |
| LG 32 | 3333,33 ± 577,35 | 125,00 ± 20,41 | 0 | 0 | 0 | 0 |
| LG 33 | 500,00 ± 86,60 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 34 | 1666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 35 | 18333,33 ± 2886,75 | 0 | 16666,67 ± 5773,50 | 166,67 ± 28,87 | 25000,00 ± 2886,75 | 0 |
| LG 36 | 6666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 37 | 1833,33 ± 144,34 | 0 | 0 | 0 | 0 | 0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| LG 38 | 6666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 39 | 3333,33 ± 577,35 | 0 | 0 | 0 | 0 | 0 |
| LG 40 | 1666,67 ± 381,88 | 0 | 0 | 0 | 0 | 0 |
| LG 41 | 333,33 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 42 | 16833,33 ± 2753,79 | 0 | 0 | 0 | 16666,67 ± 2886,75 | 0 |
| LG 43 | 166,67 ± 28,87 | 0 | 0 | 0 | 500,00 ± 50,00 | 0 |
| LG 44 | 3500,00 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 45 | 333,33 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 46 | 2500,00 ± 500,00 | 0 | 7500,00 ± 866,03 | 8333,33 ± 1154,70 | 166833,30 ± 28724,26 | 0 |
| LG 47 | 0 | 0 | 0 | 0 | 333,33 ± 28,87 | 0 |
| LG 48 | 16666,67 ± 2886,75 | 0 | 5000,00 ± 866,03 | 0 | 3833,33 ± 763,76 | 0 |
| LG 49 | 0 | 1250,00 ± 288,68 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 50 | 0 | 0 | 9000,00 ± 1322,88 | 2666,67 ± 288,68 | 1666,67 ± 288,68 | 0 |
| LG 51 | 0 | 0 | 0 | 0 | 33666,67 ± 2309,40 | 0 |
| LG 52 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 53 | 13333,33 ± 1732,05 | 0 | 0 | 833,33 ± 144,34 | 0 | 0 |
| LG 54 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 55 | 15000,00 ± 866,03 | 0 | 40000,00 ± 1000,00 | 16666,67 ± 2886,75 | 1000,00 ± 100,00 | 166,67 ± 28,87 |
| LG 56 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 57 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 | 0 |
| LG 58 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 59 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 60 | 666,67 ± 57,74 | 100000,00 ± 20412,41 | 0 | 167000,00 ± 28583,21 | 0 | 0 |
| LG 61 | 0 | 110000,00 ± 8660,25 | 0 | 0 | 2666,67 ± 288,68 | 0 |
| LG 62 | 666,67 ± 57,74 | 0 | 500,00 ± 50,00 | 0 | 0 | 0 |
| LG 63 | 666,67 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 64 | 166,67 ± 28,87 | 0 | 0 | 0 | 0 | 0 |
| LG 65 | 333,33 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 66 | 166,67 ± 28,87 | 0 | 0 | 0 | 0 | 0 |
| LG 67 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 |
| LG 68 | 0 | 662500,00 ± 75000,00 | 8666,67 ± 1527,53 | 1666,67 ± 763,76 | 0 | 0 |
| LG 69 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 70 | 0 | 0 | 0 | 0 | 5000,00 ± 500,00 | 0 |
| LG 71 | 0 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 |
| LG 72 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 73 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 |
| LG 74 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 |
| LG 75 | 0 | 0 | 0 | 6666,67 ± 577,35 | 0 | 0 |
| LG 76 | 0 | 0 | 0 | 15000,00 ± 1000,00 | 0 | 0 |
| LG 77 | 0 | 0 | 0 | 333,33 ± 28,87 | 0 | 0 |
| LG 78 | 0 | 0 | 50000,00 ± 5000,00 | 666,67 ± 144,34 | 0 | 0 |
| LG 79 | 0 | 0 | 0 | 333,33 ± 28,87 | 0 | 0 |
| LG 80 | 0 | 0 | 0 | 6666,67 ± 577,35 | 50000,00 ± 5000,00 | 0 |
| LG 81 | 133333,30 ± 14433,76 | 0 | 3333,33 ± 763,76 | 0 | 13166,67 ± 1154,70 | 166,67 ± 28,87 |
| LG 82 | 0 | 0 | 4500,00 ± 866,03 | 0 | 1166,67 ± 288,68 | 0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| LG 83 | 0 | 0 | 333,33 ± 57,74 | 0 | 166,67 ± 72,17 | 0 |
| LG 84 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 85 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 86 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 87 | 0 | 0 | 9833,33 ± 288,68 | 0 | 0 | 0 |
| LG 88 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 89 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 90 | 0 | 0 | 333,33 ± 57,74 | 0 | 0 | 0 |
| LG 91 | 0 | 375,00 ± 50,00 | 0 | 0 | 0 | 0 |
| LG 92 | 2000,00 ± 433,01 | 0 | 0 | 0 | 0 | 0 |
| LG 93 | 1500,00 ± 346,41 | 0 | 166666,70 ± 28867,51 | 0 | 0 | 0 |
| LG 94 | 3333,33 ± 288,68 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 95 | 0 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 |
| LG 96 | 0 | 0 | 0 | 0 | 28333,33 ± 7637,63 | 0 |
| LG 97 | 0 | 0 | 0 | 500,00 ± 50,00 | 0 | 0 |
| LG 98 | 0 | 0 | 500,00 ± 50,00 | 0 | 0 | 0 |
| LG 99 | 0 | 0 | 1166,67 ± 288,68 | 0 | 0 | 0 |
| LG 100 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 101 | 5000,00 ± 866,03 | 0 | 0 | 0 | 0 | 0 |
| LG 102 | 5000,00 ± 866,03 | 0 | 0 | 0 | 0 | 0 |
| LG 103 | 16666,67 ± 2886,75 | 875,00 ± 150,00 | 17166,67 ± 2565,80 | 0 | 0 | 0 |
| LG 104 | 0 | 0 | 500,00 ± 50,00 | 0 | 0 | 0 |
| LG 105 | 0 | 0 | 1166,67 ± 288,68 | 0 | 0 | 0 |
| LG 106 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 107 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 |
| LG 108 | 33333,33 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 109 | 16666,67 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 110 | 16666,67 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 111 | 16666,67 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 112 | 166666,70 ± 158771,32 | 0 | 0 | 0 | 0 | 0 |
| LG 113 | 0 | 0 | 0 | 0 | 25000,00 ± 2886,75 | 0 |
| LG 114 | 0 | 12500,00 ± 2516,61 | 16666,67 ± 2886,75 | 16666,67 ± 2886,75 | 0 | 0 |
| LG 117 | 0 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 |
| LG 118 | 0 | 150000,00 ± 8164,97 | 0 | 0 | 0 | 0 |
| LG 119 | 16666,67 ± 2886,75 | 0 | 0 | 3666,67 ± 288,68 | 0 | 0 |
| LG 120 | 0 | 1250,00 ± 300,00 | 0 | 0 | 0 | 0 |
| LG 121 | 0 | 0 | 0 | 0 | 0 | 333333,30 ± 28867,51 |
| LG 122 | 0 | 125,00 ± 33,67 | 16666,67 ± 2886,75 | 0 | 0 | 0 |
| LG 123 | 0 | 0 | 0 | 0 | 0 | 16666,67 ± 2886,75 |
| LG 126 | 0 | 0 | 16666,67 ± 2886,75 | 0 | 166666,70 ± 28867,51 | 166666,70 ± 28867,51 |
| LG 125 | 0 | 250,00 ± 40,82 | 0 | 0 | 0 | 0 |
| LG 127 | 0 | 125,00 ± 45,64 | 0 | 0 | 0 | 0 |
| LG 128 | 0 | 1250,00 ± 191,49 | 0 | 0 | 0 | 0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| LG 129 | 0 | 0 | 16666,67 ± 2886,75 | 0 | 166666,70 ± 28867,51 | 0 |
| LG 130 | 0 | 0 | 166666,70 ± 28867,51 | 0 | 0 | 0 |
| LG 131 | 0 | 0 | 166666,70 ± 28867,51 | 0 | 0 | 0 |
| LG 134 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 135 | 33333,33 ± 31754,26 | 0 | 0 | 0 | 0 | 0 |
| LG 136 | 0 | 0 | 0 | 0 | 166666,70 ± 28867,51 | 0 |
| LG 137 | 50000,00 ± 8660,25 | 0 | 0 | 0 | 0 | 0 |
| LG 138 | 0 | 0 | 0 | 0 | 333333,30 ± 28867,51 | 0 |
| LG 139 | 0 | 0 | 0 | 0 | 166666,70 ± 28867,51 | 0 |
| LG 140 | 0 | 1250,00 ± 191,49 | 0 | 0 | 0 | 0 |
| LG 141 | 0 | 2500,00 ± 95,74 | 0 | 0 | 0 | 0 |
| LG 142 | 0 | 500,00 ± 81,65 | 0 | 0 | 0 | 0 |
| LG 143 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 |
| LG 144 | 0 | 0 | 0 | 0 | 0 | 333,33 ± 28,87 |
| LG 145 | 0 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 |

**Table 2.** The cellulolytic index of bacterial isolates collected from Logending coastal mangrove sediment

| Isolate code | Cellulolytic index |
|---|---|
| LG2 | 13.96 |
| LG3 | 10.60 |
| LG7 | 6.74 |
| LG19 | 6.35 |
| LG18 | 4.83 |
| LG14 | 4.08 |
| LG73 | 3.91 |
| LG115 | 3.80 |
| LG139 | 3.64 |
| LG20 | 3.42 |
| LG13 | 3.21 |
| LG16 | 3.07 |

In the phylogenetic tree, LG2 was closely related to the *Fictibacillus* group (Figure 4), suggesting that they have a common ancestor and share some features and functionality (Table 2, Figure 4). The genus *Fictibacillus* is Gram-positive, motile, rod-shaped, endospore-forming bacteria (Pal et al. 2018); however, studies on their cellulolytic activity are still limited. A study by Chen et al. (2020) showed that *Fictibacillus* sp. YS-26 isolated from soft corals in Yongxing Island, Hainan Province, China, have high cellulolytic activity and can degrade lignocellulose. The cellulolytic activity of *Fictibacillus* from mangrove sediments has never been reported. Therefore, the results of this study are considered as new information on the cellulolytic activity of *Fictibacillus* from mangroves.

Bacterial species that adapt to mangrove ecosystems are a potential source for biotechnology. Mangroves are a rich resource for discovering new bacterial and fungal species that produce enzymes and molecules to be used in human life, i.e., agriculture, industry, and bioremediation (Dias et al. 2009; Dourado et al. 2012). The ability to convert lignocellulosic substrates into simple nutrients is crucial for the carbon cycle and microbial survival, especially in high substrates environments. Lignocellulosic materials are very difficult to degrade due to their dense, compact structures, which protect plants against microbial attack (Kumar et al. 2008). Microorganisms that can degrade plant biomass might only be found in specific environments, such as mangrove ecosystems, where the biomass is abundant.

In conclusion, bacteria with cellulolytic activity were obtained from Logending mangrove sediments. Based on the phylogenic analysis, isolate LG2 is similar to the *Fictibacillus* group. This study provides new information on the cellulolytic activity of *Fictibacillus* from mangroves. It can produce enzymes and chemicals that can be beneficial for human life, such as agriculture, industry, and bioremediation.

**Table 2.** The similarity of LG2 isolate to other related strains using BLAST-N

| Description | Max score | Total score | Query cover | E value | Per. Ident | Accession |
|---|---|---|---|---|---|---|
| *Fictibacillus nanhaiensis* strain JSM 082006 16S ribosomal RNA, partial sequence | 2571 | 2571 | 99% | 0.0 | 99.86% | NR_117524.1 |
| *Fictibacillus halophilus* strain AS8 16S ribosomal RNA, partial sequence | 2567 | 2567 | 100% | 0.0 | 99.79% | NR_149289.1 |
| *Fictibacillus phosphorivorans* strain Ca7 16S ribosomal RNA, partial sequence | 2567 | 2567 | 99% | 0.0 | 99.86% | NR_118455.1 |
| *Fictibacillus barbaricus* strain V2-BIII-A2 16S ribosomal RNA, partial sequence | 2484 | 2484 | 99% | 0.0 | 98.85% | NR_028967.1 |
| *Fictibacillus aquaticus* strain GDSW-R2A3 16S ribosomal RNA, partial sequence | 2446 | 2446 | 100% | 0.0 | 98.21% | NR_159291.1 |
| *Fictibacillus arsenicus* strain Con a/3 16S ribosomal RNA, partial sequence | 2386 | 2386 | 100% | 0.0 | 97.43% | NR_042217.1 |
| *Fictibacillus rigui* strain WPCB074 16S ribosomal RNA, partial sequence | 2350 | 2350 | 100% | 0.0 | 97.00% | NR_116518.1 |
| *Fictibacillus enclensis* strain NIO-1003 16S ribosomal RNA, partial sequence | 2320 | 2320 | 99% | 0.0 | 96.64% | NR_133744.1 |
| *Fictibacillus solisalsi* strain YC1 16S ribosomal RNA, partial sequence | 2300 | 2300 | 99% | 0.0 | 96.43% | NR_044387.1 |
| *Bacillus tianmuensis* strain B5 16S ribosomal RNA, partial sequence | 2290 | 2290 | 100% | 0.0 | 96.22% | NR_116701.1 |

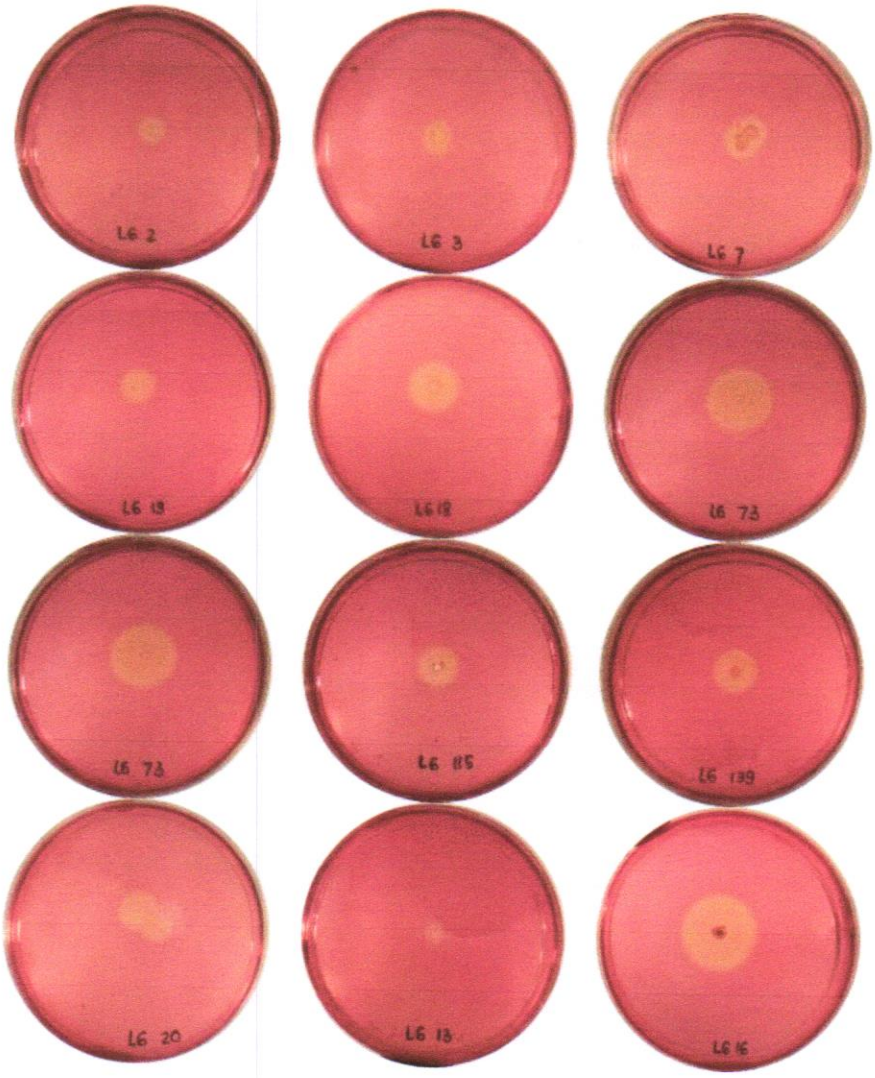


**Figure 3.** Cellulolytic activity of some bacterial isolates collected from Logending mangrove sediments

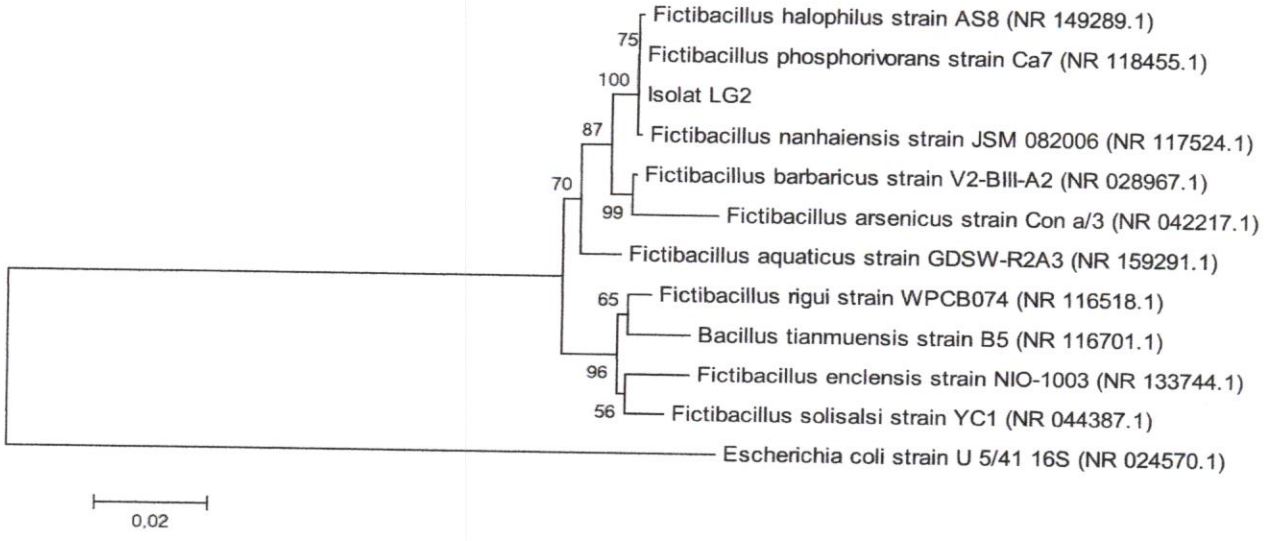


**Figure 4.** Phylogenetic tree of isolate LG2 and its closely related strains based on 16S rRNA gene sequence in the GenBank database

## ACKNOWLEDGEMENTS

We thank Soewarto, Rifda Naufalin, and Imam Widhiono for their support in obtaining research funding and to all of the students who helped collect field data. This research was supported by a Directorate General of Higher Education Research Fellowship and the Universitas Jenderal Soedirman, Purwokerto, Central Java, Indonesia. The authors declare that there are no conflicts of interest regarding the publication of this paper

## REFERENCES

Ambeng Zubair H, Oka NP, Tonggiroh A. 2019. Isolation and characterization of bacteria from mangrove sediment at coastal area in Pangkep South Sulawesi. J Phys: Conf Ser 1341(2): 022016. DOI: 10.1088/1742-6596/1341/2/022016.

Behera BC, Parida S, Dutta SK, Thatoi HN. 2014. Isolation and identification of cellulose degrading bacteria from mangrove soil of mahanadi river delta and their cellulase production ability. Am J Microbiol Res 2 (1): 41-46. DOI:10.12691/ajmr-2-1-6.

Basak P, Pramanik A, Sengupta S, Nag S, Bhattacharyya A, Pattanayak R, Ghosh A. Chattopadhyay D, Roy D, Bhattacharyya M. 2016. Bacterial diversity assessment of pristine mangrove microbial community from Dhulibhashani, Sundarbans using 16S rRNA gene tag sequencing. Genomic Data 7: 76-8. DOI: 10.1016/j.gdata.2015.11.030.

Bibi F, Ullah I, Akhtar S, Yasir M, Kensarah EA, AL-Ghamadi AAK, Azhar EI. 2017. Diversity and bioprospecting potential of rhizo and endophytic bacteria from two mangrove plants in Saudi Arabia. Afr J Microbiol Res 11 (19): 729-739. DOI: 10.5897/AJMR2017.8552.

Chantarasiri A. 2015. Aquatic *Bacillus cereus* JD0404 isolated from the muddy sediments of mangrove swamps in Thailand and characterization of its cellulolytic activity. Egypt J Aquat Res 41 (3): 257-264. DOI: 10.1016/j.ejar.2015.08.003.

Chen Y, Wang W, Zhou D, Jing T, Li K, Zhao Y, Tang W, Qi D, Zhang M, Zang X, Luo Y. 2020. Biodegradation of lignocellulosic agricultural residues by a newly isolated *Fictibacillus* sp. YS-26 improving carbon metabolic properties and functional diversity of the rhizosphere microbial community. Bioresour Technol. DOI: 10.1016/j.biortech.2020.123381.

Dias AC, Andreote FD, Dini-Andreote F, Lacava PT, Sá AL, Melo IS, Azevedo JL, Araújo WL. (2009). Diversity and biotechnological potential of culturable bacteria from Brazilian mangrove sediment. World J Microbiol Biotechnol 25 (7): 1305-1311. DOI: 10.1007/s11274-009-0013-7.

Dourado MN, Ferreira A, Araújo WL, Azevedo JL, Lacava PT. 2012. The diversity of endophytic methylotrophic bacteria in an oil-contaminated and an oil-free mangrove ecosystem and their tolerance to heavy metals. Biotechnol Res Intl. DOI:10.1155/2012/759865.

Glaeser SP, Dolt W, Busse HJ, Kampfer P. 2013. *Fictibacillus phosphorivorans* gen. nov., sp. nov. and proposal to reclassify *Bacillus arsenicus, Bacillus barbaricus, Bacillus macauensis, Bacillus nanhaiensis, Bacillus rigui, Bacillus solisalsi* and *Bacillus gelatini* in the genus *Fictibacillus*. Intl J Syst Evol Microbiol 63: 2934-2944. DOI 10.1099/ijs.0.049171-0.

Hastuti SU, Sangur K, Khasanah HN. 2015. Biodiversity and enzyme activity of indigenous cellulolytic and amylolytic bacteria in decayed mangrove stem waste product at Waai Seashore, Ambon Island. J Agric Sci Technol B 5: 252-256. DOI: 10.17265/2161-6264/2015.04.004.

Klijn NI, Weerkamp AH, de Vos WM. 1991. Identification of mesophilic lactic acid bacteria by using polymerase chain reaction-amplified variable regions of 16S rRNA and specific DNA probes. Appl Environ Microbiol 57 (11): 3390-3393.

Kumar R, Singh S, Singh OV. 2008. Bioconversion of lignocellulosic biomass: biochemical and molecular perspectives. J Ind Microbiol Biotechnol 35 (5): 377-391. DOI: 10.1007/s10295-008-0327-8.

Kurniawan A, Puspitasari S, Asriani E, Kurniawan A, Sambah AB, Prihanto AA. 2019. Molecular identification of cellulolytic bacteria from mangrove sediment at Tin Minning Region in West Bangka. Intl J Appl Biol 3 (1): 7-14. DOI: 10.20956/ijab.v3i1.5848.

Kurniawan A, Prihanto AA, Sari SP, Febriyanti D, Kurniawan A, Sambah AB, Asriani E. 2017. Isolation and Identification of cellulolytic bacteria from mangrove sediment in Bangka Island. In IOP Conf Ser: Earth Environ Sci 137 (1): 012070. DOI: 10.1088/1755-1315/137/1/012070

Liu M, Huiqin H, Bao S, Tong Y. 2019. Microbial community structure of soils in Bamenwan mangrove wetland. Sci Rep 9: 8406. DOI: 10.1038/s41598-019-44788-x.

Maharsiwi W, Astuti RI, Meryandini A, Wahyudi AT. 2020. Screening and characterization of sponge-associated bacteria from Seribu Island, Indonesia producing cellulase and laccase enzymes. Biodiversitas 21 (3): 975-981. DOI: 10.13057/biodiv/d210317.

Naresh S, Kunasundari B, Gunny AAN, Teoh YP, Shuit SH, Ng QH, Hoo PY. 2019. Isolation and partial characterisation of thermophilic cellulolytic bacteria from North Malaysian Tropical Mangrove Soil. Trop Life Sci Res 30 (1): 123-147. DOI: 10.21315/tlsr2019.30.1.8.

Nursyirwani N, Feliatra F, Tanjung A, Harjuni F. 2020. Isolation of cellulolytic bacteria from mangrove sediment in Dumai Marine Station Riau and the antibacterial activity against pathogens. IOP Conf Ser: Earth Environ Sci 430 (1): 012012. DOI: 10.1088/1755-1315/430/1/012012.

Pal D, Bhardwaj A, Kaur N, Krishnamurthi S, Sudan SK, Bisht B, Vyas B, Kumari M and Mayilraj S. 2018. *Fictibacillus aquaticus* sp. nov., isolated from downstream river water. Intl J Syst Evol Microbiol 68: 160-164. DOI: 10.1099/ijsem.0.002474.

Rajagopal G, Kannan S. 2017. Systematic characterization of potential cellulolytic marine Actinobacteria *Actinoalloteichus* sp. MHA15. Biotechnol Rep 13: 30-36. DOI: 10.1016/j.btre.2016.12.003.

Sakhia N, Sunil P, Shetty V, Bhatt S, Bhadalkal A, 2016. Study of Bacterial diversity of Mangrove Rhizosphere. Open J Mar Sci 6: 23-31. DOI: 10.4236/ojms.2016.61003.

Tamura T, Stecher G, Peterson D, Filipski A, Kumar S. 2013. MEGA6: Molecular evolutionary genetics analysis version 6.0. Mol Biol Evol 30 (12): 2725-2729. DOI: 10.1093/molbev/mst197.

Thompson CE, Beys-da-Silva WO, Santi L, Berger M, Vainstein MH, Guimarães JA, Vasconcelos ATR. 2013. A potential source for cellulolytic enzyme discovery and environmental aspects revealed through metagenomics of Brazilian mangroves. AMB Expr 3: 65. DOI: 10.1186/2191-0855-3-65.

# TURNITIN

# Short Communication: Diversity of cellulolytic bacteria isolated from coastal mangrove sediment in Logending Beach, Kebumen, Indonesia

*by* Eming Sudiana

**Submission date:** 27-Mar-2021 07:56PM (UTC+0700)
**Submission ID:** 1543771299
**File name:** BIOD_SYARAT_WAJIB_2021_REV.pdf (753.62K)
**Word count:** 5585
**Character count:** 25232

**BIODIVERSITAS**
Volume 22, Number 4, April 2021
Pages: 1869-1878

ISSN: 1412-033X
E-ISSN: 2085-4722
DOI: 10.13057/biodiv/d220433

# Short Communication: Diversity of cellulolytic bacteria isolated from coastal mangrove sediment in Logending Beach, Kebumen, Indonesia

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Manuscript received: 28 October 2020. Revision accepted: 22 March 2021.

**Abstract.** *Pramono H, Mariana A, Ryandini D, Sudiana E. 2021. Short Communication: Diversity of cellulolytic bacteria isolated from coastal mangrove sediment in Logending Beach, Kebumen, Indonesia. Biodiversitas 22: 1869-1878.* Bacteria from mangrove sediments have the potential to produce active enzymes and metabolites needed in the industry. Mangrove ecosystems in various parts of Indonesia have been explored as sources of bacteria. However, the mangrove ecosystems in Logending Beach, Kebumen District, Indonesia are still unexplored. This study aims to determine the diversity of cellulolytic bacteria using the 16S rRNA gene. Ninety-nine bacteria were isolated from Logending coastal mangrove sediments. Most of them (87.87%) had a cellulolytic activity, with the cellulolytic index values ranged from 0.25–13.96. The LG2 isolate had the highest cellulolytic activity (13.96). Molecular characterization of the LG2 isolate based on the 16S rRNA gene showed that it was similar to the *Fictibacillus nanhaiensis* strain JSM 082006. This result is new information on cellulolytic bacteria and might be used as a future source of cellulolytic enzymes.

**Keywords**: 16S rRNA gene cellulolytic, mangrove, molecular, sediments

## INTRODUCTION

Mangrove forests are very productive ecosystems and a potential source of superior bacteria that produce active metabolites and industrial enzymes. In tropical mangroves, 91% of the microbial biomass is bacteria and fungi (Naresh et al. 2019). The existence of microorganisms in mangrove ecosystems is closely related to the stability of the ecosystem. Extracellular enzymes produced by microbes are used to break down complex organic material used by cells as a nutrition source (Thompson et al. 2013). The complexity of the mangrove ecosystems and anthropogenic pressure require microbes to produce extracellular enzymes that function under these complex conditions. Enzymes are susceptible to environmental factors; however, the enzymes produced by microbes in complex ecosystems have a high tolerance to environmental factors (Pal et al. 2018). Mangroves harbor potential bacteria that produce active metabolites and enzymes (Bibi et al. 2017).

The study of microbial interactions with other ecosystem components, such as plant roots, is important for understanding the function and remediation of mangrove ecosystems (Chantarasiri 2015). The bacteria in the mangrove ecosystems break down mangrove leaf litter into organic material used as a source of nutrition by the organisms living in mangrove forests (Glaeser et al. 2013). Environmental factors strongly influence the diversity of bacteria in mangrove ecosystems. Sakhia et al. (2016) showed that bacterial species are affected by mangrove vegetation in the water, especially towards the sea (Liu et al. 2019). Although many microorganisms can degrade cellulose, only a few produce significant quantities of cell-free bioactive compounds capable of completely hydrolyzing crystalline cellulose in vitro (Rajagopal and Kannan 2015).

Mangrove bacteria can degrade cellulose (Behera et al. 2013; Basak et al. 2016). In a study of mangrove sediments in Dhulibhashani, Sundarbands, India, Basak et al. (2016) found that Proteobacteria, including Bacteroidetes, Acidobacteria, Firmicutes, Actinobacteria, Nitrospirae, Cyanobacteria, Planctomycetes, and Fusobacteria, were dominant using 16S rRNA gene tag sequencing. *Micrococcus*, *Bacillus*, *Pseudomonas*, *Xanthomonas*, and *Brucella* spp. were collected from the Mahanadi River Delta in Odisha, India, which has a high capacity carboxymethyl cellulose hydrolysis (Behera et al. 2013). In Andaman Island mangroves, Rajagopal and Kannan (2017) isolated 19 morphologically distinct Actinobacteria strains from Andaman Island mangroves and 4 strains possessed good cellulose degradation activity, of which strain MHA15 produced the highest cellulase (14,379 IU/mL). Taxonomically, the four strains are members of the genus *Actinoalloteichus* differing from *Actinoalloteichus cyanogriseus* (Behera et al. 2013). Pal et al. (2018) isolated bacteria strains from the Indian Ganges Delta that belongs to the family Bacillaceae and related to the genus *Fictibacillus* based on 16S rRNA sequencing.

Several studies have been carried out on the cellulolytic bacterial diversity of several mangrove ecosystems in Indonesia. Hastuti et al. (2015) found four cellulolytic bacteria species (*Micrococcus luteus*, *Bacillus pumilus*, *Planococcus citreus*, and *Bacillus cereus*) that originated

from mangrove bark on Waai Beach, Ambon Island. Ambeng et al. (2019) isolated 35 cellulolytic bacteria from mangrove sediments in Pangkep, South Sulawesi, belonging to seven genera, i.e., *Bacillus*, *Staphylococcus*, *Vibrio*, *Micrococcus*, *Alteromonas*, *Escherichia*, and *Listeria*. Kurniawan et al. (2017) isolated *Bacillus pumilus*, *Pseudomonas* sp., *Bacillus amyloliquefaciens*, *Bacillus alvei*, and *Bacillus coagulant* from mangrove sediment on Bangka Island, and *Pseudomonas aeruginosa,* which produced the highest cellulase. Nursyiwani et al. (2020) examined 24 isolates of cellulolytic antimicrobial pathogens from mangrove sediment samples from Dumai, Riau and found that 9 isolates could inhibit the development of pathogenic microbes; and 3 of these isolates were similar to *Bacillus toyonensis*.

Maharsiwi et al. (2020) has identified cellulolytic bacteria on mangrove sponges from the Seribu Islands similar to *Pseudomonas luteola* strain NBRC 103146, *Bacillus cereus* strain 24k, *Pseudomonas aeruginosa* strain DSM 50071, *Microbacterium maritypicum* strain DSM 20578, and *Brachybacterium conglomeratum* strain J 1015. Kurniawan et al. (2019) isolated bacteria from the tin-mining region in West Bangka and identified them as *Bacillus amyloliquefaciens*.

Since there has been no study on bacterial diversity in the mangrove area of the southern coast of Java, especially at Logending Beach, Kebumen, we conducted the study to determine the diversity of cellulolytic bacteria mangrove sediments and identified using 16S rRNA sequencing.

## MATERIALS AND METHODS

### Study site

Sediments were collected at six locations in the mangrove forest in Logending Beach, Ayah Subdistrict, Kebumen District, Central Java Province, Indonesia (7° 42' 28.0'' S and 109° 23' 20.1" E). Two locations are at the estuary and three locations along the coast (Figure 1). The samples were processed in the Microbiology Laboratory of the Faculty of Biology, Jenderal Soedirman University, Purwokerto, Banyumas, Indonesia.

### Isolation of mangrove sediment bacteria

Bacteria were isolated using the pour plate technique on nutrient agar (NA) medium. Ten g of sediment samples were homogenized in 90 mL physiological saline and then diluted up to $10^{-5}$ fold. The last three dilutions were cultured in NA medium and incubated for 24 hours at room temperature. Bacterial colonies grow on the diluted medium of $10^{-3}$, $10^{-4}$, and $10^{-5}$ were counted, and the bacterial density was reported as the total number of bacteria per gram of mangrove sediment. Bacterial diversity was observed from colony morphology. The colonies were further purified by the quadrant streak technique to isolate and characterize bacterial isolates.


**Figure 1.** Sampling locations (white circles) in the mangrove forest of Logending Beach, Ayah Subdistrict, Kebumen District, Central Java Province, Indonesia

**Cellulolytic activity test**

Bacterial isolates were inoculated on CMC agar and incubated for 24 hours. The cellulolytic activity was observed by dripping Congo Red solution on the culture surface. The formation of clear zones around a colony indicates cellulolytic activity. The clear zone and colony diameters were measured, and enzymatic activity was calculated by dividing the diameter of the clear zone by the colony diameter.

**Isolation of bacterial genomic DNA**

Bacterial chromosomal DNA was isolated using the method of Klijn et al. (1991) with some modifications. A single bacterial colony was suspended in 50 µL 10 mM Tris-HCl (pH 8) buffer solution containing 400 µg lysozyme and incubated for 1 hour at 37°C. Cell lysis was improved by adding 50 µL 10% SDS and 250 µL buffer. The mixture was centrifuged at 5,000 × *g* for 10 minutes. The supernatant was transferred to a new tube, and 60 mL 3 M sodium acetate and 1 mL 96% chilled ethanol were added. After centrifuging at 12,000 × *g* for 15 minutes, the DNA pellet was placed in 300 *µ*L 70% ethanol and centrifuged at 8,000 × *g* for 5 minutes. The pellet was dried at room temperature until all the ethanol evaporated, and then it was dissolved in 50 µL TE buffer. The chromosomal DNA was analyzed by agarose gel electrophoresis.

**16S rRNA gene amplification and phylogenetic analysis**

The 16S rRNA gene was amplified by polymerase chain reaction (PCR) using a DNA chromosome template. The PCR mixture consisted of 8.55 *µ*L $ddH_2O$, 3 *µ*L 10× PCR buffer, 0.45 *µ*L 5 mM dNTP, 1.05 *µ*L 0.6 mM $MgCl_2$, 2 *µ*L primers, 0.15 *µ*L *Taq* polymerase, and 0.6 *µ*L chromosomal DNA. PCR analysis was performed on a thermocycler, with an initial denaturation step at 94°C for 4 min, followed by 34 cycles of 94°C for 30 s, 50°C for 1 min, and 72°C for 2 min. The primers used to amplify the 16S rRNA gene were Bact27F (5'-AGAGTTTGATCATG GCTCAG-3') and Uni1492R (5'-GGTTACCTTGT TACGACTT-3'). The PCR products were verified by agarose gel electrophoresis and then sequenced.

**Data analysis**

Using a BLAST search, selected 16S rRNA sequences were compared to other sequences in GenBank to determine the level of similarity. Then, a phylogenetic tree of the 16S rRNA sequences was constructed using MEGA ver. 6. (Tamura et al. 2013)

## RESULTS AND DISCUSSION

**Diversity of cellulolytic bacteria in coastal mangrove sediment from Logending**

In total, there were 99 bacteria strains isolated from the Logending mangrove sediments (Table 1). The highest number of isolates was obtained from the first sampling location (58 isolates), followed by the 3rd and 5th sampling locations (53 isolates from each location), and the lowest number was obtained from the 6th sampling location (14 isolates) (Figure 2). The first sampling point is directly opposite the river flow direction, while the 6th sampling location faces the sea. The difference in the number of isolates among sampling points is probably due to the sediment type. The result showed that 87 bacterial isolates have cellulolytic activity, as indicated by a clear zone around the colony on CMC agar containing Congo Red (Figure 3).

The cellulolytic index (CI) reflects the ability of the cellulase produced by bacterial isolates to degrade cellulose on CMC agar. The CI values ranged from 0.25 to 13.96 (Table 1), and 12 isolates had a CI > 3.0. The highest CI (13.96) was produced by the LG2 isolate (Table 1).

Mangrove sediments in Logending Beach have a high potential for cellulolytic bacteria, and only a few isolates have lower CI values than in other mangrove areas in Indonesia. Kurniawan et al. (2017) only found five cellulolytic bacteria from mangrove sediments at Sungailiat and Tukak Sadai on Bangka Island. Nursyirwani et al. (2020) obtained 24 cellulolytic bacterial isolates with CI values ranging from 1.00 to 2.86.

Many cellulolytic bacteria have been isolated from sediments and tropical mangrove soils. Functional metagenomic analyses have shown that mangrove land has high cellulolytic enzyme activity. Specific sequences involved in cellulose degradation were identified in mangroves from Brazil (Thompson et al. 2013). High cellulolytic activity can be found in sediments with high mangrove litter levels, which creates a cellulose-rich environment for bacterial populations.

**Identification of 16S rRNA genes**

Based on 16S rRNA sequences and using BLAST from NCBI, it showed the LG2 isolate had the highest similarity (99.86%) to *Fictibacillus nanhaiensis* strain JSM 082006 in GenBank (Table 2). Other closely related strains were *F. halophilus* (99.79%), *F. phosphorivorans* (99.86%), *F. barbaricus* (98.21%), *F. aquaticus* (98.21%), *F. arsenicus* (97.43.0%), *F. rigui* (97%), *F. enclensis* (96.64%), *F. solisalsi* (96.43%), and *Bacillus tianmuensis* (96.22%) (Table 2).

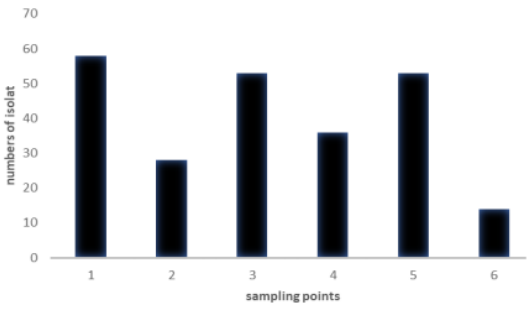


**Figure 2.** Number of isolates in each sampling locations

**Table 1.** Isolat richness and densities (means + stdev)

| Isolate name | Sampling points (n = 6) | | | | | |
|---|---|---|---|---|---|---|
| | 1 | 2 | 3 | 4 | 5 | 6 |
| LG 1 | 0 | 0 | 0 | 0 | 2500,00 ± 577,35 | 0 |
| LG 2 | 633,33 ± 115,47 | 25125,00 ± 3544,36 | 3000,00 ± 433,01 | 338333,30 ± 17559,42 | 17833,33 ± 2565,80 | 16833,33 ± 288,68 |
| LG 3 | 0 | 0 | 0 | 2166,67 ± 577,35 | 5000,00 ± 500,00 | 0 |
| LG 4 | 350000,00 ± 28867,51 | 1250,00 ± 204,12 | 16666,67 ± 1443,38 | 0 | 453333,30 ± 45092,50 | 500,00 ± 86,60 |
| LG 5 | 0 | 0 | 0 | 0 | 5000,00 ± 500,00 | 0 |
| LG 6 | 0 | 250000,00 ± 40824,83 | 33333,33 ± 5773,50 | 4166,67 ± 577,35 | 18333,33 ± 2886,75 | 16833,33 ± 288,68 |
| LG 7 | 40000,00 ± 8231,04 | 19969125,00 ± 1353518,77 | 58333,33 ± 5773,50 | 183333,30 ± 28867,51 | 54000,00 ± 3605,55 | 16666,67 ± 2886,75 |
| LG 8 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 9 | 0 | 0 | 166,67 ± 14,43 | 0 | 0 | 0 |
| LG 10 | 0 | 0 | 333,33 ± 43,30 | 0 | 16666,67 ± 2886,75 | 0 |
| LG 11 | 0 | 0 | 2333,33 ± 144,34 | 0 | 35000,00 ± 5000,00 | 0 |
| LG 12 | 0 | 0 | 17500,00 ± 1322,88 | 0 | 0 | 0 |
| LG 13 | 13333,33 ± 2886,75 | 62500,00 ± 5000,00 | 8333,33 ± 2886,75 | 5000,00 ± 288,68 | 18333,33 ± 2886,75 | 0 |
| LG 14 | 1666,67 ± 288,68 | 50375,00 ± 3685,56 | 500,00 ± 43,30 | 500,00 ± 50,00 | 17833,33 ± 2565,80 | 0 |
| LG 15 | 0 | 0 | 0 | 0 | 19000,00 ± 1732,05 | 0 |
| LG 16 | 0 | 125000,00 ± 28867,51 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 17 | 46166,67 ± 5392,90 | 3000,00 ± 408,25 | 103333,30 ± 2886,75 | 15000,00 ± 2500,00 | 8500,00 ± 866,03 | 334000,00 ± 27712,81 |
| LG 18 | 0 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 |
| LG 19 | 0 | 125000,00 ± 41231,06 | 0 | 0 | 173333,30 ± 46188,02 | 0 |
| LG 20 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 21 | 0 | 0 | 666,67 ± 57,74 | 1666,67 ± 288,68 | 0 | 0 |
| LG 22 | 0 | 0 | 0 | 11666,67 ± 2886,75 | 16666,67 ± 2886,75 | 0 |
| LG 23 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 |
| LG 24 | 0 | 0 | 0 | 7666,67 ± 577,35 | 0 | 0 |
| LG 25 | 1328333,00 ± 183462,08 | 149625,00 ± 33623,84 | 38333,33 ± 7637,63 | 1666,67 ± 288,68 | 9333,33 ± 2081,67 | 17666,67 ± 3511,88 |
| LG 26 | 0 | 0 | 1666,67 ± 152,75 | 166666,70 ± 28867,51 | 0 | 0 |
| LG 27 | 1666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 28 | 1666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 29 | 5000,00 ± 0,00 | 0 | 0 | 15000,00 ± 2000,00 | 0 | 0 |
| LG 30 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 31 | 0 | 0 | 0 | 3333,33 ± 288,68 | 3500,00 ± 500,00 | 0 |
| LG 32 | 3333,33 ± 577,35 | 125,00 ± 20,41 | 0 | 0 | 0 | 0 |
| LG 33 | 500,00 ± 86,60 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 34 | 1666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 35 | 18333,33 ± 2886,75 | 0 | 16666,67 ± 5773,50 | 166,67 ± 28,87 | 25000,00 ± 2886,75 | 0 |
| LG 36 | 6666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 37 | 1833,33 ± 144,34 | 0 | 0 | 0 | 0 | 0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| LG 38 | 6666,67 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 39 | 3333,33 ± 577,35 | 0 | 0 | 0 | 0 | 0 |
| LG 40 | 1666,67 ± 381,88 | 0 | 0 | 0 | 0 | 0 |
| LG 41 | 333,33 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 42 | 16833,33 ± 2753,79 | 0 | 0 | 0 | 16666,67 ± 2886,75 | 0 |
| LG 43 | 166,67 ± 28,87 | 0 | 0 | 0 | 500,00 ± 50,00 | 0 |
| LG 44 | 3500,00 ± 288,68 | 0 | 0 | 0 | 0 | 0 |
| LG 45 | 333,33 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 46 | 2500,00 ± 500,00 | 0 | 7500,00 ± 866,03 | 8333,33 ± 1154,70 | 166833,30 ± 28724,26 | 0 |
| LG 47 | 0 | 0 | 0 | 0 | 333,33 ± 28,87 | 0 |
| LG 48 | 16666,67 ± 2886,75 | 0 | 5000,00 ± 866,03 | 0 | 3833,33 ± 763,76 | 0 |
| LG 49 | 0 | 1250,00 ± 288,68 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 50 | 0 | 0 | 9000,00 ± 1322,88 | 2666,67 ± 288,68 | 1666,67 ± 288,68 | 0 |
| LG 51 | 0 | 0 | 0 | 0 | 33666,67 ± 2309,40 | 0 |
| LG 52 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 53 | 13333,33 ± 1732,05 | 0 | 0 | 833,33 ± 144,34 | 0 | 0 |
| LG 54 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 55 | 15000,00 ± 866,03 | 0 | 40000,00 ± 1000,00 | 16666,67 ± 2886,75 | 1000,00 ± 100,00 | 166,67 ± 28,87 |
| LG 56 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 57 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 | 0 |
| LG 58 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 59 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 | 0 |
| LG 60 | 666,67 ± 57,74 | 100000,00 ± 20412,41 | 0 | 167000,00 ± 28583,21 | 0 | 0 |
| LG 61 | 0 | 110000,00 ± 8660,25 | 0 | 0 | 2666,67 ± 288,68 | 0 |
| LG 62 | 666,67 ± 57,74 | 0 | 500,00 ± 50,00 | 0 | 0 | 0 |
| LG 63 | 666,67 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 64 | 166,67 ± 28,87 | 0 | 0 | 0 | 0 | 0 |
| LG 65 | 333,33 ± 57,74 | 0 | 0 | 0 | 0 | 0 |
| LG 66 | 166,67 ± 28,87 | 0 | 0 | 0 | 0 | 0 |
| LG 67 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 |
| LG 68 | 0 | 662500,00 ± 75000,00 | 8666,67 ± 1527,53 | 1666,67 ± 763,76 | 0 | 0 |
| LG 69 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 70 | 0 | 0 | 0 | 0 | 5000,00 ± 500,00 | 0 |
| LG 71 | 0 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 |
| LG 72 | 0 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 |
| LG 73 | 0 | 0 | 0 | 1666,67 ± 288,68 | 0 | 0 |
| LG 74 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 |
| LG 75 | 0 | 0 | 0 | 6666,67 ± 577,35 | 0 | 0 |
| LG 76 | 0 | 0 | 0 | 15000,00 ± 1000,00 | 0 | 0 |
| LG 77 | 0 | 0 | 0 | 333,33 ± 28,87 | 0 | 0 |
| LG 78 | 0 | 0 | 50000,00 ± 5000,00 | 666,67 ± 144,34 | 0 | 0 |
| LG 79 | 0 | 0 | 0 | 333,33 ± 28,87 | 0 | 0 |
| LG 80 | 0 | 0 | 0 | 6666,67 ± 577,35 | 50000,00 ± 5000,00 | 0 |
| LG 81 | 133333,30 ± 14433,76 | 0 | 3333,33 ± 763,76 | 0 | 13166,67 ± 1154,70 | 166,67 ± 28,87 |
| LG 82 | 0 | 0 | 4500,00 ± 866,03 | 0 | 1166,67 ± 288,68 | 0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| LG 83 | 0 | 0 | 333,33 ± 57,74 | 0 | 166,67 ± 72,17 | 0 |
| LG 84 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 85 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 86 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 87 | 0 | 0 | 9833,33 ± 288,68 | 0 | 0 | 0 |
| LG 88 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 89 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 90 | 0 | 0 | 333,33 ± 57,74 | 0 | 0 | 0 |
| LG 91 | 0 | 375,00 ± 50,00 | 0 | 0 | 0 | 0 |
| LG 92 | 2000,00 ± 433,01 | 0 | 0 | 0 | 0 | 0 |
| LG 93 | 1500,00 ± 346,41 | 0 | 166666,70 ± 28867,51 | 0 | 0 | 0 |
| LG 94 | 3333,33 ± 288,68 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 |
| LG 95 | 0 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 |
| LG 96 | 0 | 0 | 0 | 0 | 28333,33 ± 7637,63 | 0 |
| LG 97 | 0 | 0 | 0 | 500,00 ± 50,00 | 0 | 0 |
| LG 98 | 0 | 0 | 500,00 ± 50,00 | 0 | 0 | 0 |
| LG 99 | 0 | 0 | 1166,67 ± 288,68 | 0 | 0 | 0 |
| LG 100 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 101 | 5000,00 ± 866,03 | 0 | 0 | 0 | 0 | 0 |
| LG 102 | 5000,00 ± 866,03 | 0 | 0 | 0 | 0 | 0 |
| LG 103 | 16666,67 ± 2886,75 | 875,00 ± 150,00 | 17166,67 ± 2565,80 | 0 | 0 | 0 |
| LG 104 | 0 | 0 | 500,00 ± 50,00 | 0 | 0 | 0 |
| LG 105 | 0 | 0 | 1166,67 ± 288,68 | 0 | 0 | 0 |
| LG 106 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 107 | 0 | 0 | 0 | 3333,33 ± 288,68 | 0 | 0 |
| LG 108 | 33333,33 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 109 | 16666,67 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 110 | 16666,67 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 111 | 16666,67 ± 2886,75 | 0 | 0 | 0 | 0 | 0 |
| LG 112 | 166666,70 ± 158771,32 | 0 | 0 | 0 | 0 | 0 |
| LG 113 | 0 | 0 | 0 | 0 | 25000,00 ± 2886,75 | 0 |
| LG 114 | 0 | 12500,00 ± 2516,61 | 16666,67 ± 2886,75 | 16666,67 ± 2886,75 | 0 | 0 |
| LG 117 | 0 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 |
| LG 118 | 0 | 150000,00 ± 8164,97 | 0 | 0 | 0 | 0 |
| LG 119 | 16666,67 ± 2886,75 | 0 | 0 | 3666,67 ± 288,68 | 0 | 0 |
| LG 120 | 0 | 1250,00 ± 300,00 | 0 | 0 | 0 | 0 |
| LG 121 | 0 | 0 | 0 | 0 | 0 | 333333,30 ± 28867,51 |
| LG 122 | 0 | 125,00 ± 33,67 | 16666,67 ± 2886,75 | 0 | 0 | 0 |
| LG 123 | 0 | 0 | 0 | 0 | 0 | 16666,67 ± 2886,75 |
| LG 126 | 0 | 0 | 16666,67 ± 2886,75 | 0 | 166666,70 ± 28867,51 | 166666,70 ± 28867,51 |
| LG 125 | 0 | 250,00 ± 40,82 | 0 | 0 | 0 | 0 |
| LG 127 | 0 | 125,00 ± 45,64 | 0 | 0 | 0 | 0 |
| LG 128 | 0 | 1250,00 ± 191,49 | 0 | 0 | 0 | 0 |

| | | | | | | |
|---|---|---|---|---|---|---|
| LG 129 | 0 | 0 | 16666,67 ± 2886,75 | 0 | 166666,70 ± 28867,51 | 0 |
| LG 130 | 0 | 0 | 166666,70 ± 28867,51 | 0 | 0 | 0 |
| LG 131 | 0 | 0 | 166666,70 ± 28867,51 | 0 | 0 | 0 |
| LG 134 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 | 0 |
| LG 135 | 33333,33 ± 31754,26 | 0 | 0 | 0 | 0 | 0 |
| LG 136 | 0 | 0 | 0 | 0 | 166666,70 ± 28867,51 | 0 |
| LG 137 | 50000,00 ± 8660,25 | 0 | 0 | 0 | 0 | 0 |
| LG 138 | 0 | 0 | 0 | 0 | 333333,30 ± 28867,51 | 0 |
| LG 139 | 0 | 0 | 0 | 0 | 166666,70 ± 28867,51 | 0 |
| LG 140 | 0 | 1250,00 ± 191,49 | 0 | 0 | 0 | 0 |
| LG 141 | 0 | 2500,00 ± 95,74 | 0 | 0 | 0 | 0 |
| LG 142 | 0 | 500,00 ± 81,65 | 0 | 0 | 0 | 0 |
| LG 143 | 0 | 0 | 0 | 166,67 ± 28,87 | 0 | 0 |
| LG 144 | 0 | 0 | 0 | 0 | 0 | 333,33 ± 28,87 |
| LG 145 | 0 | 0 | 0 | 0 | 0 | 166,67 ± 28,87 |

**Table 2.** The cellulolytic index of bacterial isolates collected from Logending coastal mangrove sediment

| Isolate code | Cellulolytic index |
|---|---|
| LG2 | 13.96 |
| LG3 | 10.60 |
| LG7 | 6.74 |
| LG19 | 6.35 |
| LG18 | 4.83 |
| LG14 | 4.08 |
| LG73 | 3.91 |
| LG115 | 3.80 |
| LG139 | 3.64 |
| LG20 | 3.42 |
| LG13 | 3.21 |
| LG16 | 3.07 |

In the phylogenetic tree, LG2 was closely related to the *Fictibacillus* group (Figure 4), suggesting that they have a common ancestor and share some features and functionality (Table 2, Figure 4). The genus *Fictibacillus* is Gram-positive, motile, rod-shaped, endospore-forming bacteria (Pal et al. 2018); however, studies on their cellulolytic activity are still limited. A study by Chen et al. (2020) showed that *Fictibacillus* sp. YS-26 isolated from soft corals in Yongxing Island, Hainan Province, China, have high cellulolytic activity and can degrade lignocellulose. The cellulolytic activity of *Fictibacillus* from mangrove sediments has never been reported. Therefore, the results of this study are considered as new information on the cellulolytic activity of *Fictibacillus* from mangroves.

Bacterial species that adapt to mangrove ecosystems are a potential source for biotechnology. Mangroves are a rich resource for discovering new bacterial and fungal species that produce enzymes and molecules to be used in human life, i.e., agriculture, industry, and bioremediation (Dias et al. 2009; Dourado et al. 2012). The ability to convert lignocellulosic substrates into simple nutrients is crucial for the carbon cycle and microbial survival, especially in high substrates environments. Lignocellulosic materials are very difficult to degrade due to their dense, compact structures, which protect plants against microbial attack (Kumar et al. 2008). Microorganisms that can degrade plant biomass might only be found in specific environments, such as mangrove ecosystems, where the biomass is abundant.

In conclusion, bacteria with cellulolytic activity were obtained from Logending mangrove sediments. Based on the phylogenic analysis, isolate LG2 is similar to the *Fictibacillus* group. This study provides new information on the cellulolytic activity of *Fictibacillus* from mangroves. It can produce enzymes and chemicals that can be beneficial for human life, such as agriculture, industry, and bioremediation.

**Table 2.** The similarity of LG2 isolate to other related strains using BLAST-N

| Description | Max score | Total score | Query cover | E value | Per. Ident | Accession |
|---|---|---|---|---|---|---|
| *Fictibacillus nanhaiensis* strain JSM 082006 16S ribosomal RNA, partial sequence | 2571 | 2571 | 99% | 0.0 | 99.86% | NR_117524.1 |
| *Fictibacillus halophilus* strain AS8 16S ribosomal RNA, partial sequence | 2567 | 2567 | 100% | 0.0 | 99.79% | NR_149289.1 |
| *Fictibacillus phosphorivorans* strain Ca7 16S ribosomal RNA, partial sequence | 2567 | 2567 | 99% | 0.0 | 99.86% | NR_118455.1 |
| *Fictibacillus barbaricus* strain V2-BIII-A2 16S ribosomal RNA, partial sequence | 2484 | 2484 | 99% | 0.0 | 98.85% | NR_028967.1 |
| *Fictibacillus aquaticus* strain GDSW-R2A3 16S ribosomal RNA, partial sequence | 2446 | 2446 | 100% | 0.0 | 98.21% | NR_159291.1 |
| *Fictibacillus arsenicus* strain Con a/3 16S ribosomal RNA, partial sequence | 2386 | 2386 | 100% | 0.0 | 97.43% | NR_042217.1 |
| *Fictibacillus rigui* strain WPCB074 16S ribosomal RNA, partial sequence | 2350 | 2350 | 100% | 0.0 | 97.00% | NR_116518.1 |
| *Fictibacillus enclensis* strain NIO-1003 16S ribosomal RNA, partial sequence | 2320 | 2320 | 99% | 0.0 | 96.64% | NR_133744.1 |
| *Fictibacillus solisalsi* strain YC1 16S ribosomal RNA, partial sequence | 2300 | 2300 | 99% | 0.0 | 96.43% | NR_044387.1 |
| *Bacillus tianmuensis* strain B5 16S ribosomal RNA, partial sequence | 2290 | 2290 | 100% | 0.0 | 96.22% | NR_116701.1 |

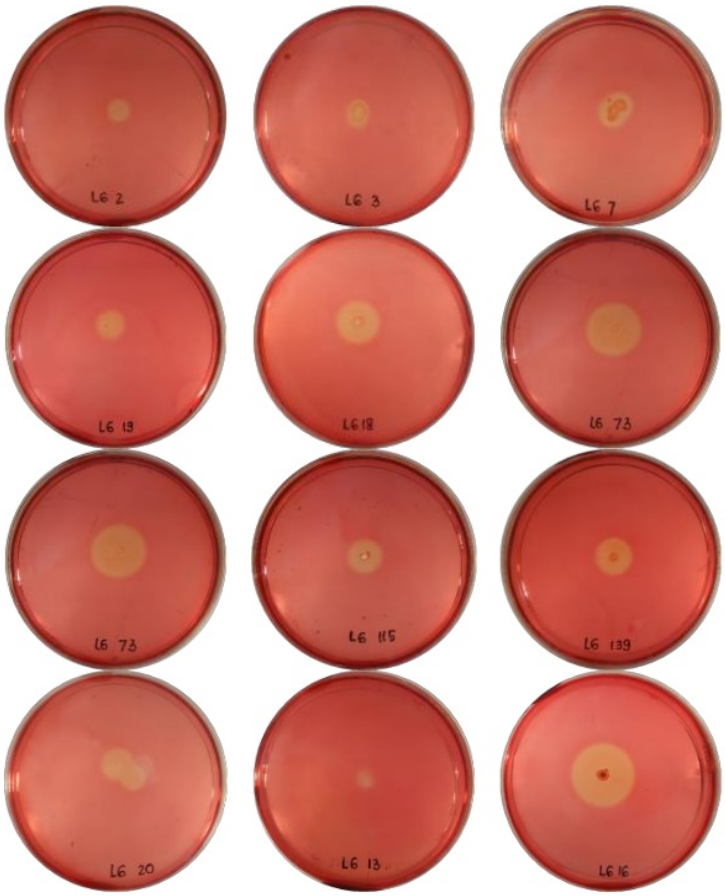


**Figure 3.** Cellulolytic activity of some bacterial isolates collected from Logending mangrove sediments

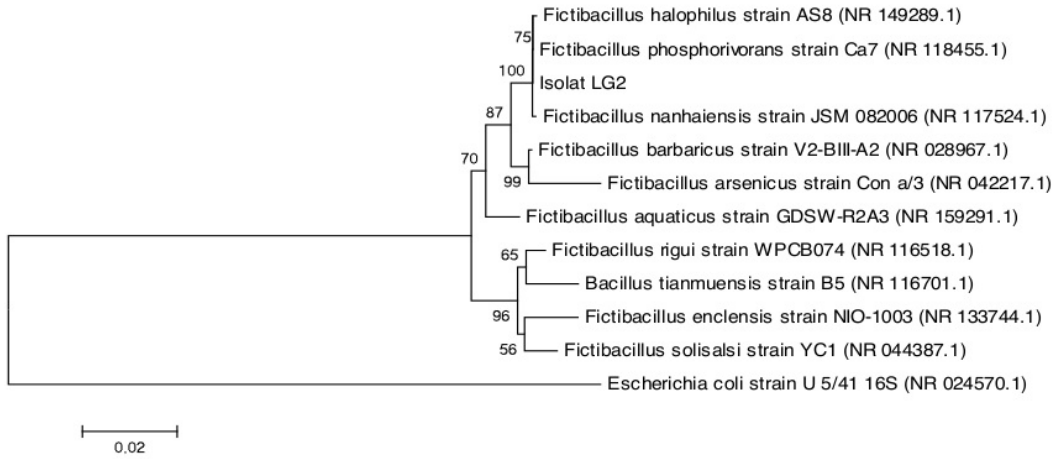


**Figure 4.** Phylogenetic tree of isolate LG2 and its closely related strains based on 16S rRNA gene sequence in the GenBank database

## ACKNOWLEDGEMENTS

We thank Soewarto, Rifda Naufalin, and Imam Widhiono for their support in obtaining research funding and to all of the students who helped collect field data. This research was supported by a Directorate General of Higher Education Research Fellowship and the Universitas Jenderal Soedirman, Purwokerto, Central Java, Indonesia. The authors declare that there are no conflicts of interest regarding the publication of this paper

## REFERENCES

Ambeng Zubair H, Oka NP, Tonggiroh A. 2019. Isolation and characterization of bacteria from mangrove sediment at coastal area in Pangkep South Sulawesi. J Phys: Conf Ser 1341(2): 022016. DOI: 10.1088/1742-6596/1341/2/022016.

Behera BC, Parida S, Dutta SK, Thatoi HN. 2014. Isolation and identification of cellulose degrading bacteria from mangrove soil of mahanadi river delta and their cellulase production ability. Am J Microbiol Res 2 (1): 41-46. DOI:10.12691/ajmr-2-1-6.

Basak P, Pramanik A, Sengupta S, Nag S, Bhattacharyya A, Pattanayak R, Ghosh A. Chattopadhyay D, Roy D, Bhattacharyya M. 2016. Bacterial diversity assessment of pristine mangrove microbial community from Dhulibhashani, Sundarbans using 16S rRNA gene tag sequencing. Genomic Data 7: 76-8. DOI: 10.1016/j.gdata.2015.11.030.

Bibi F, Ullah I, Akhtar S, Yasir M, Kensarah EA, AL-Ghamadi AAK, Azhar EI. 2017. Diversity and bioprospecting potential of rhizo and endophytic bacteria from two mangrove plants in Saudi Arabia. Afr J Microbiol Res 11 (19): 729-739. DOI: 10.5897/AJMR2017.8552.

Chantarasiri A. 2015. Aquatic *Bacillus cereus* JD0404 isolated from the muddy sediments of mangrove swamps in Thailand and characterization of its cellulolytic activity. Egypt J Aquat Res 41 (3): 257-264. DOI: 10.1016/j.ejar.2015.08.003.

Chen Y, Wang W, Zhou D, Jing T, Li K, Zhao Y, Tang W, Qi D, Zhang M, Zang X, Luo Y. 2020. Biodegradation of lignocellulosic agricultural residues by a newly isolated *Fictibacillus* sp. YS-26 improving carbon metabolic properties and functional diversity of the rhizosphere microbial community. Bioresour Technol. DOI: 10.1016/j.biortech.2020.123381.

Dias AC, Andreote FD, Dini-Andreote F, Lacava PT, Sá AL, Melo IS, Azevedo JL, Araújo WL. (2009). Diversity and biotechnological potential of culturable bacteria from Brazilian mangrove sediment. World J Microbiol Biotechnol 25 (7): 1305-1311. DOI: 10.1007/s11274-009-0013-7.

Dourado MN, Ferreira A, Araújo WL, Azevedo JL, Lacava PT. 2012. The diversity of endophytic methylotrophic bacteria in an oil-contaminated and an oil-free mangrove ecosystem and their tolerance to heavy metals. Biotechnol Res Intl. DOI:10.1155/2012/759865.

Glaeser SP, Dolt W, Busse HJ, Kampfer P. 2013. *Fictibacillus phosphorivorans* gen. nov., sp. nov. and proposal to reclassify *Bacillus arsenicus, Bacillus barbaricus, Bacillus macauensis, Bacillus nanhaiensis, Bacillus rigui, Bacillus solisalsi* and *Bacillus gelatini* in the genus *Fictibacillus*. Intl J Syst Evol Microbiol 63: 2934-2944. DOI 10.1099/ijs.0.049171-0.

Hastuti SU, Sangur K, Khasanah HN. 2015. Biodiversity and enzyme activity of indigenous cellulolytic and amylolytic bacteria in decayed mangrove stem waste product at Waai Seashore, Ambon Island. J Agric Sci Technol B 5: 252-256. DOI: 10.17265/2161-6264/2015.04.004.

Klijn NI, Weerkamp AH, de Vos WM. 1991. Identification of mesophilic lactic acid bacteria by using polymerase chain reaction-amplified variable regions of 16S rRNA and specific DNA probes. Appl Environ Microbiol 57 (11): 3390-3393.

Kumar R, Singh S, Singh OV. 2008. Bioconversion of lignocellulosic biomass: biochemical and molecular perspectives. J Ind Microbiol Biotechnol 35 (5): 377-391. DOI: 10.1007/s10295-008-0327-8.

Kurniawan A, Puspitasari S, Asriani E, Kurniawan A, Sambah AB, Prihanto AA. 2019. Molecular identification of cellulolytic bacteria from mangrove sediment at Tin Minning Region in West Bangka. Intl J Appl Biol 3 (1): 7-14. DOI: 10.20956/ijab.v3i1.5848.

Kurniawan A, Prihanto AA, Sari SP, Febriyanti D, Kurniawan A, Sambah AB, Asriani E. 2017. Isolation and Identification of cellulolytic bacteria from mangrove sediment in Bangka Island. In IOP Conf Ser: Earth Environ Sci 137 (1): 012070. DOI: 10.1088/1755-1315/137/1/012070

Liu M, Huiqin H, Bao S, Tong Y. 2019. Microbial community structure of soils in Bamenwan mangrove wetland. Sci Rep 9: 8406. DOI: 10.1038/s41598-019-44788-x.

Maharsiwi W, Astuti RI, Meryandini A, Wahyudi AT. 2020. Screening and characterization of sponge-associated bacteria from Seribu Island, Indonesia producing cellulase and laccase enzymes. Biodiversitas 21 (3): 975-981. DOI: 10.13057/biodiv/d210317.

Naresh S, Kunasundari B, Gunny AAN, Teoh YP, Shuit SH, Ng QH, Hoo PY. 2019. Isolation and partial characterisation of thermophilic cellulolytic bacteria from North Malaysian Tropical Mangrove Soil. Trop Life Sci Res 30 (1): 123-147. DOI: 10.21315/tlsr2019.30.1.8.

Nursyirwani N, Feliatra F, Tanjung A, Harjuni F. 2020. Isolation of cellulolytic bacteria from mangrove sediment in Dumai Marine Station Riau and the antibacterial activity against pathogens. IOP Conf Ser: Earth Environ Sci 430 (1): 012012. DOI: 10.1088/1755-1315/430/1/012012.

Pal D, Bhardwaj A, Kaur N, Krishnamurthi S, Sudan SK, Bisht B, Vyas B, Kumari M and Mayilraj S. 2018. *Fictibacillus aquaticus* sp. nov., isolated from downstream river water. Intl J Syst Evol Microbiol 68: 160-164. DOI: 10.1099/ijsem.0.002474.

Rajagopal G, Kannan S. 2017. Systematic characterization of potential cellulolytic marine Actinobacteria *Actinoalloteichus* sp. MHA15. Biotechnol Rep 13: 30-36. DOI: 10.1016/j.btre.2016.12.003.

Sakhia N, Sunil P, Shetty V, Bhatt S, Bhadalkal A, 2016. Study of Bacterial diversity of Mangrove Rhizosphere. Open J Mar Sci 6: 23-31. DOI: 10.4236/ojms.2016.61003.

Tamura T, Stecher G, Peterson D, Filipski A, Kumar S. 2013. MEGA6: Molecular evolutionary genetics analysis version 6.0. Mol Biol Evol 30 (12): 2725-2729. DOI: 10.1093/molbev/mst197.

Thompson CE, Beys-da-Silva WO, Santi L, Berger M, Vainstein MH, Guimarães JA, Vasconcelos ATR. 2013. A potential source for cellulolytic enzyme discovery and environmental aspects revealed through metagenomics of Brazilian mangroves. AMB Expr 3: 65. DOI: 10.1186/2191-0855-3-65.

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