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Home (<https://smujo.id/biodiv/index>)
 / Archives (<https://smujo.id/biodiv/issue/archive>)
 / Vol. 22 No. 2 (2021)



(<https://smujo.id/biodiv/issue/view/287>)

Vol. 22 No. 2 (2021)

Full Issue

Front Cover (<https://smujo.id/biodiv/issue/view/287/140>)

Articles

Freshwater pond microalgae for biofuel: Strain isolation, identification, cultivation and fatty acid content
 (<https://smujo.id/biodiv/article/view/5821>)

BAYU AFNOVANDRA PERDANA, ABDI DHARMA, INDRA JUNAIDI ZAKARIA, SYAFRIZAYANTI

PDF (<https://smujo.id/biodiv/article/view/5821/4542>)

Sustainability analysis of tomato jellyfish (Crambione mastigophora) fisheries resources management in Saleh Bay Waters, Sumbawa Island, Indonesia
 (<https://smujo.id/biodiv/article/view/4939>)

EVRON ASRIAL, MUHAMMAD MARZUKI, HAMID, RULY ISFATUL KHASANAH

PDF (<https://smujo.id/biodiv/article/view/4939/4543>)

Medicinal plants used by the indigenous Ati tribe in Tobias Fornier, Antique, Philippines
 (<https://smujo.id/biodiv/article/view/7286>)

CECILIA S. CORDERO, GRECEBIO JONATHAN D. ALEJANDRO

PDF (<https://smujo.id/biodiv/article/view/7286/4544>)

Drought adaptive prediction in potato (Solanum tuberosum) using in vitro and in vivo approaches
 (<https://smujo.id/biodiv/article/view/7139>)

JANE KATHRYNE JOLANDA LAISINA, AWANG MAHARIJAYA, SOBIR, AGUS PURWITO

PDF (<https://smujo.id/biodiv/article/view/7139/4545>)

Information

For Readers
 (<https://smujo.id/biodiv/information/readers>)

For Authors
 (<https://smujo.id/biodiv/information/authors>)

For Librarians
 (<https://smujo.id/biodiv/information/librarians>)

Journals List

Biodiversitas Journal of Biological Diversity
 (<https://smujo.id/biodiv>)

Nusantara Bioscience
 (<https://smujo.id/nb>)

Prosiding Seminar Nasional Masyarakat Biodiversitas Indonesia
 (<https://smujo.id/psnmbi>)

Asian Journal of Agriculture
 (<https://smujo.id/aja>)

Asian Journal of Ethnobiology
 (<https://smujo.id/aje>)

Asian Journal of Forestry
 (<https://smujo.id/ajf>)

Asian Journal of Natural Product Biochemistry
 (<https://smujo.id/jnpb>)

Asian Journal of Tropical Biotechnology
 (<https://smujo.id/bbs>)

International Journal of Bonorowo Wetlands
 (<https://smujo.id/bw>)

Cell Biology and Development
 (<https://smujo.id/cbd>)

Asia Pacific Journal of Ocean Life
 (<https://smujo.id/ol>)

International Journal of Tropical Drylands
 (<https://smujo.id/td>)

Reviewers List

Reviewers
 (<https://smujo.id/biodiv/reviewers/index>)

Visitor Statistics

Statistics
 (<https://smujo.id/info/stats>)

Classical and molecular cytogenetics of *Belontia hasselti* (Perciformes: Osphronemidae): Insights into the ZZ/ZW sex chromosome system (<https://smujo.id/biodiv/article/view/7110>)

PATCHARAPORN CHAIYASAN, BOONYADA MINGKWAN, SITTHISAK JANTARAT, CHATMONGKON SUWANNAPOOM, MARCELO DE BELLO CIOFFI, THOMAS LIEHR, SUCHEELA TALUMPHAI, ALONGKLOD TANOMTONG, WEERAYUTH SUPIWONG

PDF (<https://smujo.id/biodiv/article/view/7110/4546>)



(<https://info.flagcounter.com/JKAr>)

Antimicrobial susceptibility and molecular species identification of clinical carbapenem-resistant bacteria (<https://smujo.id/biodiv/article/view/7189>)

MAULIN INGGRAINI, SITI NURFAJRIAH, JEPRI AGUNG PRIYANTO, NOOR ANDRYAN ILSAN

PDF (<https://smujo.id/biodiv/article/view/7189/4547>)

Vegetative anatomy of three potted *Chrysanthemum* varieties under various paclobutrazol concentrations (<https://smujo.id/biodiv/article/view/7241>)

INTANI QUARTA LAILATY, LAURENTIUS HARTANTO NUGROHO

PDF (<https://smujo.id/biodiv/article/view/7241/4548>)

Short Communication: Presence of the vulnerable freshwater goby *Sicyopus auxilimentus* (Gobiidae, Sicydiinae) on Sangihe Island, Indonesia (<https://smujo.id/biodiv/article/view/7121>)

VERYL HASAN, FITRI SIL VALEN, R. ADHARYAN ISLAM, MAHENO SRI WIDODO, ADITYA MIRZAPAHLEVI SAPTADAJA, IZZUL ISLAM

PDF (<https://smujo.id/biodiv/article/view/7121/4549>)

Spatial and temporal overlaps of top predators: Dhole, tiger and leopard, and their potential preys in Huai Kha Khaeng Wildlife Sanctuary, Thailand (<https://smujo.id/biodiv/article/view/7340>)

KHWANRUTAI CHARASPET, RONGLARP SUKMASUANG, NORASET KHOEWSREE, MANANYA PLA-ARD, PAANWARIS PAANSRI, BOONYATIPORN KEAWDEE, YUWALUK CHANACHAI, NARIS BHUMPAKPHAN

PDF (<https://smujo.id/biodiv/article/view/7340/4550>)

Genetic diversity of indigenous catfish from Indonesia based on mitochondrial Cytochrome Oxidase Subunit II gene (<https://smujo.id/biodiv/article/view/7400>)

VISTA BUDIARIATI, TRINI SUSMIATI, SITI MUNAWAROH, RACHMAWATI CAHYANINGTYAS ARIE PUTRI, RINI WIDAYANTI

PDF (<https://smujo.id/biodiv/article/view/7400/4551>)

Phytochemicals and antimicrobial analysis of selected medicinal plants from Brunei Darussalam (<https://smujo.id/biodiv/article/view/7450>)

HUSSEIN TAHA, ZULHAMIZAN AWANG-JAMIL, MUHAMMAD FAIRUZ AMINUDDIN, AIDA MARYAM BASRI, BARIRAH QURATULAIN ZAIDI, NORHAYATI AHMAD

PDF (<https://smujo.id/biodiv/article/view/7450/4552>)

Morphotype diversity of *Prorocentrum* lima in the western part of Indonesian waters
(<https://smujo.id/biodiv/article/view/7195>)

RIANI WIDIARTI, NEVIATY PUTRI ZAMANI, DIETRICH GEOFFREY BENGEN, HAWIS MADDUPPA

PDF (<https://smujo.id/biodiv/article/view/7195/4553>)

Genetic Analysis and Pathogenic Characterization of *Alternaria tenuissima* Induced Fruit Rot of Bitter Gourd
(<https://smujo.id/biodiv/article/view/7262>)

SEHRISH IFTIKHAR, WAHEED ANWAR, ADNAN AKHTER, SAJID ALI, HAFIZ AZHAR ALI KHAN, MUHAMMAD KHURSHID, MUHAMMAD SALEEM HAIDER

PDF (<https://smujo.id/biodiv/article/view/7262/4554>)

Stock status of coconut crab (*Birgus latro*) in Daeo, Morotai Island District, North Maluku, Indonesia
(<https://smujo.id/biodiv/article/view/7479>)

RUGAYA H. SEROSERO, SULISTIONO, NURLISA A. BUTET, ETTY RIANI

PDF (<https://smujo.id/biodiv/article/view/7479/4555>)

Vegetation structure of Sumatran Orangutan (*Pongo abelii*) habitat in North Sumatra, Indonesia
(<https://smujo.id/biodiv/article/view/6670>)

ANITA ZAITUNAH, SAMSURI, SATIA RAS

PDF (<https://smujo.id/biodiv/article/view/6670/4556>)

The antagonistic activity of marine actinomycetes from mangrove ecosystem against phytopathogenic fungi *Colletotrichum* sp. KA
(<https://smujo.id/biodiv/article/view/7162>)

QONITA GINA FADHILAH, IMAN SANTOSO, YASMAN

PDF (<https://smujo.id/biodiv/article/view/7162/4557>)

The role of coastal biodiversity conservation on sustainability and environmental awareness in mangrove ecosystem of southern Malang, Indonesia
(<https://smujo.id/biodiv/article/view/7272>)

ZAINAL ABIDIN, BUDI SETIAWAN, ABDUL WAHIB MUHAMMIN, AGUSTINA SHINTA

PDF (<https://smujo.id/biodiv/article/view/7272/4558>)

Ethnoecological study on the utilization of plants in Ciletuh-Palabuhanratu Geopark, Sukabumi, West Java, Indonesia
(<https://smujo.id/biodiv/article/view/7484>)

INDRI WULANDARI, BUDIAWATI SUPANGKAT ISKANDAR, PARIKESIT, TEGUH HUDOSO, JOHAN ISKANDAR, ERRI NOVIAR MEGANTARA, ELMA FAUZIAH GUNAWAN, SYA SYA SHANIDA

PDF (<https://smujo.id/biodiv/article/view/7484/4559>)

Genetic differentiation of dengue vector *Aedes aegypti* in the small geographical scale of Banyumas District, Indonesia based on

Cytochrome Oxidase I
(<https://smujo.id/biodiv/article/view/5543>)

MOHAMMED A. MOHAMMED, AGUS NURYANTO,
ENDANG SRIMURNI KUSMINTARSIH

PDF (<https://smujo.id/biodiv/article/view/5543/4560>)

The characteristics of fermented purple sweet potato (*Ipomoea batatas*) and black rice (*Oryza sativa*) using UV-irradiated *Monascus purpureus* (<https://smujo.id/biodiv/article/view/7309>)

ISWIYANTI NOVITA, OEDJIJONO, ARI ASNANI

PDF (<https://smujo.id/biodiv/article/view/7309/4561>)

Assessing the conservation value of medicinal plant collections in Bogor Botanic Gardens, Indonesia

(<https://smujo.id/biodiv/article/view/7485>)

SYAMSUL HIDAYAT, ERVIZAL A.M. ZUHUD, DIDIK
WIDYATMOKO, BAHRUNI

PDF (<https://smujo.id/biodiv/article/view/7485/4562>)

The role of bacterial symbionts in the biodegradation of chlorpyrifos in the digestive tract of *Plutella xylostella* larvae

(<https://smujo.id/biodiv/article/view/6942>)

MOHAMMAD SYAMSUL HADI, ABDUL LATIEF ABADI,
TOTO HIMAWAN, MASRURI, SAFIRA RIZKA LESTARI,
BAMBANG TRI RAHARDJO, LUQMAN QURATA AINI,
YOGO SETIAWAN, HAGUS TARNO

PDF (<https://smujo.id/biodiv/article/view/6942/4563>)

Genetic structuring of *Ephedra* an important medicinal plant from low to high altitudinal zones of Balochistan, Pakistan

(<https://smujo.id/biodiv/article/view/7154>)

ABDUL BASIT, SHAZIA SAEED, ALIA AHMED, NIZAM
BALOCH, ASIF ZAHID ALI

PDF (<https://smujo.id/biodiv/article/view/7154/4564>)

The use of alternative liquid media for propagation of pathogenic fungi and their effect on weeds

(<https://smujo.id/biodiv/article/view/7220>)

LOEKAS SOESANTO, ENDANG MUGIASTUTI, ABDUL
MANAN

PDF (<https://smujo.id/biodiv/article/view/7220/4565>)

Short communication: Savanna-forest boundary on Mount Rinjani, Lombok Island, West Nusa Tenggara, Indonesia

(<https://smujo.id/biodiv/article/view/5979>)

SUTOMO, EDDIE VAN ETEN, RAJIF IRYADI

PDF (<https://smujo.id/biodiv/article/view/5979/4566>)

Bioactivity of carotenoid produced by soft coral symbiotic microorganisms from Panjang and Karimunjawa Island, Central Java, Indonesia (<https://smujo.id/biodiv/article/view/7151>)

LIA KUSMITA, HANDUNG NURYADI, PRASTYO ABI
WIDYANANTO, SAKTI MUCHLISSIN, AGUS SABDONO,
AGUS TRIANTO, OCKY KARNA RADJASA

PDF (<https://smujo.id/biodiv/article/view/7151/4567>)

Sequence and expression analysis of glucokinase mRNA from herbivorous Giant gourami (*Osphronemus goramy*) (<https://smujo.id/biodiv/article/view/7266>)

DIAN NOVITA SARI, HASAN NASRULLAH, JULIE EKASARI, MUHAMMAD AGUS SUPRAYUDI, ALIMUDDIN ALIMUDDIN

PDF (<https://smujo.id/biodiv/article/view/7266/4568>)

The allometric relationships for estimating above-ground biomass and carbon stock in an abandoned traditional garden in East Kalimantan, Indonesia (<https://smujo.id/biodiv/article/view/7253>)

KARYATI, KUSNO YULI WIDIATI, KARMINI, RACHMAD MULYADI

PDF (<https://smujo.id/biodiv/article/view/7253/4569>)

Species diversity and abundance of scorpions in Ahvaz city, Southwest Iran (<https://smujo.id/biodiv/article/view/7289>)

NAHID JALILI SHAH MANSOURI, KAMRAN AKBARZADEH, ELHAM JAHANIFARD, BABAK VAZIRIANZADEH, JAVAD RAFINEJAD

PDF (<https://smujo.id/biodiv/article/view/7289/4570>)

Molecular characterization of a farmer-preferred maize landrace population from a multiple-stress-prone subtropical lowland environment (<https://smujo.id/biodiv/article/view/7374>)

FORTUNATE MAKORE, EDMORE GASURA, CALEB SOUTA, UPENYU MAZARURA, JOHN DERERA, MELULEKI ZIKHALI, CASPER N. KAMUTANDO, COSMOS MAGOROKOSHO, SHORAI DARI

PDF (<https://smujo.id/biodiv/article/view/7374/4571>)

Identification of fermentative bacteria on local microorganisms of golden snail (*Pomacea canaliculata* Lamarck, 1822) (<https://smujo.id/biodiv/article/view/6881>)

YULIANA RETNOWATI, ABUBAKAR SIDIK KATILI

PDF (<https://smujo.id/biodiv/article/view/6881/4572>)

Identification of Drought Tolerant and Molecular Analysis of DREB2A and BADH2 Genes and Yield Potensial of Lines from Single Crossing Bengkulu Local Rice Varieties (<https://smujo.id/biodiv/article/view/6884>)

RENY HERAWATI, ALNOPRI, MASDAR, MARULAK SIMARMATA, SIPRIYADI, MIMI SUTRAWATI

PDF (<https://smujo.id/biodiv/article/view/6884/4573>)

First genetic record and the phylogenetic relationship of *Osteochilus spilurus* (Cyprinidae: Labeoninae) originating from Bangka and Belitung Islands, Indonesia (<https://smujo.id/biodiv/article/view/7285>)

ARDIANSYAH KURNIAWAN, ANIK M. HARIATI, ANDI KURNIAWAN, DEWA G.R. WIADNYA

PDF (<https://smujo.id/biodiv/article/view/7285/4574>)

Mangrove diversity and suitability assessments for ecotourism in Cimalaya Wetan Coast, Karawang District, Indonesia

(<https://smujo.id/biodiv/article/view/7438>)

TJIONG GIOK PIN, JATNA SUPRIATNA, NOVERITA DIAN
TAKARINA, RUDY PARLUHUTAN TAMBUNAN

PDF (<https://smujo.id/biodiv/article/view/7438/4575>)

Morphological and genetic variation in
populations of *Desmos chinensis* Lour.
(Annonaceae)

(<https://smujo.id/biodiv/article/view/7618>)

ISNA AROFATUN NIKMAH, RUGAYAH RUGAYAH, TATIK
CHIKMAWATI

PDF (<https://smujo.id/biodiv/article/view/7618/4576>)

Diversity and genetic parameter of chili pepper
(*Capsicum annuum*) based on yield component
in three location

(<https://smujo.id/biodiv/article/view/7386>)

TRI WAHONO DYAH AYU SAYEKTI, MUHAMAD SYUKUR,
SRI HENDRASTUTI HIDAYAT, AWANG MAHARIJAYA

PDF (<https://smujo.id/biodiv/article/view/7386/4578>)

Distribution of polyprenol and dolichol in oil
palm genotype (*Elaeis guineensis*) involving
lipase activity

(<https://smujo.id/biodiv/article/view/7415>)

TENGGU SITI HABSYAH, MOHAMMAD BASYUNI, LUTHFI
A.M. SIREGAR, DADANG AFANDI, INDRA SYAHPUTRA

PDF (<https://smujo.id/biodiv/article/view/7415/4579>)

Phytoremediation of iron in ex-sand mining
waters by water hyacinth (*Eichhornia crassipes*)
(<https://smujo.id/biodiv/article/view/7476>)

QADAR HASANI, NIKEN T.M. PRATIWI, YUSLI
WARDIATNO, HEFNI EFFENDI, ARTHO NUGRAHA
MARTIN, PURNA PIRDAUS, WAGIRAN

PDF (<https://smujo.id/biodiv/article/view/7476/4580>)

Length-weight relationship, sex ratio, mortality
and growth condition of natural stock of
Macrobrachium rosenbergii from the estuarine
systems of North Kalimantan, Indonesia
(<https://smujo.id/biodiv/article/view/7331>)

AGUS INDARJO, GAZALI SALIM, CHRISTINE DYTA
NUGRAENI, MUFRIDA ZEIN, JULIAN RANSANGAN,
LUKMAN YUDHO PRAKOSO, SUHIRWAN, SUTRISNO
ANGGORO

PDF (<https://smujo.id/biodiv/article/view/7331/4581>)

Semi-natural regeneration and conservation in
agroforestry system models on small-scale
farmers

(<https://smujo.id/biodiv/article/view/7403>)

PRIYONO SURYANTO, RONGGO SADONO, AISYA
YOHANIFA, MUHAMMAD HABIB WIDYAWAN, TAUFAN
ALAM

PDF (<https://smujo.id/biodiv/article/view/7403/4582>)

Isolation and identification of phytate-degrading
yeast from traditional fermented food
(<https://smujo.id/biodiv/article/view/7371>)

ADE ERMA SURYANI, AYU SEPTI ANGGRAENI, LUSTY
ISTIQOMAH, EMA DAMAYANTI, MOHAMMAD FAIZ
KARIMY

PDF (<https://smujo.id/biodiv/article/view/7371/4583>)

Short Communication: Histopathology of red chilli fruit (*Capsicum annum*) infected with *Colletotrichum acutatum* of Java, Indonesia isolates

(<https://smujo.id/biodiv/article/view/7280>)

JUNI SAFITRI MULJOWATI, LOEKAS SOESANTO, LAURENTIUS HARTANTO NUGROHO

PDF (<https://smujo.id/biodiv/article/view/7280/4584>)

Distribution of multidrug-resistant *Salmonella* spp. recovered from aquatic environment of Banda Aceh, Indonesia

(<https://smujo.id/biodiv/article/view/7358>)

SUHARTONO SUHARTONO, YULIA SARI ISMAIL, ZAH RATUL AINI

PDF (<https://smujo.id/biodiv/article/view/7358/4585>)

Diversity and distribution of epiphytic lichens on *Cedrus atlantica* and *Quercus faginea* in Mount Babor Forest, Algeria

(<https://smujo.id/biodiv/article/view/7697>)

AMINA BELGUIDOUM, TAKIA LOGRADA, MESSAOUD RAMDANI

PDF (<https://smujo.id/biodiv/article/view/7697/4586>)

DNA intensity and genetic diversity of oil palm (*Elaeis guineensis*) to determine an elite low lipase line

(<https://smujo.id/biodiv/article/view/6897>)

NINA UNZILA ANGKAT, LUTHFI AZIZ MAHMUD SIREGAR, MOHAMMAD BASYUNI, DADANG AFANDI, INDRA SYAHPUTRA

PDF (<https://smujo.id/biodiv/article/view/6897/4587>)

Composition and diversity of bacteria from giant Asian honeybee *Apis dorsata* gut

(<https://smujo.id/biodiv/article/view/7077>)

NURDJANNAH JANE NIODE, ARYANI ADJI, JIMMY RIMBING, MAX TULUNG, TRINA EKAWATI TALLEI

PDF (<https://smujo.id/biodiv/article/view/7077/4588>)

Phytochemical screening and cytotoxic evaluation of *Bauhinia scandens* leaf extracts using HeLa and T47D cell lines

(<https://smujo.id/biodiv/article/view/6954>)

LIANAH LIANAH, RITA ARIYANA NUR KHASANAH, DWIMEI AYUDEWANDARI PRANATAMI, KRISANTINI KRISANTINI

PDF (<https://smujo.id/biodiv/article/view/6954/4589>)

Correlation between landscape structure and distribution of Javan Pangolin (*Manis javanica*) in an extreme landscape

(<https://smujo.id/biodiv/article/view/7307>)

SUSANTI WITHANINGSIH, PARIKESIT, ANWAR NASRUDIN

PDF (<https://smujo.id/biodiv/article/view/7307/4590>)

Chemical composition and antimicrobial activity of *Myrtus communis* essential oils from Algeria

(<https://smujo.id/biodiv/article/view/7327>)

YACINE MOHAMADI, TAKIA LOGRADA, MESSAOUD
RAMDANI, GILLES FIGUEREDO, PIERRE CHALARD

PDF (<https://smujo.id/biodiv/article/view/7327/4591>)

A pictorial key for the identification of beetle
(Order: Coleoptera) and diversity study in
selected area within Pelangai Forest Reserve,
Negeri Sembilan, Malaysia
(<https://smujo.id/biodiv/article/view/7072>)

NURUL HUDDA ABDULLAH, IZZATI ADILAH AZMIR

PDF (<https://smujo.id/biodiv/article/view/7072/4592>)

Baseline susceptibility of Philippine *Ostrinia
furnacalis* (Lepidoptera: Crambidae) populations
to insecticidal Cry1A.105 and Cry2Ab2 proteins
and validation of candidate diagnostic
concentration for monitoring resistance
(<https://smujo.id/biodiv/article/view/7087>)

EDWIN P. ALCANTARA, MARNELLE M. ATIENZA, LUIS
CAMACHO, SRINIVAS PARIMI

PDF (<https://smujo.id/biodiv/article/view/7087/4593>)

Fishing ground and tuna productivity by tuna
longline based on Benoa Bay, Bali, Indonesia
(<https://smujo.id/biodiv/article/view/7228>)

MOHAMMAD IMRON, MUHAMMAD IRSYAD TAWAQAL,
ROZA YUSFIANDAYANI

PDF (<https://smujo.id/biodiv/article/view/7228/4594>)

Maternal effect of agronomic and morphological
characters on cluster structure of F3 soybean
lines (<https://smujo.id/biodiv/article/view/7510>)

PANJI HANDOKO BADIARAJA, SITI ZUBAIDAH, HERU
KUSWANTORO

PDF (<https://smujo.id/biodiv/article/view/7510/4595>)

Short communication: Environment and
morphometric of sea hare *Dolabella auricularia*
from shrimp pond, Sorong, West Papua,
Indonesia
(<https://smujo.id/biodiv/article/view/7035>)

ACHMAD SOFIAN, ACHMAD SUHERMANTO, SAIDIN,
MOHAMMAD SAYUTI, DIAN NOVIANTO, FERLIANA
WIDYASARI

PDF (<https://smujo.id/biodiv/article/view/7035/4596>)

Seagrass-associated fish species' richness:
evidence to support conservation along the
south coast of Lombok Island, Indonesia
(<https://smujo.id/biodiv/article/view/7061>)

ABDUL SYUKUR, AGIL AL-IDRUS, LALU ZULKIFLI

PDF (<https://smujo.id/biodiv/article/view/7061/4597>)

Determinants of Nile tilapia's (*Oreochromis
niloticus*) growth in aquaculture pond in Batu,
Indonesia
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The characteristics of fermented purple sweet potato (*Ipomoea batatas*) and black rice (*Oryza sativa*) using UV-irradiated *Monascus purpureus*

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Abstract. Novita I, Oedjijono, Asnani A. 2021. The characteristics of fermented purple sweet potato (*Ipomoea batatas*) and black rice (*Oryza sativa*) using UV-irradiated *Monascus purpureus*. *Biodiversitas* 22: 684-690. This research aimed to produce *Monascus* fermented product (MFP) with purple sweet potato (*Ipomoea batatas* (L.) Lam.) and black rice (*Oryza sativa* L.) using UV-irradiated *Monascus purpureus* Went and evaluated the characteristic of its antibacterial activity against *Salmonella typhi*. *M. purpureus* was irradiated with UV at λ_{254} nm for 0, 2, 3, and 4 min. The solid-state fermentation process was carried out for 7, 14, and 21 days. The pigments were measured at λ_{390} nm for yellow and λ_{500} nm for red. The ethanol extracts of MFP were analyzed for their antibacterial activity against *S. typhi* using the Kirby–Bauer method. The results showed that the highest yield of MFP was obtained from MFP–black rice (51.88%) that used UV-irradiated *M. purpureus* for 2 min and fermentation for 21 days. The highest absorbance value of MFP–purple sweet potato was obtained from UV-irradiated *M. purpureus* for 3 min, whereas the highest absorbance value of MFP–black rice was obtained with UV-irradiated *M. purpureus* for 2 min. Ethanol extracts of both MFP3–purple sweet potato and MFP2–black rice showed antibacterial activities against *S. typhi* with minimum inhibitory concentration values of 0.2 and 0.15 g/mL, respectively. Thin-layer chromatography analysis of the ethanol extract from MFP2–black rice revealed the presence of bioactive saponin and flavonoid. These findings suggest that UV-irradiated *M. purpureus* was able to use both purple sweet potato and black rice substrates to produce MFP with antibacterial activity against *S. typhi*.

Keywords: Black rice, fermentation, *Monascus*, purple sweet potato, UV-irradiated

INTRODUCTION

Monascus purpureus Went is a filamentous fungus with known potential for natural pigment production through submerged or solid-state fermentation (Christiana 2016). During fermentation, *M. purpureus* will utilize starchy substrates and produce secondary metabolites in the form of pigments. Based on the color, *Monascus* pigments are classified into three categories, including red (monascorubramin and rubropunctamin), yellow (monascin and ankaflavin), and orange (monascorubrin and rubropunctatin) (Kim and Ku 2018). These pigments are widely used in the Asian region as a natural food colorant and as health products because of their bioactivities (Srianta et al. 2014).

Production of *Monascus* fermented product (MFP) is affected by the type of nutrients as well as physicochemical conditions such as pH, temperature, moisture content, dissolved oxygen concentration, and light. Bühler et al. (2015) observed that light intensity is an essential factor in the mycelium growth and pigment production of *Monascus*. Indeed, monacolin K produced by ultraviolet (UV)-irradiated *Monascus* was three times greater than the control culture (Sun et al. 2011). Monacolin K, commercially known as lovastatin, is a potent competitive

inhibitor of 3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase, inhibits cholesterol biosynthesis, and lowers blood cholesterol levels in both humans and animals. UV-mutated *M. purpureus* TISTR 3179 was used to produce a yellow pigment at a single λ_{max} 370 nm (Yongsmith et al. 2013). Huang et al. (2019) mutated *Monascus* spores using UV irradiation and ethyl methane sulfonate (EMS) for higher monacolin K and red pigment productions. The importance of light intensity in the fermentation process indicates that *Monascus* probably has photoreceptors that influence the physiological responses.

Rice is mainly used as a substrate for centuries (Pattanagul et al. 2007), but alternative substrates have been extensively explored. Yongsmith et al. (2013) used rice, corn, mung bean, soybean, potato, sweet potato, and cassava tubers as substrates to produce exclusively yellow pigments. Bühler et al. (2015) reported the utilization of pear juice, jackfruit seeds, ethanol, fructose, maltose, sucrose, lactose, corn syrup, grape waste, cornflour, cassava starch, and glycerol as alternative substrates for pigment production of *Monascus* sp. *Monascus* FJ46 produced MFP with a lower yield of citrinin using various carbon sources, including cereals, tuber crops, and agro-industrial residues, compared with rice flour as control (Mu et al. 2015). Citrinin is a mycotoxin produced by *Monascus*

species, which has been a common problem in MFP production. Srianta et al. (2017) reported the use of rice, corn, and sorghum to produce MFP that has antioxidant activities. So, far, purple sweet potato and black rice have not been used as substrates for MFP. Thus, in this research, we proposed purple sweet potato (*Ipomoea batatas* (L.) Lam.) and black rice (*Oryza sativa* L.) as two new alternative substrates for MFP.

Purple sweet potato and black rice are considered functional foods because they are rich in anthocyanin, which also serves as their natural pigment (Ginting et al. 2015; Sompong et al. 2011). Anthocyanin is reported to have various biological activities, including strong antioxidant, anti-inflammatory, and antimicrobial properties (Khoo et al. 2017; Esatbeyoglu et al. 2017; Zhi et al. 2020). Black rice also contains active phytochemicals such as tocopherol, tocotrienols, oryzanols, vitamin B complex, and phenolic compounds (Jang et al. 2012). It also has phenolic-active ingredients four times higher than white rice (Thanuja and Parimalavalli 2020). Hence, the bioactive components in purple sweet potato and black rice make them potential substrates to increase the nutraceutical uses of MFP.

The application of MFP as a coloring additive provides additional advantages such as preservatives and food supplements (Srianta et al. 2014). The pigments produced by *Monascus* are used as food preservatives because of their antimicrobial activities. The pigments have been reported to inhibit the growth of fungi such as *Aspergillus*, *Trichoderma*, *Mucor*, *Penicillium*, and *Fusarium* and bacteria such as *Bacillus*, *Pseudomonas*, *Escherichia*, and *Streptomyces* (Ungureanu and Ferdes 2010). Rojsuntornkitti et al. (2010) noted that *Monascus* red pigment is used as a substitute for nitrite inhibited *Salmonella* spp., *Clostridium perfringens*, and *Staphylococcus aureus* in Thai sausage. *Salmonella* causes foodborne diseases because of food and water contamination. Until now, typhoid fever caused by *Salmonella typhi* is a global burden with 11.0–17.8 million typhoid fever illnesses occurring annually worldwide (Crump 2019). Recently, *S. typhi* resistances include antibiotics ampicillin, trimethoprim-sulfamethoxazole (TMP-SMX), ciprofloxacin, and ceftriaxone (Wong et al. 2019). The increasing threat of antimicrobial resistance is the reason for the search for alternative antibacterials for *S. typhi* from natural sources. Therefore, the objectives of this research were evaluating the use of purple sweet potato and black rice as new substrates for MFP production using UV-irradiated *M. purpureus* and analyzing their antibacterial activity against *S. typhi*.

MATERIALS AND METHODS

Procedures

Substrate preparation

Purple sweet potato (*Ipomoea batatas* L.) and organic black rice (*Oryza sativa* L.) Hotel® were purchased from a local market, that is, Pasar Wage, Purwokerto. Individually, purple sweet potato and black rice were prepared as

described by Ginting et al. (2015), with modification. The tubers of purple sweet potatoes were thinly sliced, dried at 60°C for 32 h, ground, and then sieved with a 60 mesh to obtain a fine powder of purple sweet potato. Similarly, the black rice was thoroughly washed, dried at 60°C for 12 h, ground, and then sieved with a 60 mesh to obtain a fine powder of black rice.

Evaluation of *Monascus* growth pattern

Monascus purpureus was obtained from the Indonesian Culture Collection (InaCC) LIPI Bogor. *M. purpureus* was cultivated in potato dextrose agar (PDA) and incubated at 30°C for 7 days before being used. The growth pattern of *M. purpureus* for each substrate was evaluated based on mycelial growth. The medium used was modified from Permana et al. (2004). It consisted of fine powder of purple sweet potato or black rice (5%), KH₂PO₄ (0.25%), NaNO₃ (0.15%), MgSO₄·7H₂O (0.1%), monosodium glutamate (0.1%), CaCl₂ (0.001%), and distilled water up to 100 mL. The pH was adjusted to 6.0, and the medium was sterilized at 121°C for 15 min. After cooling, the medium was inoculated with *M. purpureus* and incubated at 30°C. The mycelial growth was measured as dry weight every day for 7 days. The incubation time with the highest mycelial weight was used for inoculum preparation.

UV irradiation and inoculum preparation

M. purpureus was inoculated with PDA in a Petri dish. Aseptically, the Petri disk cover was opened, and the culture was irradiated with ultraviolet light at λ254 nm with exposure times of 0, 2, 3, and 4 min. The control was *M. purpureus* without UV irradiation (0 min). All irradiated *M. purpureus* samples were then incubated at 30°C for 7 days. The sterilized medium, as described earlier, was inoculated with irradiated *M. purpureus* and incubated at 30°C for 4 days to produce an inoculum.

Solid-state fermentation

Solid-state fermentation was performed using Completely Randomized Factorial Design with three factorials. The first factor was two types of substrate (S1 = purple sweet potato and S2 = black rice), the second factor was four lengths of UV irradiation at λ254 nm (R0 = 0 min, R1 = 2 min, R2 = 3 min, and R3 = 4 min), and the third factor was three incubation times (T1 = 7 days, T2 = 14 days, and T3 = 21 days). The choice of incubation times followed Yongsmith et al. (2013) with modification. Each treatment was repeated two times, so there were 48 experimental units. The parameters measured were the yield of the *Monascus* fermented products (MFPs) and the absorbance of the ethanol extracts of MFP at λ390 and λ500 nm for pigment evaluation.

100 g of purple sweet potato (S1) or black rice powder (S2) was sterilized at 121°C for 15 min. After cooling, each substrate was inoculated with 2% inoculum. The mixture was stirred aseptically and then closed tightly with sterilized parchment paper. The mixture was incubated at 30°C with three incubation times (T1, T2, and T3). After each incubation time, all MFPs were dried in an oven at

60°C to yield irradiated MFP types, which were MFP–purple sweet potato and MFP–black rice.

0.05 g of MFP products were extracted with 10 mL of 95% ethanol. The mixture was centrifuged, and the filtrate obtained was measured with UV–Vis spectrophotometry at λ 390 nm for the yellow pigment and λ 500 nm for the red pigment (Kasim et al. 2005). The MFP product with the highest absorbance was evaluated for antibacterial activity against *Salmonella typhi*.

Evaluation of antibacterial activity

The MFP product with the highest absorbance was extracted with 95% ethanol with a ratio of 1:5 (w/v) by maceration. After 3×24 h of macerations, each mixture was filtered, and the filtrate obtained was evaporated to give the ethanol extract. The antibacterial activity of the ethanol extract against *Salmonella typhi* was evaluated by disk diffusion or Kirby–Bauer method (Hudzicki 2009). *S. typhi* was from the Microbiology Laboratory, Faculty of Biology, Universitas Jenderal Soedirman, Purwokerto. *S. typhi* on nutrient agar slant was serially diluted with sterile 0.9% NaCl into 10^{-5} dilution. 100 μ L of 10^{-5} suspension of *S. typhi* was uniformly spread over the NA medium and incubated 37°C overnight. Paper disks (ϕ 6 mm, thickness of 0.5 mm) were impregnated with 15 μ L of four concentrations (0.05, 0.10, 0.15, and 0.20 g/mL) of ethanol extracts. Then, each disk was put on NA plates inoculated with *S. typhi* and incubated at 37°C for 24 h. The positive control was 5% (w/v) of chloramphenicol, and the negative one was a sterile aqueous solution. The inhibition zone of each extract was calculated based on the diameter inhibition zone minus the diameter of the paper disk used.

Analysis of ethanol extract

The ethanol extract with the highest antibacterial activity was analyzed using thin-layer chromatography (TLC), followed by bioautography. The ethanol extract was spotted on the TLC sheet (Silica gel 60–F254 nm) and eluted with n-butanol: acetic acid: water (5:1:4). After elution, each TLC was sprayed with the following reagents: 1% ethanolic solution of AlCl_3 for flavonoid, *p*-anisaldehyde–sulfuric acid for terpenoid, 1% FeCl_3 for tannin, Dragendorff reagent for alkaloid, and Liebermann–Burchard reagent for saponins. All TLCs were observed under UV light at λ 365 nm for color changes, and its R_f value was calculated (Waldi 1965). Each separated spot from TLC was carefully scraped with a sterile spatula and then impregnated directly on the NA inoculated with *S. typhi* for bioautography analysis. The plates were incubated at 37°C for 24 h. The inhibition zone observed was calculated and compared to the TLC results (Nostro et al. 2000).

Data analysis

The data from solid-state fermentation were analyzed using analysis of variance. The results that showed significant diversity were further analyzed using Duncan's multiple range test (DMRT) with a 95% confidence level ($\alpha = 0.05$). The lowest concentration of ethanol extracts capable of inhibiting *S. typhi* was considered the minimum inhibitory concentration (MIC).

RESULTS AND DISCUSSION

The morphology of *M. purpureus*

The morphology development of *M. purpureus* cultivated on PDA was observed for 7 days at 27°C–32°C. The culture rapidly grew and spread on the surface of the media. The colony has a diameter of 20–30 mm, the texture was wooly, and the surface was *flocculent superficial*. In the early stage, the color of the mycelium was white. It rapidly turned into pink, subsequently into yellow-orange, and finally became crimson red as the colony ages (Figure 1). The red pigment was observed both in the mycelium and diffused pigment in the media.

Monascus purpureus showed different growth characteristics because of UV irradiation at λ 254 nm. The colony changed in size, shape, and color of the mycelium. The colony's size was reduced to 2–6 mm in diameter, the shape became round, and the color of the mycelium was white to reddish-orange (Figure 2). The growth of *M. purpureus* after 2 min of UV irradiation was better compared to others.

Analysis of growth pattern

The growth of *M. purpureus* in liquid medium was evaluated as the increment in mycelium weight (g) during incubation time (d). The research results showed that the growth curve of *M. purpureus* on both purple sweet potato and black rice substrates followed the growth pattern of fungi in general. *M. purpureus* grew quickly up to the fourth day of cultivation and slowed down after that. The highest mycelium weight (1.86 g) using purple sweet potato substrate was achieved on the fourth day, and the black rice substrate has the highest mycelium weight (2.32 g) after the fourth day of incubation. Thus, the inoculum of *M. purpureus* was prepared by the cultivation of spores in a liquid medium for 4 days. Mycelium weight clearly indicated that the growth of *M. purpureus* was better in black rice than in purple sweet potato as a substrate (Figure 3). Besides, purple sweet potato formed sticky clumps, which might hamper the dispersal of oxygen and nutrients for the effective growth of *M. purpureus*.

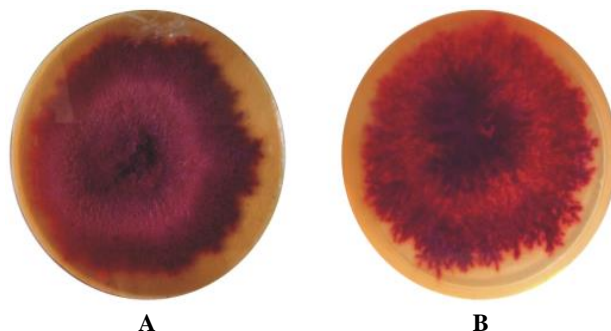


Figure 1. The colony of *M. purpureus*. A. View from the top, B. View from the bottom

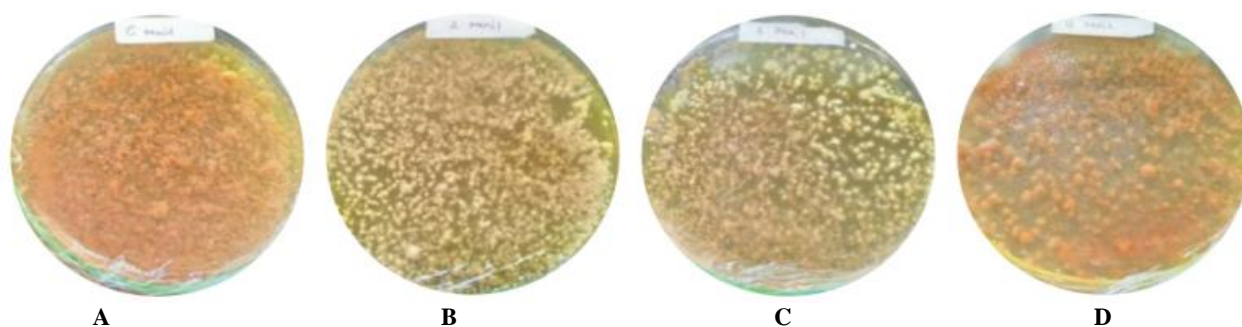


Figure 2. The colony of *M. purpureus* after UV irradiation for: A. 0, B. 2, C. 3, D. 4 min

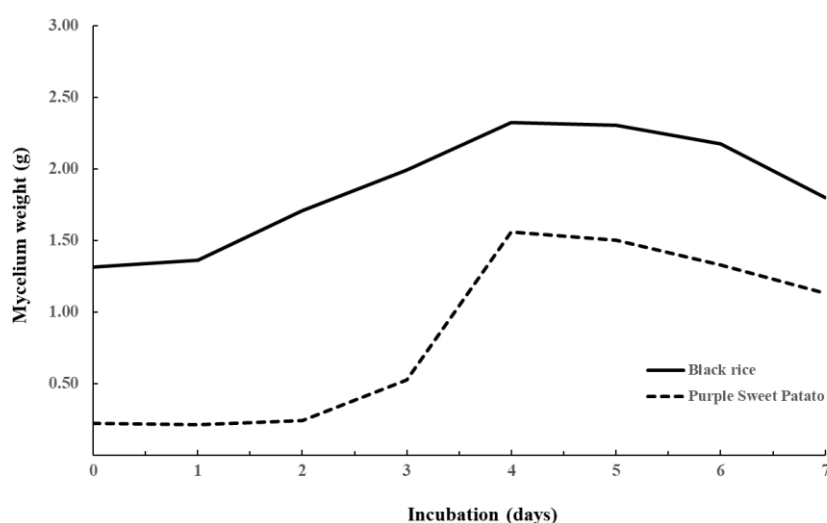


Figure 3. The growth curve of *M. purpureus* on purple sweet potato and black rice substrates

Monascus fermented product (MFP)

Fermentation processes were conducted for 21 days with an interval of 7 days. MFP–purple sweet potato was sticky solid and reddish-black, had a sweet odor, and caramelized, whereas MFP–black rice was sticky and reddish-black and had a grainy form. The yield of MFP–purple sweet potato varied from 5.97% to 26.48%. The highest yield (26.48%) was obtained from fermentation with UV-irradiated *M. purpureus* for 3 min and incubation for 21 days. The yield of MFP–black rice varied from 42.47% to 51.88%. The highest yield (51.88%) was obtained with UV-irradiated *M. purpureus* for 2 min and incubation for 21 d. The overall results indicated that MFP–black rice had a higher yield than MFP–purple sweet potato (Figure 4).

The absorbance values of the ethanol extract from MFP–purple sweet potato and MFP–black rice are shown in Table 1 and Table 2, respectively. The fermentation process with a longer incubation time produced a higher absorbance value. These results applied for both MFP–purple sweet potato and MFP–black rice. The absorbance of the yellow pigment at λ 390 nm was varied from 0.17 ± 0.01 to 3.90 ± 0.12 , whereas the absorbance of the red pigment at λ 500 nm was varied from 0.03 ± 0.00 to 3.98 ± 0.02 . The highest absorbance of both pigments in MFP–purple sweet potato was obtained from UV-irradiated *M. purpureus* for 3 min (MFP3–purple sweet potato), whereas

that in MFP–black rice was obtained from UV-irradiated *M. purpureus* for 2 min (MFP2–black rice). Both fermentations occurred in 21 days. The result of the variance analysis showed that the interaction of substrates, length of UV irradiation, and incubation time were significantly different at a 95% confidence level ($p > 0.05$). This result indicated that the type of substrate, length of UV irradiation, and fermentation time affect MFP production.

Antibacterial activity of ethanol extracts

Ethanol extracts from both MFP3–purple sweet potato and MFP2–black rice showed antibacterial activity toward *S. typhi* with inhibition zones of 1.0 and 1.5 mm, respectively. The minimum inhibition concentrations (MICs) of ethanol extracts were 0.2 g/L for MFP3–purple sweet potato and 0.15 g/mL for MFP2–black rice. This finding suggested the possible use of MFP3–purple sweet potato and MFP2–black rice as functional food ingredients not only for food colorants but also for the prevention of *S. typhi* infection. The T-test analysis result at a 95% confidence level indicated the difference in the antibacterial activities of MFP3–purple sweet potato and MFP2–black rice to *S. typhi*. Correlation test (r) between concentrations of fermented substrates showed that MFP2–black rice had a better antibacterial activity to *S. typhi* than MFP3–purple sweet potato.

Since MFP2-black rice has the highest antibacterial activity, its ethanol extract was used for TLC and bioautography. The TLC results gave three spots with R_f values of 0.23, 0.85, and 0.96. The results from spray reagents indicated the presence of saponin (first spot), flavonoid (second spot), and an alkaloid (third spot) compounds. Each spot was scraped carefully and subjected

to media inoculated with *S. typhi* for bioautography analysis using an aqueous solution as the negative control. After incubation, the first and second spots showed inhibition against *S. typhi* growth, with inhibition zones of 14.7 and 19.0 mm, respectively (Figure 5). These results suggested that MFP2-black rice contained saponin and flavonoid, which have antibacterial activity toward *S. typhi*.

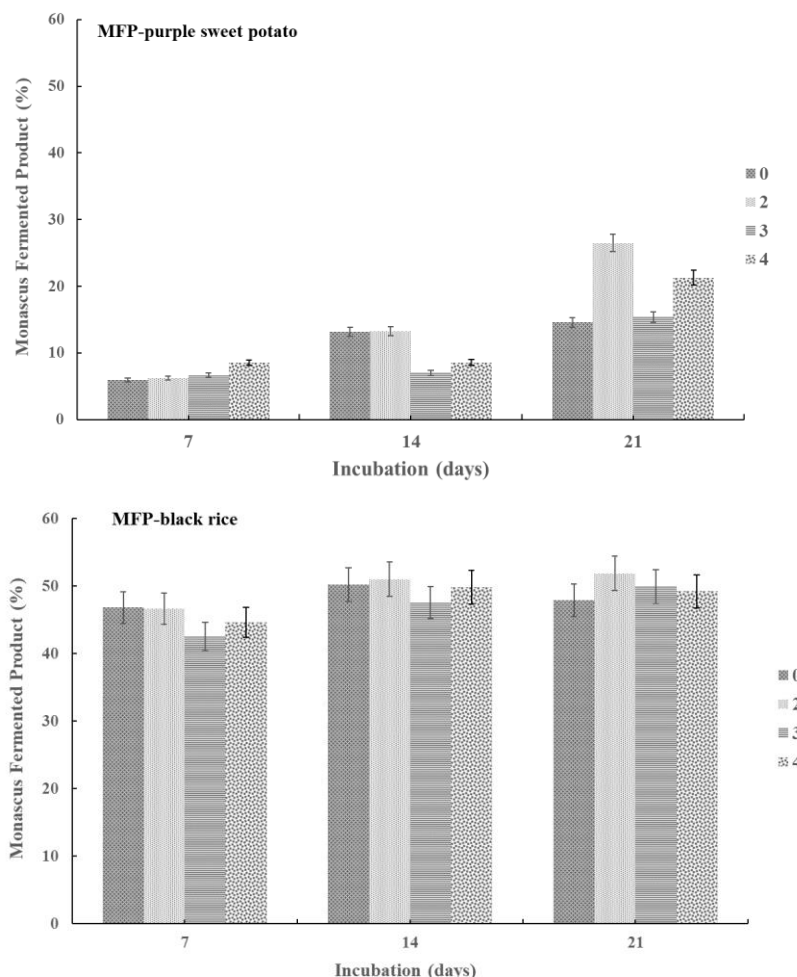


Figure 4. *Monascus* fermented product from purple sweet potato and black rice substrates

Table 1. The absorbance of ethanol extracts from MFP-purple sweet potato

Sample	Absorbance λ_{390nm}			Absorbance λ_{500nm}		
	7 days	14 days	21 days	7 days	14 days	21 days
0 min	0.30 ± 0.20	0.78 ± 0.00	3.59 ± 0.08	0.12 ± 0.05	0.53 ± 0.00	2.26 ± 0.01
2 min	0.42 ± 0.00	1.78 ± 0.00	3.78 ± 0.26	0.19 ± 0.00	1.52 ± 0.00	3.22 ± 0.01
3 min	0.17 ± 0.01	0.82 ± 0.00	3.90 ± 0.12	0.03 ± 0.00	0.56 ± 0.00	3.98 ± 0.03
4 min	0.08 ± 0.05	0.53 ± 0.00	2.37 ± 0.00	0.03 ± 0.00	0.30 ± 0.00	1.55 ± 0.01

Table 2. The absorbance of ethanol extracts from MFP-black rice

Sample	Absorbance λ_{390nm}			Absorbance λ_{500nm}		
	7 days	14 days	21 days	7 days	14 days	21 days
0 min	0.87 ± 0.01	0.43 ± 0.00	3.65 ± 0.25	0.55 ± 0.01	0.28 ± 0.01	3.60 ± 0.00
2 min	0.78 ± 0.00	2.78 ± 0.02	3.85 ± 0.17	0.45 ± 0.00	2.05 ± 0.00	3.98 ± 0.02
3 min	0.74 ± 0.01	2.36 ± 0.01	3.74 ± 0.30	0.44 ± 0.00	2.09 ± 0.01	3.48 ± 0.01
4 min	1.06 ± 0.01	1.20 ± 0.01	3.83 ± 0.06	0.67 ± 0.00	1.13 ± 0.00	3.60 ± 0.00

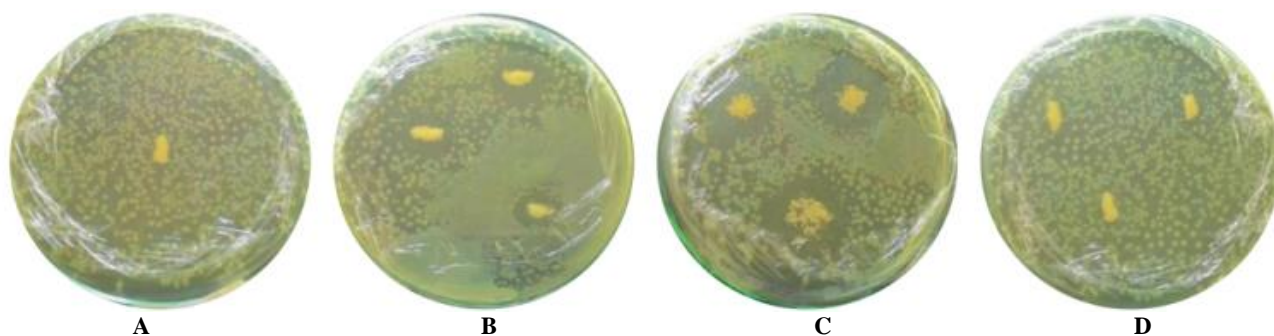


Figure 5. Bioautography results of three TLC spots from the ethanol extract of MFP2–black rice. A. The negative control, B. First spot, C. Second spot, D. Third spot

Discussion

The growth of *M. purpureus* was affected by UV irradiation. Bühler et al. (2015) stated that the average radial growth of *M. purpureus* was between 2.3 and 3.1 mm. He observed a decrease in the radial growth rate of *Monascus ruber* exposed to direct illumination. The radial growth rate was 1.50 mm in darkness, whereas it was reduced to 0.59 mm with direct illumination.

Various preparations of inoculum have been reported. Inoculum for lovastatin production was prepared with the inoculation of *M. purpureus* spore in rice substrate and incubated for 14 days (Kasim et al. 2005). Inoculum from various cereals, tuber crops, and agro-industrial residues was prepared by cultivating the spore of *Monascus* strain FJ46 for 24 h in a rotary shaker incubator (Mu et al. 2015). The inoculum of *M. ruber* CCT3802 was obtained by the germination of *M. ruber* spores for 60 h (Bühler et al. 2015). *M. purpureus* starter for MFP with antioxidant activity was prepared by inoculating *M. purpureus* M9 and incubated for 7 days (Srianta et al. 2017). The inoculum preparation varies, possibly because of the different intended utilization.

MFP–purple sweet potato gave lower yields than MFP–black rice. The difference in yield might be due to the variation in carbohydrate content. Black rice contains 74.09% to 75.71% of total carbohydrates (Sompong et al. 2011), whereas the purple sweet potato contains 22.64% of carbohydrates (Ginting et al. 2015). During fermentation, *M. purpureus* consumes starchy substrates as a source of energy for the growth and developmental needs of secondary metabolites. It produces glucoamylase and α -amylase enzymes, the key enzymes for starch hydrolysis (Yoshizaki et al. 2010). Thus, the difference in carbohydrate content apparently results in a different fermentation product yield.

Monascus produces pigments at various incubation times. *M. purpureus* M9 produced MFP with antioxidant activity after 14 days of incubation (Srianta et al. 2017). Mutant *Monascus* sp. TISTR 3179 rapidly produced yellow pigments after 5 days of cultivation using rice, corn, mung bean, soybean, potato, sweet potato, and cassava tubers as substrates (Yongsmith et al. 2013). Huang et al. (2019) observed that the cell growth of mutant *Monascus* U-2 increased rapidly and reached its maximum at the 10th–11th

day of incubation. During the exponential phase of cell growth, monacolin K was produced and reached its maximum during the 17th–18th day of incubation. These observations indicated that *Monascus* produced secondary metabolites during the stationary phase of cell growth. However, a shorter incubation time for monacolin K production was reported by Sun et al. (2011). Monacolin K level reached the highest on day 13 when *Sporobolomyces huaxiensis* was introduced as a fungal elicitor in the fermentation system.

In this research, *M. purpureus* converted the substrate into primary metabolite products for cell growth and had sufficient time to form pigments as secondary metabolites during stationary phases from 7 to 21 days of incubation. UV irradiation toward *M. purpureus* increased the intensity of the pigment until it reached a certain exposure time. Longer exposure to UV irradiation decreased the intensity of pigments, which might alter the microbial ability to produce pigments. MFPs with the highest absorbance values, which were MFP3–purple sweet potato and MFP2–black rice, were further analyzed for antibacterial activity.

Monascus pigments produced by *M. ruber* CCT 3802, *M. purpureus*, *M. purpureus* N11S, and *Monascus* M3428 showed antimicrobial activity against *Escherichia coli*, *Salmonella enteritidis*, *B. subtilis*, *S. aureus*, and yeast, respectively. These antimicrobial activities of *Monascus* pigments indicate their potential as food preservatives (Kim and Ku 2018). Christiana (2016) mentioned that the antimicrobial activities of *Monascus* pigments vary with the type of pigment. Ungureanu and Ferdes (2010) reported that the red pigment from red yeast rice inhibited the growth of *Bacillus subtilis*, *Pseudomonas aeruginosa*, and *Streptomyces albus*. The bacteriostatic effects suggested the preservative value of the *Monascus* pigment. Rojsuntornkitti et al. (2010) used *M. purpureus* TISTR 3080 to produce Chinese red broken rice with antibacterial activity against *C. perfringens*, *Salmonella* spp., and *S. aureus*. The yellow pigment produced by mutant *Monascus* sp. TISTR 3642 was useful for the protection of food products, such as fresh Chinese noodles, or non-food products, such as cosmetics (Yongsmith et al. 2013).

Purple sweet potato and black rice served as potential substrates for MFP using UV-irradiated *M. purpureus* for 3 and 2 min, respectively. Solid-state fermentation was

carried out for 21 days to obtain the highest yield. The ethanol extracts from both MFP–purple sweet potato and MFP–black rice showed antibacterial activities against *S. typhi*. Further studies on the elucidation of the bioactive compounds from MFP–purple sweet potato and MFP–black rice will be considered for a better understanding of their pharmaceutical benefits.

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Our decision is: Revisions Required

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Reviewer E:

Line 73. Could you please avoid using objectives and aim in the same sentence.

Line 103. Why did the authors choose to use a 21 days fermentation?

Recommendation: Revisions Required

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