

The Second International Conference on Genetic Resources and Biotechnology

Harnessing Technology for Conservation and Sustainable Use of Genetic Resources for Food and Agriculture

Bogor, Indonesia • 24–25 May 2021

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Rerenstradika Tizar Terryana and Dani Satyawan



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Preface: The Second International Conference on Genetic Resources and Biotechnology

The Second International Conference on Genetic Resources and Biotechnology, which is the continuation of the first event held in 2018, focuses on topics related to advances in biotechnology to create more opportunities for effective conservation and sustainable utilization of genetic resources for food and agriculture. This year conference's theme is Harnessing Technology for Conservation and Sustainable Use of Genetic Resources for Food and Agriculture. The conference was organized by Indonesian Agency for Agricultural Research and Development (IAARD), Ministry of Agriculture, Indonesia, in collaboration with Indonesian Biotechnology Consortium and held on 24th-25th of May 2021 virtually due to the pandemic of COVID-19.

The conference aims to share and exchange current scientific information and technological developments on biotechnology and their applications for conservation and sustainable use of genetic, to encourage and promote quality, efficiency, and modernization of management and utilization of genetic resources, and to facilitate national and international collaboration among participants. There are five scopes discussed in this conference. They are effective management of conservation and sustainable use of genetic resources for food and agriculture, application of genomics and molecular markers for genetic resource conservation and crop adaptation to climate change, application of innovative crop improvement techniques for conservation and sustainable use of plant genetic resources for food and agriculture, plant cell and tissue culture for conservation and effective utilization of genetic resources, and the use of microbial genetic resources as biological control agents of agricultural pests and diseases, and for soil bioremediation.

Five speakers from the United States of America, Japan, India and Indonesia were invited to discuss about their expertise and knowledge on relevant subjects in the plenary sessions. This conference was attended by more than 100 participants including 75 presenters and 44 listeners worldwide. They came from diverse governmental, private, or academic institutions and also scientific communities. The presented materials have undergone peer review processes and only qualified papers were selected. Furthermore, all papers were subjected to double blind peer-review and expected to meet the scientific criteria of significance and academic excellence to be published in a conference proceedings indexed in a well-known, reputable service.

We would like to express our sincere gratitude to our speakers, presenters and all participants for their contributions in this conference. We would also like to express our appreciation for the generosity of our sponsors that support this conference: PT CropLife, PT ITS Science Indonesia, PT Fajar Mas Murni and PT Prima Instrument Analitika. Lastly, special thanks to all committee members for their exceptional work and contributions in the conference and publication.

Chair of Organizing Committee

Dr. Toto Hadiarto

Table of Contents

THE SECOND INTERNATIONAL CONFERENCE ON GENETIC RESOURCES AND BIOTECHNOLOGY: Harnessing Technology for Conservation and Sustainable Use of Genetic Resources for Food and Agriculture

[_PREV](#)



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DISPLAY :

- [20](#)
 - [50](#)
 - [100](#)
 - [all](#)
-

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EFFECTIVE MANAGEMENT OF CONSERVATION AND SUSTAINABLE USE OF GENETIC RESOURCES FOR FOOD AND AGRICULTURE

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Harnessing plant genetic resources through biotechnology for food security in Indonesia

[Mastur](#), [Reflinur](#), [Nurul Hidayatun](#), [Sustiprijatno](#), [Fatimah](#), [Tri Puji Priyatno](#) and [Puji Lestari](#)
AIP Conference Proceedings **2462**, 020001 (2022); <https://doi.org/10.1063/5.0075671>

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DNA barcoding of *Vatica bantamensis*, a critically endangered tree endemic to Banten, Indonesia

[Muhammad Rifqi Hariri](#), [Iyan Robiansyah](#), [Dipta Sumeru Rinandio](#), [Dodo](#), [Desi Siti Sundari](#), [Cecep H. Sukmawan](#) and [Bayuntoro Ardi](#)
AIP Conference Proceedings **2462**, 020002 (2022); <https://doi.org/10.1063/5.0075529>

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Genetic parameters of agronomic traits in soybean (*Glycine max* [L.] Merrill) genotypes tolerant to drought

[Made J. Mejaya](#), [Suhartina](#), [Purwantoro](#), [Novita Nugrahaeni](#) and [Titik Sundari](#)
AIP Conference Proceedings **2462**, 020003 (2022); <https://doi.org/10.1063/5.0075159>

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Yield stability performance of soybean (*Glycine max* [L.] Merrill) lines tolerant to drought

[Suhartina](#), [Purwantoro](#), [Novita Nugrahaeni](#), [Abdullah Taufiq](#) and [Made Jana Mejaya](#)
AIP Conference Proceedings **2462**, 020004 (2022); <https://doi.org/10.1063/5.0075158>

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FreeJanuary 2022

Polymorphisms and associations of the *RACK1* genes with antibody response to Newcastle disease in KUB chickens

[Ifa Manzila](#), [Puji Lestari](#), [Tike Sartika](#), [Tri Puji Priyatno](#), [Risa Indriani](#), [Kristianto Nugroho](#) and [Rerenstradika Tizar Terryana](#)

AIP Conference Proceedings **2462**, 020005 (2022); <https://doi.org/10.1063/5.0075622>

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Rice grain quality evaluation of promising lines of rice under irrigation and for salinity tolerance

[Dody D. Handoko](#), [Nafisah](#), [Aris Hairmansis](#), [Trias Sitaresmi](#), [Heni Safitri](#), [Satoto](#), [Ali Imamuddin](#), [Cucu Gunarsih](#) and [Untung Susanto](#)

AIP Conference Proceedings **2462**, 020006 (2022); <https://doi.org/10.1063/5.0075956>

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Existing diversity profile for kernel characteristics of maize germplasm in IAARD-ICABIOGRAD gene bank

[Andari Risliawati](#), [Sobir](#), [Trikoesoemaningtyas](#), [Willy B. Suwarno](#) and [Puji Lestari](#)
AIP Conference Proceedings **2462**, 020007 (2022); <https://doi.org/10.1063/5.0075178>

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Characterization of Japansche citroen rootstock somaclones and *in vitro* selection for aluminium tolerance

[Deden Sukmadjaja](#), [Mia Kosmiatin](#) and [Tiwi Wati](#)
AIP Conference Proceedings **2462**, 020008 (2022); <https://doi.org/10.1063/5.0077888>

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Resistance to brown planthoppers (*Nilaparvata lugens* Stål) in rice accessions originated from Sumatra Island, Indonesia

[Dodin Koswanudin](#), [Nurul Hidayatun](#) and [Muhamad Ace Suhendar](#)
AIP Conference Proceedings **2462**, 020009 (2022); <https://doi.org/10.1063/5.0075680>

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Morphological identification of underutilized local fruits in Kutai Barat Regency to support their conservation and sustainable use

[Fitri Handayani](#), [Nurbani](#) and [Asep Pebriandi](#)

AIP Conference Proceedings **2462**, 020010 (2022); <https://doi.org/10.1063/5.0075594>

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Genetic resources of adlay (*Coix lacryma-jobi* L.) in East Kalimantan as source of functional food

[Fitri Handayani](#), [Muhammad Amin](#) and [Muhammad Taufiq Ratule](#)

AIP Conference Proceedings **2462**, 020011 (2022); <https://doi.org/10.1063/5.0075593>

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Screening of soybean genotypes resistance to rust disease (*Phakopsora pachyrhizi*)

[Sumartini](#) and [Kurnia Paramita Sari](#)

AIP Conference Proceedings **2462**, 020012 (2022); <https://doi.org/10.1063/5.0075674>

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Identification of soybean promising lines resistant to pod-sucking bug, *Riptortus linearis* (Fabricius)

[M. Muchlish Adie](#), [Titik Sundari](#), [Kurnia Paramita Sari](#) and [Ayda Krisnawati](#)
AIP Conference Proceedings **2462**, 020013 (2022); <https://doi.org/10.1063/5.0075343>

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Variation in pod shattering resistance among black soybean genotypes associated with agronomic traits

[Ayda Krisnawati](#), [Titik Sundari](#) and [M. Muchlish Adie](#)
AIP Conference Proceedings **2462**, 020014 (2022); <https://doi.org/10.1063/5.0075338>

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Preliminary characterization and identification of genetic integrity of velvet bean germplasm in IAARD-ICABIOGRAD gene bank

[Nurwita Dewi](#), [Andari Risliawati](#) and [Nurul Hidayatun](#)

AIP Conference Proceedings **2462**, 020015 (2022); <https://doi.org/10.1063/5.0076355>

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Plant parasitic nematodes infesting three minor legumes (velvet bean, lablab bean, and jack bean)

[Chaerani](#), [Try Zulchi P. Hariyadi](#) and [Nurwita Dewi](#)

AIP Conference Proceedings **2462**, 020016 (2022); <https://doi.org/10.1063/5.0075204>

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Proactive management approach of seed PGRFA conservation during the pandemic of coronavirus disease (COVID-19) in Indonesia

[Nurul Hidayatun](#), [Andari Risliawati](#), [Nurwita Dewi](#), [Lina Herlina](#) and [Dodin Koswanudin](#)

AIP Conference Proceedings **2462**, 020017 (2022); <https://doi.org/10.1063/5.0075531>

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Evaluation of mung bean accessions in saline soil based on quantitative morphological characters

[Trustinah](#), [Ratri Tri Hapsari](#), [Rudi Iswanto](#) and [Rudy Soehendi](#)

AIP Conference Proceedings **2462**, 020018 (2022); <https://doi.org/10.1063/5.0075324>

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Screening and evaluation of 100 upland rice accessions for developing high-yielding upland rice varieties tolerant against acid soil

[Lina Herlina](#) and [Yusi N. Andarini](#)

AIP Conference Proceedings **2462**, 020019 (2022); <https://doi.org/10.1063/5.0075550>

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Morphological characters of sugarcane mutant (*Saccharum officinarum* L.) from *in vitro* selection for drought stress

Rr. Sri Hartati, Sri Suhesti and Nurya Yuniyati

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Identifying potential seedless citrus accessions through floral structure and pollen performance

Baiq Dina Mariana, Anis Andrini and Sri Andayani

AIP Conference Proceedings **2462**, 020021 (2022); <https://doi.org/10.1063/5.0076922>

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Secondary characters based selection of Indonesian kenaf (*Hibiscus cannabinus* L.) germplasm for developing superior varieties

Taufiq Hidayat R. S., Marjani, Nurindah, Muhammad Rasyidur Ridho, Cynthia Lestari Hertianti and Widya Fatriasari

AIP Conference Proceedings **2462**, 020022 (2022); <https://doi.org/10.1063/5.0075716>

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Genetic relationship of pigmented rice (*Oryza sativa* L.) collected from Eastern Indonesia based on morpho-agronomical traits and SSR markers

[Yusi Nurmalita Andarini](#), [Willy Bayuardi Suwarno](#), [Hajrial Aswidinnoor](#) and [Hakim Kurniawan](#)
AIP Conference Proceedings **2462**, 020023 (2022); <https://doi.org/10.1063/5.0075706>

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Rejuvenation and morphological characterization of local rice from the province of Yogyakarta

[Setyorini Widayanti](#), [Sutarno](#), [Endang Wisnu Wiranti](#) and [Kristamtini](#)
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Characterization of plant architecture and yield trait of castor (*Ricinus communis* L.) germplasm suitable for mechanical harvesting

[Tantri Dyah Ayu Anggraeni](#) and [Rully Dyah Purwati](#)

AIP Conference Proceedings **2462**, 020025 (2022); <https://doi.org/10.1063/5.0075155>

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Characterization and interrelationships of the number of vessel bundles with yield components in various genotypes of soybean (*Glycine max* [L.] Merrill)

[Anna S. Karyawati](#) and [Dyah P. Fitrawantio](#)

AIP Conference Proceedings **2462**, 020026 (2022); <https://doi.org/10.1063/5.0075693>

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Tuber starch content of edible canna (*Canna indica* L.) from different geographical origins

[Surya Diantina](#), [Randy Sanjaya](#), [Kristina Dwi Atmini](#), [Ace Suhendar](#) and [Dodin Koswanudin](#)

AIP Conference Proceedings **2462**, 020027 (2022); <https://doi.org/10.1063/5.0075922>

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The diversity of morpho-agronomic characters and identification of early maturity cassava (*Manihot esculenta* Crantz.) germplasm

[Tinuk Sri Wahyuni](#), [Kartika Noerwijati](#) and [Made J. Mejaya](#)

AIP Conference Proceedings **2462**, 020028 (2022); <https://doi.org/10.1063/5.0075658>

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Radiosensitivity and phenotypic characterization of gamma ray-induced mutant population of four *Capsicum annum* L. cultivars grown in screen house

[Andri Fadillah Martin](#), [Dyah Retno Wulandari](#), [Tri Muji Ermayanti](#), [Betolini Widhi Hapsari](#), [Erwin Al Hafiih](#) and [Laela Sari](#)

AIP Conference Proceedings **2462**, 020029 (2022); <https://doi.org/10.1063/5.0075173>

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Morphological performances of mutant butterfly pea (*Clitoria ternatea* L.)

[Try Zulchi](#), [Ali Husni](#), [Dwinita Wikan Utami](#), [Reflinur](#), [Mia Kosmiatin](#), [Tarkus Suganda](#) and [Agung Karuniawan](#)

AIP Conference Proceedings **2462**, 020030 (2022); <https://doi.org/10.1063/5.0075592>

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Screening of beta carotene and its correlation with tuber flesh color in sweet potato

[Kristina Dwi Atmini](#), [Surya Diantina](#), [Muhamad Sabda](#) and [Dodin Koswanudin](#)

AIP Conference Proceedings **2462**, 020031 (2022); <https://doi.org/10.1063/5.0075618>

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Evaluation of morpho-agronomical characters and grain quality of red rice lines

[Heni Safitri](#) and [Puji Lestari](#)

AIP Conference Proceedings **2462**, 020032 (2022); <https://doi.org/10.1063/5.0078807>

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Growth variation and relationship of clove progenies of high-yielding mother trees collected from various regions in Indonesia

[Mariana Susilowati](#), [Sri Wahyuni](#), [Adi Setiadi](#) and [Nurliani Bermawie](#)

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Screening on bast fiber plants resistant to spiral stem borer, *Agilus acutus* (Coleoptera: Buprestidae)

[Sujak](#), [Nurindah](#), [Dwi Adi Sunarto](#), [Marjani](#) and [Nurul Hidayah](#)

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Characteristic of indigenous *Leuconostoc mesenteroides* EN 17-11 protease and its stability during storage at cold and freezing temperatures

Tatik Khusniati, Ika, Harry Noviard and Sulistiani

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Performance of introduced lines based on morphological markers for diversity enrichment of Indonesian chili pepper (*Capsicum annuum* L.) varieties

Rinda Kirana, Catur Hermanto, Reflinur and Derek W. Barchenger

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Chotimatul Azmi, Imas Rita Saadah, Nazly Aswani and Asih Kartasih Karjadi

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Genetic diversity analysis of *Castanopsis argentea* using random amplified polymorphic DNA markers

Muhammad Imam Surya, Lily Ismaini, Decky Indrawan Junaedi, Aisyah Handayani, Taufikurrahman Nasution, Muhammad Efendi, Andes Hamuraby Rozak, Zaenal Mutaqien, Musyarofah Zuhri, Imawan Wahyu Hidayat, Fitri Kurniawati, Vandra Kurniawan, Dwinda Mariska Putri and Risha Amilia Pratiwi
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APPLICATION OF GENOMICS AND MOLECULAR MARKERS FOR GENETIC RESOURCE CONSERVATION AND CROP ADAPTATION TO CLIMATE CHANGE

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Muhamad Sabran, Dwinita Wikan Utami and Susilawati
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[Edy Listanto](#), [Ahmad Warsun](#), [Ahmad Dadang](#), [Eny Ida Riyanti](#), [Saptowo Jumali](#)

[Pardal](#), [Sustiprijatno](#) and [Mastur](#)

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Molecular diversity comparison in local rice accessions originated from Kalimantan and other islands of Indonesia

[Puji Lestari](#), [Rerenstradika Tizar Terryana](#), [Kristianto Nugroho](#), [Andari Risliawati](#), [Nurul](#)

[Hidayatun](#), [Priatna Sasmita](#), [Yudhi Sastro](#), [I. Gusti Komang Dana Arsana](#) and [Ikhwani](#)

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Genetic variation of Adan, a Krayan local rice mutant, using microsatellite markers

[Joko Prasetyono](#), [Tio Fadel Rafsanjani](#), [Tri Aminingsih](#), [Taslihah](#) and [Sugiono Moeljopawiro](#)

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[Ida Rosdianti](#), [Dani Satyawati](#), [Muhamad Yunus](#) and [Dwinita Wikan Utami](#)

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Field adaptation and molecular characterization of Code-*qTSN4* and Code-*qDTH8* rice lines at two different locations

[Tasliyah](#), [Kurniawan Rudi Trijatmiko](#) and [Joko Prasetyono](#)

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Hybrid purity assessment in F₁ hybrids segregating for phytophthora root rot resistance genes of chili pepper (*Capsicum annuum* L.)

[Fatimah](#), [Reflinur](#), [Joko Prasetyono](#), [Wartono](#), [Kristianto Nugroho](#), [Rinda Kirana](#), [Dani Satyawan](#), [Rerenstradika Tizar Terryana](#), [Aqwin Polosoro](#), [Puji Lestari](#) and [I. Made Tasma](#)
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Cytological and molecular identifications of seedless tangerine derived from endosperm culture

[Chaireni Martasari](#), [Mia Kosmiatin](#), [Ali Husni](#), [Kurniawan Budiarto](#) and [Innez Candri Gilang Purnama](#)
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Reflinur, Ma'sumah, Namira Nur Arfa, Budi Setiadi Daryono and Azis Natawijaya
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- [all](#)

APPLICATION OF INNOVATIVE CROP IMPROVEMENT TECHNIQUES FOR CONSERVATION AND SUSTAINABLE USE OF PLANT GENETIC RESOURCES FOR FOOD AND AGRICULTURE

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Design and *in vitro* test of sgRNA for the CRISPR/Cas9 plasmid construct of the *SQS* gene of *Artemisia annua* L.

Sri Koerniati

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The efficacy of genetically modified (GM) corn Bt11 against *Ostrinia furnacalis* (Guenee) and *Helicoverpa armigera* (Hubner)

[Bahagiawati](#) and [Diani Damayanti](#)

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Construction and introduction of OsAER1::LeAlaAT cassette to improve the nitrogen use efficiency in rice cv. Mekongga

[Atmitri Sisharmini](#), [Aniversari Apriana](#), [Intan Kamila](#), [Aqwin Polosoro](#), [Wening Enggarini](#), [Tri Joko Santoso](#), [Toto Hadiarto](#), [Bahagiawati A. Husin](#) and [Kurniawan Rudi Trijatmiko](#)

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Environmental safety assessment of genetically engineered potato resistant to late blight caused by *Phytophthora infestans*

[Alberta Dinar Ambarwati](#), [Eny Ida Riyanti](#), [Edy Listanto](#), [Tri Joko Santoso](#), [Toto Hadiarto](#) and [Kusmana](#)

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[Saptowo J. Pardal](#), [Amalia Prihaningsih](#), [Suharsono](#), [Ratna Utari](#) and [Riri Sundasari](#)
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Phenotypic and genetic stability evaluation of the targeted *GA20ox-2* gene mutation in CRISPR/Cas9 mutant rice derived from Mentong cultivar

[Aniversari Apriana](#), [Tri Joko Santoso](#), [Atmitri Sisharmini](#), [Reflinur](#), [A. Dinar Ambarwati](#), [Toto Hadiarto](#), [Sustiprijatno](#) and [Nuryati](#)
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Transformation of *csp* gene into tobacco plant mediated by *Agrobacterium tumefaciens*

[Sustiprijatno](#), [Seagames Waluyo](#) and [Suharsono](#)

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PLANT CELL AND TISSUE CULTURE FOR CONSERVATION AND EFFECTIVE UTILIZATION OF GENETIC RESOURCES

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The application of gamma ray irradiation to increase triterpenoid compounds in embryogenic calli of *Centella asiatica* L. Urban

[Ika Roostika](#), [Suci Rahayu](#) and [Nurliani Bermawie](#)

AIP Conference Proceedings **2462**, 050001 (2022); <https://doi.org/10.1063/5.0076402>

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The effect of FeSO₄ concentration on the callus growth of two chili (*Capsicum annuum* L.) varieties

[Rossa Yunita](#), [Endang Gati Lestari](#), [Iswari S. Dewi](#), [Mastur](#) and [Bambang Sapta Purwoko](#)

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Evaluation of ratooning ability in several sweet sorghum (*Sorghum bicolor* [L.] Moench) mutant lines

[Endang Gati Lestari](#), [Iswari Saraswati Dewi](#), [Rossa Yunita](#) and [Amin Nur](#)
AIP Conference Proceedings **2462**, 050003 (2022); <https://doi.org/10.1063/5.0075542>

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Response of gamma ray irradiation derived-cultures of three sugarcane varieties to drought stress induced by polyethylene glycol

[Ragapadmi Purnamaningsih](#) and [Suci Rahayu](#)
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Sucrose and putrescine increased callus induction in tomato anther culture

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Field evaluation of elephant grass mutant lines (*Pennisetum purpureum* Schumach.) in highlands

Ali Husni, Muhammad Rifay, Mia Kosmiatin and Vyta W. Hanifah
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Increasing drought tolerance of sugarcane through gamma ray irradiation and *in vitro* selection

Sri Suhesti, Syafaruddin, I. Ketut Ardana, Endang Hadipoentyanti and Rr. Sri Hartati
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Cells density affects cell production of *Citrus limonia* in flask and air-lift bioreactor cultures and limonin farming

[Dita Agisimanto](#), [Farida Yulianti](#) and [Hidayatul Arisah](#)

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THE USE OF MICROBIAL GENETIC RESOURCES AS BIOLOGICAL CONTROL AGENTS OF AGRICULTURAL PESTS AND DISEASES, AND FOR SOIL BIOREMEDIATION

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In Silico functional prediction of CAS2, a protein specifically expressed in appressorium and required for pathogenicity of *Colletotrichum gloeosporioides*

[Tri Puji Priyatno](#), [Farah Diba Abu Bakar](#), [Rohaiza Ahmad Redzuan](#), [Abdul Munir Abdul Murad](#) and [Ifa Manzila](#)

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Biofertilizer increases nutrient use efficiency (NUE) of nitrogen, phosphorus, and potassium at leaves level of *Artemisia annua* L.

[Wiguna Rahman](#), [Arthur A. Lelono](#), [Erwin Al Hafiizh](#) and [Tri Muji Ermayanti](#)

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Effect of nitrogen fixation and phosphate solubilizing bacteria on growth and yield of lowland rice in different soil type

[Ikhwani](#), [Higa Afza](#), [Siti Yuriyah](#) and [Waluyo](#)

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Morphological, physiological, and molecular identification and characterization of yeast isolated from Indonesian fruits and woods

[Rerenstradika Tizar Terryana](#), [Nazhirotul Ilmiyah](#), [Inda Setyawati](#), [Titin Haryati](#), [Karden Mulya](#), [Eny Ida Riyanti](#), [Yudi Sastro](#) and [Puji Lestari](#)

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The effect of coating application using chitosan enzymatic depolymerization on anthracnose disease suppression in mango (*Mangifera indica* L.) cv. ‘Arumanis’

[Yadi Suryadi](#), [Dwi Ningsih Susilowati](#), [I. Made Samudra](#), [Alina Akhdiya](#) and [Karsinah](#)
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Understanding yeast tolerance as cell factory for bioethanol production from lignocellulosic biomass

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Isolation and pathogenicity test of fusarium basal rot and purple blotch fungal pathogens from shallot and *Allium* spp

[Chaerani](#), [Ragapadmi Purnamaningsih](#) and [Suci Rahayu](#)

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[Yusmani Prayogo](#), [Marida Santi Yudha Ika Bayu](#), [Sri Wahyuni Indiaty](#) and [Made Jana Mejaya](#)

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[Heny Herawati](#), [Diana Nur Afifah](#), [Eni Kusumaningtyas](#), [Sri Usmiati](#), [Agus S. Soemantri](#), [Miskiyah](#), [Elmi Kamsiati](#) and [Muchamad Bachtiar](#)

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[Laksmita P. Santi](#), [Haryo T. Prakoso](#) and [Donny N. Kalbuadi](#)
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Application of phosphate solubilizing microbes to promote the effectiveness of rock phosphate on cacao seedling growth in acid soil

[Kurnia Dewi Sasmita](#), [Iswandi Anas](#), [Syaiful Anwar](#), [Sudirman Yahya](#) and [Gunawan Djajakirana](#)
AIP Conference Proceedings **2462**, 060011 (2022); <https://doi.org/10.1063/5.0075843>

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Morphological Performances of Mutant Butterfly Pea (*Clitoria ternatea* L.)

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Abstract. Butterfly pea (locally known as *kembang telang*) could be used in many functions that are valuable in the field as forage, cover crop, cut for hay, and natural dyes. This study aimed to characterize morphological performance and petal color of butterfly pea putative mutants. The research was carried out in a greenhouse of Cikeumeuh Experimental Station, ICABIOGRAD, Bogor from March to July 2020. The mutant plants were grown in completely randomized design. The results showed a relatively diverse morphology of the butterfly pea putative mutants. The plant height ranged from 51 to 270 cm, shoot diameter of 4–13 mm, early flowering appeared after 36 to 43 days, fresh weight of 18.5–109 g/plant and dry weight reached 16.1 to 95.44 g/plant. The number of seeds per pod ranged from 2 to 8 seeds, and the pod length was between 3–8 cm. The butterfly pea petal colors have been ranged from white, blue and purple. At that point, the number of branches have a correlation with plant height, while shoot diameter correlates with fresh and dry weight, and the number of pods is related to pod weight, seed number and seed weight. The butterfly pea mutant numbers 179, 182, 185, 187, 226, and 228 were selected for their good plant performance.

INTRODUCTION

Butterfly pea (*Clitoria ternatea* L.) is locally known as *kembang telang* or *kembang biru* in Indonesia, belonging to the family Fabaceae, which is deep-rooted, slender, tall, climbing plant with five leaflets, and beautiful flower color. Butterfly pea is tolerant to a various soil types (pH 5.5–8.9) such as calcareous soils. The plants can survive in both regions with excess of rainfall and prolonged drought periods. They can adapt to marginal land and wide range of regions. It can grow well in dry condition [1, 2], and can be harvested between 110 until 150 days [3]. Butterfly pea has multipurposes, such as food color agent, ornamental plants [4–6], medicinal plants [7], ground cover crops, organic fertilizers, and forages plants [1, 3]. These fodder crops can be developed with a focus on productivity, good nutrition, and adaptation ability in dryland.

The potential of butterfly pea has been used to produce biomass and quality product. The yield have high biomass, as well as nutritional value with a good protein content [2]. The characters flower of butterfly pea has various color, i.e. blue, mauve, and white color petal [8–10].

As a forage crop, this legume, when mixed with grass, has provided positive values in animal feeding. Butterfly pea has also been used as a substitute for grass. Local butterfly pea cultivar from Bali which has been used as cover crop and forage for ruminant is able to produce a dry weight of 34 g per pot [1]. The morphology characters consist of 2–4 pairs, small leaves, and deep root [8, 11]. Dry matter production ranged around 4–6 t/ha or 8–12 t/ha/year [12], providing the dry weight of forage that reaches 2.5–3.5 t/ha and seed production of 2.7 t/ha at 42 days [3]. Nutritional value of butterfly pea has high protein content (18–29%), crude fat, fiber, and good digestibility. Therefore the use of mixed fodder crops will sustain extensive livestock growth and development [3, 12, 13].

Development of superior varieties in an effort to support crop productivity can be done by doubling chromosomes plant. Plants with double chromosomes will have bigger shape and sturdier, stronger organ, as well as higher nutrient content [14]. The treatment of colchicine solution to shoot apex of buds, callus, embryo, or other growing organs may increase the number of plant chromosomes, as well as the size of the cells and nucleus [15]. Polyploid plants have large cells because the number of chromosomes is more than diploid plants [14, 15]. Moreover, naturally polyploid plants seem to enlarge the size and increase the weight of stems, leaves, roots, flowers, and fruits in mung bean [15] and can increase yields [14]. Therefore, plants with double chromosomes will be potential to become giant plants with large biomass and increase nutrition.

The potential advantages of this butterfly pea will be new opportunities in the forage plant breeding program. Based upon these considerations, the preliminary study through agronomic characterization is the first step to be carried out to determine butterfly pea mutants. The aim of this study was to characterize morphological performance and petal color of butterfly pea putative mutants.

MATERIALS AND METHODS

The research was carried out in a greenhouse of Cikeumeuh Experimental Station, Indonesian Center for Agricultural Biotechnology and Genetic Resources Research and Development (ICABIOGRAD), Bogor from March until July 2020. The planting materials of butterfly pea local cultivar from East Nusa Tenggara (ENT) were grown from ratoons which were previously induced by chemical mutagens of 0.5 mM colchicine or 12 μ M oryzalin. Sample ratoons were treated with colchicine or oryzalin mutagen by immersion for 24, 36, 72, 96, and 120 h. A total of 60 putative mutants were selected from well-survived plants as planting materials. Each butterfly pea putative mutants were grown in a pot. The experimental design of this study was arranged using a randomized block design and two replications with the treatment of mutant materials.

Each butterfly pea putative mutants and control cultivar (untreated ENT local cultivar) was cultivated in a pot with a capacity of 10 kg of soils per pot. Furthermore, each plant was given 0.5 kg of manure and inorganic fertilizers in a doses of 1.5 g of urea, 3.0 g of TSP, and 3.0 g of KCl at planting. Then, butterfly pea putative mutants were maintained in the pot and given stacks of bamboo stands, until the harvest of pods and seeds. The butterfly pea genotypes were observed based on the characters of plant height, number of branches, flower petal color, fresh and dry weight, and seed weight per plant. The data was analyzed statistically by Pearson correlation coefficient.

RESULTS AND DISCUSSIONS

The colchicine and oryzalin mutagens have different effects on plant viability (Table 1). Butterfly pea plants did not grow at the colchicine immersion for more than 72 h, while oryzalin-treated plants still grew after up to 144 h treatment. The butterfly pea survived better at longer duration of immersion by oryzalin than that by colchicine. The use of mutagens can increase or inhibit growth depending on plant materials and types [8, 13]. The optimum concentration of a solution, immersion time, pH, and adventitious organs could cause chromosome doubling [15]. The combinations of colchicine or oryzalin (i.e. the type of mutagens) and immersion time could be effective for variance morphology.

At the vegetative stage, ENT local cultivars showed a higher yield of fresh and dry biomass than mutant plants. However, at generative stage (after flowering), the productions and chlorophyll content of pod and seed of the putative mutants showed higher rates than those of ENT local cultivar. Butterfly pea mutant genotypes were late in flowering time, and this might cause increased pod and seed productions. The effect of environment and genetic variation influenced the individual in the population, which led to the variance of growth [16].

Observation on the color of the flower petal and the type of flower showed relative similarity. The color character of the flower petal has been grouped into mauve, blue, and white (Fig. 1), which is similar to petal color of *C. ternatea* cultivar from Northern Australia [9]. Meanwhile, the flower type belong to a single petal type (Fig. 1). The use of chemical mutagens of colchicine or oryzalin suggested to establish the flower color. The establishment of flower color can be obtained by crossing plants. Butterfly pea is predominated by a self-pollinated species with petal colors of white, blue to purple flowers, but often, it can be considered as cross-pollinated species due to the appearance of segregating genotypes in some populations [9, 13, 16]. Progenies of artificial crosses resulted in variation of flower color viz., light pink, cream flower with blue borders, medium blue, dark blue with velvety appearance, violet, and dark violet as compared to the parental colors [16].

TABLE 1. Morphological characters and yield of control and putative mutants of butterfly pea local cultivar from East Nusa Tenggara (ENT) in Cikeumeuh Experimental Station, ICABIOGRAD, Bogor.

Characters	Means	
	ENT local cultivar	Putative mutants
Plant height (cm)	147.40	131.90
Branches number	17.26	11.89
Shoot diameter (mm)	8.57	9.06
Flowering time (days)	36.00	43.00
Total fresh biomass (g)	137.77	89.68
Total dry biomass (g)	51.62	48.09
Chlorophyll content	38.66	41.77
Pod number	9.57	34.41
Total number of seeds	47.19	57.45
Pod weight (g)	16.10	17.28
Seed weight (g)	1.035	2.37



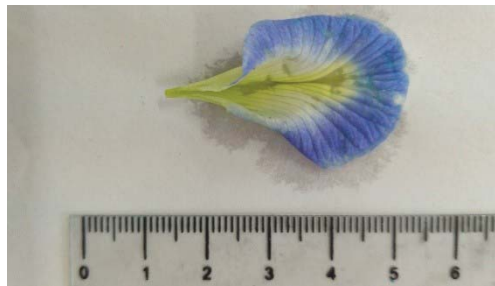
(a)



(b)



(c)



(d)



(e)



(f)

FIGURE 1. Petal flower performance of butterfly pea cultivars (a, b) and color variance of putative mutants: mauve (c), pale blue (d), blue (e), and white (f).

Pearson correlation coefficient in morphology and agronomy of butterfly pea showed relative moderate correlation rate and positive rate among biomass yields, i.e. fresh and dry weight, which were contributed by branches number and shoot diameter (Table 2). Moreover, the number of branches has a moderate correlation and positive rate to the shoot diameter. However, low negative value was shown by flowering time. The pod and seed yield in butterfly pea and pod number generated strong positive correlation to pod weight, number seed, and seed weight respectively.

TABLE 2. Correlation coefficient rate of morphological characters and yield of butterfly pea.

	PH	BN	SD	FT	TFB	TDB	PN	CC	SN	PW	SW
PH	1										
BN	0.108	1									
SD	0.015	0.343**	1								
FT	0.094	-0.203	-0.139	1							
TFB	0.009	0.326*	0.347**	-0.281	1						
TDB	0.011	0.352*	0.345*	-0.431**	0.779**	1					
PN	-0.044	0.080	-0.034	-0.250	-0.209	0.007	1				
CC	-0.113	-0.078	-0.109	0.016	-0.138	-0.030	-0.141	1			
SN	0.066	-0.032	-0.116	-0.113	-0.239	-0.069	0.742**	0.011	1		
PW	-0.081	0.057	-0.049	-0.249*	-0.247	-0.044	0.973**	-0.090	0.695**	1	
SW	0.064	-0.034	-0.079	-0.120	-0.230	-0.034	0.782**	0.000	0.979**	0.733**	1

PH = plant height, BN = branches number, SD = shoot diameter, FT = flowering time, TFB = total fresh biomass, TDB = total dry biomass, PN = pod number, CC = chlorophyll content, SN = seed number per pod, PW = pod weight, SW = seed weight. *Correlation is significant at the 0.05 level. **Correlation is significant at the 0.01 level.

According to the early growth, butterfly pea was upright and slender until the plant reached the height ranged from 30–40 cm. During the growth stage, number of branches increased and plant height grew horizontally to form tight stands and covered the ground [5, 7]. Branching was significantly correlated with foliage, while seed number was correlated with seed weight [9]. The implications of the results are that genetic improvements of polyploid butterfly pea can be performed on the characters of plant height, number of branches, and shoot diameter at vegetative stage, while the characters of pod weight and seed yield are suggested for reproduction. The blue-petaled butterfly pea mutants of 179, 182, 185, 187, 226, and 228 were selected for biomass production due to good plant performance.

CONCLUSION

Butterfly pea of ENT local cultivar have a higher yield of fresh and dry biomass than mutant plants at vegetative stage, while at generative stage the production of pods, seeds, and chlorophyll content of mutant plants were higher than that of the local cultivar.

Correlation coefficient in butterfly pea showed relative moderate correlation value among biomass yield, which contributed to the branches number and shoot diameter. Moreover, the number of branches has a positive correlation with plant height. The number of pods is related strong correlation to pod weight, number seed, and weight seed. This study showed the effect of genetic polyploid improvement to the plant organ performance. The butterfly pea mutant numbers 179, 182, 185, 187, 226, and 228 were selected due to their good plant performance.

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