

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

Editors • I Made Tasma, Dwinita Winkan Utami, Ika Roostika,
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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



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- [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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FreeJanuary 2022

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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-
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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

AIP Conference Proceedings **2462**, 020020 (2022); <https://doi.org/10.1063/5.0075656>

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Identifying potential seedless citrus accessions through floral structure and pollen performance

Baiq Dina Mariana, Anis Andrini and Sri Andayani

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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 Aswidinnor](#) 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 Widyayanti](#), [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, *Agrilus acutus* (Coleoptera: Buprestidae)

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

AIP Conference Proceedings **2462**, 020034 (2022); <https://doi.org/10.1063/5.0075691>

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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 annum* L.) varieties

[Rinda Kirana](#), [Catur Hermanto](#), [Reflinur](#) and [Derek W. Barchenger](#)

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The growth of garlic internal sprout on different storage condition

[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, Dwindi 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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Current status of tidal swamp rice varieties and its improvement for Fe toxicity tolerance and biofortification

Muhamad Sabran, Dwinita Wikan Utami and Susilawati
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Agroforensic, a new emerging study using molecular marker technique

[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](#), [Tasliah](#) and [Sugiono Moeljopawiro](#)

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The genome sequence of Ciherang, an Indonesian rice mega variety, revealed the footprints of modern rice breeding

[Ida Rosdianti](#), [Dani Satyawan](#), [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

[Tasliah](#), [Kurniawan Rudi Trijatmiko](#) and [Joko Prasetyono](#)
AIP Conference Proceedings **2462**, 030006 (2022); <https://doi.org/10.1063/5.0075661>

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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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Characterization of genomic variation on three Indonesian oil palm genotypes analyzed using next-generation sequencing HiSeq

I. Made Tasma, Habib Rijzaani, Dani Satyawan, Ida Rosdianti, Edy Supriyanto and Razak Purba
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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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Improvement of sex determination of salak plant using sequence characterized amplified regions

Reflinur, Ma'sumah, Namira Nur Arfa, Budi Setiadi Daryono and Azis Natawijaya
 AIP Conference Proceedings **2462**, 030010 (2022); <https://doi.org/10.1063/5.0075698>

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



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Volume number: 2462

Published: Jan 19, 2022

DISPLAY :

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

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

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

FreeJanuary 2022

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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FreeJanuary 2022

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

Bahagiawati and Diani Damayanti

AIP Conference Proceedings **2462**, 040002 (2022); <https://doi.org/10.1063/5.0075312>

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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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FreeJanuary 2022

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

AIP Conference Proceedings **2462**, 040004 (2022); <https://doi.org/10.1063/5.0075612>

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Backcrossing of soybean lines containing aluminium tolerance gene into superior soybean variety, Biosoy

[Saptowo J. Pardal](#), [Amalia Prihaningsih](#), [Suharsono](#), [Ratna Utari](#) and [Riri Sundasari](#)
AIP Conference Proceedings **2462**, 040005 (2022); <https://doi.org/10.1063/5.0075187>

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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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FreeJanuary 2022

Transformation of *csp* gene into tobacco plant mediated by *Agrobacterium tumefaciens*

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

AIP Conference Proceedings **2462**, 040007 (2022); <https://doi.org/10.1063/5.0075571>

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

FreeJanuary 2022

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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FreeJanuary 2022

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

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

AIP Conference Proceedings **2462**, 050002 (2022); <https://doi.org/10.1063/5.0075223>

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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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-
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FreeJanuary 2022

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

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

AIP Conference Proceedings **2462**, 050004 (2022); <https://doi.org/10.1063/5.0075185>

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Sucrose and putrescine increased callus induction in tomato anther culture

Iswari Saraswati Dewi, Imam Nur Kholis, Bambang Sapta Purwoko and Ratna Ningsih
AIP Conference Proceedings **2462**, 050005 (2022); <https://doi.org/10.1063/5.0075666>

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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
AIP Conference Proceedings **2462**, 050006 (2022); <https://doi.org/10.1063/5.0076418>

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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
AIP Conference Proceedings **2462**, 050007 (2022); <https://doi.org/10.1063/5.0076155>

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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

AIP Conference Proceedings **2462**, 050008 (2022); <https://doi.org/10.1063/5.0075651>

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

FreeJanuary 2022

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

AIP Conference Proceedings **2462**, 060001 (2022); <https://doi.org/10.1063/5.0075625>

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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 Hafiih and Tri Muji Ermayanti

AIP Conference Proceedings **2462**, 060002 (2022); <https://doi.org/10.1063/5.0075503>

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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

AIP Conference Proceedings **2462**, 060004 (2022); <https://doi.org/10.1063/5.0075170>

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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](#)
AIP Conference Proceedings **2462**, 060005 (2022); <https://doi.org/10.1063/5.0075183>

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Understanding yeast tolerance as cell factory for bioethanol production from lignocellulosic biomass

[Eny Ida Riyanti and Edy Listanto](#)
AIP Conference Proceedings **2462**, 060006 (2022); <https://doi.org/10.1063/5.0075157>

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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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Morphological characters and efficacy of thirteen entomopathogenic fungi of *Aschersonia aleyrodis* Webber isolates on whitefly (*Bemisia tabaci* Gennadius)

Yusmani Prayogo, Marida Santi Yudha Ika Bayu, Sri Wahyuni Indiati and Made Jana Mejaya

AIP Conference Proceedings **2462**, 060008 (2022); <https://doi.org/10.1063/5.0076067>

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Physicochemical characteristics of yoghurt from various beans and cereals

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

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The potential use of zeolite and exopolysaccharide bacteria for reduction of degradation and carbon emission on oil palm plantation in tropical peatland

[Laksmita P. Santi](#), [Haryo T. Prakoso](#) and [Donny N. Kalbuadi](#)
AIP Conference Proceedings **2462**, 060010 (2022); <https://doi.org/10.1063/5.0075506>

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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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Evaluation of Ratooning Ability in Several Sweet Sorghum (*Sorghum bicolor* [L.] Moench) Mutant Lines

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Abstract. Sweet sorghum (*Sorghum bicolor* [L.] Moench) is a multipurpose plant which can be grown for grain, forage, silage, syrup and/or sugar, and bioethanol production. Ratooning plant is an additional double ratooning scheme whereby the plant is harvested two or more times from a single planting during the growing season. The character of the main plant in the sorghum plant will determine the yield of the ratoon plant, however, information on the ratooning ability is lacking. The research objective was to determine the ratooning ability in the mutant lines of Numbu sorghum. The plant material consisted of 20 genotypes of the M₆ mutant line which developed from gamma ray irradiation of Numbu variety. This research was arranged using completely randomized design which consisted of three replications with a cutting height of 10 cm. Each mutant line as a treatment was planted in a plot size of 3 m × 2.5 m, with 70 cm spacing between rows and a 25 cm spacing in row. The observation was conducted on plant height, stalk diameter, panicle weight, and length, and sugar content (brix) of the stalk sap. The results indicated that mutant lines were having more or similar values when compared to the main plant in plant height, stalk diameter, and sugar content of stalk sap. The mutant lines N17-2, N17-3, and N17-5 that have high ratooning ability which is a desirable trait, as it reduces overall inputs in terms of seed and labor for field preparation, will be further used in research concerning their utilization for feeds and the fermentation industry.

INTRODUCTION

Sweet sorghum (*Sorghum bicolor* [L.] Moench) is a multipurpose plant. Their stalk and seed contain high level of fermented lignocellulose and saccharides [1, 2]. In several countries, sorghum has been used as a source of material for food and feed industry [3, 4]. Among the parts of the sorghum, their stalk plays a major role in sap production which used as raw material to produce bioethanol [1, 3, 5, 6], through fermentation process [7]. Extracted sap from sorghum stalks can be processed into liquid sugar owing to the its high sucrose content, specifically 30% sucrose, 18.64% fructose, and 12.45% glucose [1, 7–9]. Sorghum plants have advantages, one of them is an ability to adapt to suboptimal environmental conditions such as drought, therefore they can be cultivated in less-fertile fields and is well suited to be used as a source of bioethanol [10, 11]. The leaves and stalks of sweet sorghum contain nutrients which identical to Napier grass, hence they are suitable to be processed as raw material for green fodder [5, 6]. Prospect for sweet sorghum as a green fodder and bioethanol ingredient needs to be supported by findings sweet sorghum varieties that possess the advantages of biomass, ratooning ability, and high stalk sap sugar content [11, 12]. One of many problems faced by ranchers is the increasing amount of raw materials needed for animal feed as livestock population increases, therefore research is deemed necessary to find lines that possess high ratooning ability and superior quality [11].

The advantages of cultivating sorghum, among other things, are ability to produce new shoots from the stump of trimmed stalks [12]. The stumps could produce new tillers which then be harvested again, these new tillers are known as ratoon plant [12]. According to Meliala *et al.* [12] and Tsuchihashi and Goto [13], sorghum plants can be

harvested two to three times, including the main plant and the ratoons, which allows production increase in the next harvest season.

The advantages of ratoon cultivation on sorghum plants includes a relatively shorter growing time compared to the main plant, less water requirements and lower production costs [12, 14, 15]. Efendi *et al.* [15] showed that sorghum of the Numbu variety has a high ratooning ability with the percentage of first ratoon emergence exceeding 75%. Research conducted by Lestari *et al.* [16] showed a similar result that sorghum of the Numbu variety demonstrate a considerable ratooning ability. The advantage of high ratooning ability can be utilized to reduce costs (energy, labor, and time) because one planting session can provide enough sustenance for two or three harvest seasons [15]. The material used in this experiment were 6th generation mutant (M₆) sourced from selected seed, based on their performance on seed production and brix value of stalk sugar content. The research objective was to determine the ratooning ability in 20 mutant lines of Numbu sorghum.

MATERIALS AND METHODS

The research was conducted at the Cikeumeuh Experimental Station, Indonesian Center for Agricultural Biotechnology and Genetic Resources Research and Development (ICABIOGRAD), Bogor from March to September 2018. The planting material used in this study was 20 sorghum mutant lines of Numbu variety. Every plant was planted within a plot size of 3 m × 2.5 m, with 70 cm spacing between rows and 25 cm spacing within rows. Each plot contains 4 rows, and 10 planting holes were set in each row. The total plants grown for each plot were 40 plants.

Fertilization is performed twice, the first fertilization dosage consists of 100 kg/ha urea and 150 kg/ha Phonska at 12 days after sowing, while second fertilization was given at 35–40 days after sowing with a dosage of 150 kg/ha urea and 150 kg/ha Phonska. Sorghum plant that grows from seed is called the main plant. After harvesting the main plant at 105 days, ratoon is performed once by cutting the main stalk and leaving one node from the surface of the soil. The plant that grows from the node that has been trimmed is called ratoon. For ratoon maintenance, the amount of fertilizer dosage is similar with the main plant, and fertilizer is given three weeks after cutting.

Five plants from each line were observed as samples. Variables of vegetative character observed in the yield of ratoon plants were plant height (cm), stalk diameter (mm), and panicle length. Plant height measured from the base of the stalk to the point where the panicles emerged. Stalk diameter (bottom, middle, and top; mm) measured using Vernier calipers. Reproductive characters observed at 105 days consisted of the weight of full seed per panicle, fresh weight, and dry weight per panicle, and stalk sugar content (brix) measured using refractometer. There were five tillers produced, but only one of each tiller was observed in this experiment. The experiments were arranged in a complete randomized design. Data was analyzed using ANOVA test in SAS program version 12.0, further analysis was conducted using Duncan's Multiple Range Test (DMRT).

RESULTS AND DISCUSSIONS

As observed in the growth of plant height and stalk diameter in main and ratoon plants, there were five mutant lines that grew larger than Numbu, namely N17-1, N17-2, N17-3, N17-4, and N17-5. These five lines also showed optimal growth in their ratoon plants (Table 1). N17-2 line was the highest plant at 289.2 cm which is not significantly different from line N17-3 with a height difference of 5.6 cm. Those two lines were significantly different from lines N17-1, N17-4, and N17-5. In plant height variable, ratoon line N17-3 produced the highest plant at 262 cm.

Results of correlation analysis indicate the efficacy for biomass production per ha has a significant to plant height with a correlation coefficient value of 0.56 [10]. This analysis shows that the higher the plant, the higher the productivity of fresh biomass per ha, and *vice versa*, the lower the plant, the lower the fresh biomass per ha.

As for stalk diameter variable, it showcase done stable line, the main plant and ratoon plant produced larger stalks than Numbu, specifically line N17-5. The stalk diameter of the main plant was measured at 18.4 mm, its ratoon plant was 17.2 mm, whereas in Numbu it was 14.6 mm.

In this study, the yield components (panicle length and panicle weight) were also observed to see a decrease in yield on ratoon plants. Main plants showcased multiple lines that grow longer panicle than Numbu, namely N6-3, N25-2, N2-1, N5-1, and N21-3 with panicle length ranging from 22.5–23.4 cm, while on Numbu it was only 21.8 cm. For the ratoon plants, there were 8 lines which had different panicle length from the parent plants (Table 2). Out of those 8 lines, 3 lines produced panicles with the same length as the main plant, these lines were N1-4, N2-1,

N2-2, N7-4, N9-1, N17-1, N25-2, N5-1, N21-2, and N17-5. In wet panicle weight variable, there was a decrease in both Numbu parent and tested lines. Of the 20 lines tested, one line namely N17-5 yielded the same results as the main plant and was superior to Numbu.

TABLE 1. Plant height and stalk diameter of main and ratoon plants of sorghum mutant lines.

No.	Lines	Height of main plant (cm)	Height of ratoon plant (cm)	Stalk diameter of main plant (mm)	Stalk diameter of ratoon plant (mm)
1	N1-1	249.6de	204.0 de	15.8 bc	13.0 fgh
2	N1-4	230.0 ef	227.0 bcd	15.6 bc	12.0cdef
3	N2-1	241.6 de	245.6 ab	16.0 bc	12.2 gh
4	N2-2	248.8 de	235.8 ab	16.0 bc	15.6 abcd
5	N6-2	257.2 bcd	221.8 bcd	15.0 bc	15.8 abc
6	N6-3	252.4 cd	228.0 bcd	14.2 c	13.8 efg
7	N7-4	245.6 g	223.6 bcd	15.4 bc	15.0 bcde
8	N8-1	252.6 cd	241.0 ab	16.0 bc	14.0 def
9	N9-1	246.6 de	247.6 ab	16.0 bc	14.0 def
10	N17-1	276.6 ab	241.0 ab	17.0 ab	14.6 bcdef
11	N19-1	240.2 def	244.8 ab	16.2 b	15.0 bcde
12	N25-2	209.0 g	176.2 f	16.6 ab	14.4 cdef
13	N2-1	221.8 ef	194.8 ef	16.6 ab	11.4 h
14	N5-1	240.2 def	207.4 cde	16.6 ab	14.4 cdef
15	N7-3	246.0 de	226.8 bcd	15.8 bc	13.6 efg
16	N21-2	239.8 def	231.8 bc	16.2 b	14.0 def
17	N17-2	289.2 a	247.4 ab	17.0 ab	14.2 cdef
18	N17-3	283.6 a	262.0 a	16.8 ab	16.2 ab
19	N17-4	271.2 abc	244.6 ab	16.2 b	14.2 cdef
20	N17-5	272.8 ab	259.0 a	18.4 a	17.2 a
21	Numbu (control)	241.8 de	228.0 bcd	15.2 bc	14.6 bcdef

Numbers in the same column followed by the same letter are not significantly different by DMRT at $P \leq 0.05$.

TABLE 2. Panicle length and panicle weight of main and ratoon plants of sorghum mutant lines.

No.	Lines	Panicle length of main plant (cm)	Panicle length of ratoon plant (cm)	Panicle weight of main plant (g)	Panicle weight of ratoon (g)
1	N1-1	22.1 abcdef	19.5 fghi	66.3 bcdef	58.4 abcd
2	N1-4	23.1 abcd	21.1 abcdefg	61.8 defgh	57.2 abcd
3	N2-1	21.9 cdef	21.7 abcdefg	58.5 fgh	58.4 abcd
4	N2-2	21.7 f	21.2 abcde	66.4 bcdef	61.2 abc
5	N6-2	22.8 abcde	20.6 abcdefghi	70.6 abcdefg	56.6 abcd
6	N6-3	22.5 abc	29.4 bcdefghi	53.6 h	54.2 bcd
7	N7-4	21.9 cdef	21.7 abcdef	58.8 fgh	59.8 abcd
8	N8-1	21.1 f	19.6 defghi	67.3 bcdefgh	61.8 abc
9	N9-1	22.5 abcde	21.2 abcdefgh	65.1 cdefg	60.2 abc
10	N17-1	22.5 abcde	22.1 ab	63.7 defgh	61.4 abc
11	N9-1	22.1 abcdef	18.7 i	76.8 abcd	60.0 abcd
12	N25-2	23.4 a	21.9 abc	76.4 abcd	60.4 abc
13	N2-1	23.3 ab	19.5 efghi	81.2 abc	54.8 abcd
14	N5-1	23.3 ab	22.9 a	81.7 ab	63.6 a
15	N7-3	22.4 abcdef	20.8 abcdefghi	75.1 abcde	53.8 cd
16	N21-2	23.2 ab	21.8 abcd	72.9 abcdef	60.0 abcd
17	N17-2	22.5 abcde	19.4 ghi	59.5 efg	54.2 bcd
18	N17-3	22.5 abcde	19.0 bcdefghi	54.2 h	59.4 abcd
19	N17-4	22.0 bcdef	20.9 abcdefghi	54.4 gh	61.6 abc
20	N17-5	22.9 abcde	21.6 abcdefg	86.3 a	63.2 ab
21	Numbu (control)	21.8 def	19.1 hi	66.0 bcdefgh	50.7 d

Numbers in the same column followed by the same letter are not significantly different by DMRT at $P \leq 0.05$.

In this study, the measurement of stalk sugar content (brix) was carried out to see the efficacy of ratoon plant as a raw material for liquid sugar and bioethanol production. The results showed that the main plants had 13 lines with higher sugar content than Numbu, while in ratoon plants it decreased to 8 lines. The range of sugar content in those 8 lines were 10.1–13.3%, while in Numbu it was only 8.9%. Out of 20 lines tested, there were 7 stable lines with significantly higher results than Numbu, namely N1-4, N2-2, N25-2, N5-1, N21-2, N17-2, and N17-3 (Table 3). The results obtained showed that in ratoon plants, the sugar content (brix) remains high, thus these lines have the potential to be used as a ratoon plant. The high stalk sugar content provides an opportunity to process the sap into liquid sugar or alcohol. The higher sugar content in the stalk, the more level of bioethanol produced [5].

TABLE 3. Brix value (%) of main and ratoon plants of sorghum mutant lines.

No.	Lines	Brix value of main plant	Brix value of ratoon plant
1	N1-1	9.0 efg	10.1 bcde
2	N1-4	11.0 abcde	11.3 abcd
3	N2-1	12.1 abc	12.1 ab
4	N2-2	9.6 efg	6.0 g
5	N6-2	10.4 bcdef	5.4 f
6	N6-3	10.5 bcde	5.4 g
7	N7-4	9.8 defg	6.6 fg
8	N8-1	9.6 efg	9.4 cde
9	N9-1	9.9 defg	10.1 bcde
10	N17-1	10.2 cdef	9.3 de
11	N9-1	6.4 hi	10.7 bcde
12	N25-2	8.0 gh	12.4 ab
13	N2-1	8.0 gh	11.8 ab
14	N5-1	8.3 fgh	11.3 abcd
15	N7-3	7.8 ghi	10.5 bcde
16	N21-2	6.7 hi	11.8 ab
17	N17-2	13.3 a	13.3 a
18	N17-3	11.9 abcd	11.8 abc
19	N17-4	9.6 efg	10.1 bcde
20	N17-5	12.5 ab	9.3 de
21	Numbu (control)	5.71 i	8.9 ef

Numbers in the same column followed by the same letter are not significantly different by DMRT at $P \leq 0.05$.

In the variables of plant height, stalk diameter, and sugar content (brix), there were mutant lines with higher, similar, and lower yields compared to main plant. As for variables of plant height and sugar content (brix), three lines showed great potential to be used as ratoon plants, namely N17-2, N17-3, and N17-5, which could be further selected into superior lines of ratoon plants. One way to increase the productivity of sorghum is through the use of ratoon plants, which is expected to produce cheaper product while avoid competition with food plants. Yield and quality evaluation of ratoon plants that were carried out by Liu *et al.* [14] and Vinutha *et al.* [17] obtained a similar results that showed a decrease in ratoon plant yields.

Sugar content increase of ratoon plant in this study is assumed to occur because it was harvested in a period of less rain, which happened around September, while the harvest of main plant is conducted in May. Mathur *et al.* [1] stated that sugar production and seed yields depend on the grown varieties, growth environment conditions, and the growing season. In dry climate condition, there is a tendency for higher production than in a period of heavy rain, therefore it is suitable to be developed during water shortage in the dry season [13]. Tsuchihashi and Goto [13] stated that ratoon plants are suitable to be developed during dry season when rain frequency starts to decrease, in these conditions, the roots of the main plants can function optimally.

Correlation coefficient between agronomic characters in ratoon plants can be seen in Table 4. Lines N8-1 and N9-1 showed a correlation between plant height and stalk diameter. Lines N6-3, N7-4, and N19-1 also showed a significant correlation between panicle length and wet panicle weight.

TABLE 4. Correlation coefficients between agronomic characters of sorghum mutant lines.

No.	Lines	MD×MB	MD×MB	PL×PW
1	N1-1	(-)0.04 ns	0.25 ns	0.39 ns
2	N1-4	(-)0.25 ns	0.06 ns	0.31 ns
3	N2-1	0.532 ns	0.10 ns	0.68 ns
4	N2-2	0.15 ns	0.10 ns	0.85 ns
5	N6-2	0.35 ns	0.98 ns	0.39 ns
6	N6-3	0.17 ns	0.24 ns	0.88*
7	N7-4	0.05 ns	0.07 ns	0.89*
8	N8-1	0.97**	(-)0.49 ns	0.55 ns
9	N9-1	0.98**	(-)0.82 ns	0.39 ns
10	N17-1	0.78 ns	0.65 ns	(-)0.62 ns
11	N19-1	(-)0.42 ns	0.31 ns	0.95**
12	N25-2	(-)0.46 ns	(-)0.45 ns	0.49 ns
13	N2-1	0.32 ns	(-)0.39 ns	0.51 ns
14	N5-1	(-)0.82 ns	0.35 ns	0.75 ns
15	N7-3	0.82 ns	0.66 ns	0.08 ns
16	N21-2	0.39 ns	0.54 ns	0.53 ns
17	N17-2	(-)0.07 ns	0.51 ns	0.94**
18	N17-3	0.51 ns	(-)0.53 ns	0.63 ns
19	N17-4	0.56 ns	0.16 ns	(-)0.49 ns
20	N17-5	0.85 ns	(-)0.67 ns	0.92**
21	Numbu (control)	(-)0.82 ns	(-)0.08 ns	0.13 ns

MD = middle stalk diameter, MB = middle stalk brik, PL= panicle length, PW= panicle weight, ns = not significant. *Significant at $P<0.05$. **Significant at $P\leq 0.01$.

CONCLUSION

Based on the variables of plant height and sugar content (brix), mutant lines N17-2, N17-3, and N17-5 can be used as ratoon plants. Line N17-5 showed stalk diameter in primer and ratoon plants larger than the control. The selected lines are expected to be able to provide the needs for carbohydrate raw materials, animal forage or bioethanol in sustainable manner.

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