



Rinaldi Sjahril <rinaldi.sjahril@gmail.com>

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Cc: rinaldi.sjahril@gmail.com

MS ID#: HAYATIJB/2018/001164

MS TITLE: Selection Of Early Maturing And High Yielding Mutants Of Toraja Local Red Rice Grown From M2-M3 Population After Ion Beam Irradiation

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Thank you very much for submitting to the HAYATI Journal of Biosciences.

Sincerely,
Dr. Berry Juliandi

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Thu, Mar 14, 2019 at 5:54 PM

To: rinaldi.sjahril@gmail.com

Cc: rinaldi.sjahril@gmail.com

MS ID#: HAYATIJB/2018/001164

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Dear Rinaldi Sjahril;

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Dear Rinaldi Sjahril;

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Sincerely,
Dr. Berry Juliandi



Rinaldi Sjahril <rinaldi.sjahril@gmail.com>

Jurnal HAYATI

14 messages

hayati_j_biosci@cbn.net.id <hayati_j_biosci@cbn.net.id>
To: rinaldi.sjahril@gmail.com

Wed, Jun 3, 2020 at 3:00 PM

Dear Rinaldi Sjahril, P.hD

Artikel yang berjudul

Selection of early maturing and high yielding mutants of Toraja local red rice grown from M2-M3 population after ion beam irradiation

akan terbit di Jurnal HAYATI Vol. 27 No. 2, 2020. Saat ini artikel tersebut sedang proses diantrian layout, mohon untuk dikirimkan gambar dengan format jpg untuk dijadikan kandidat cover Jurnal HAYATI. Terima kasih.

Salam,
Admin HAYATI J Biosci

Rinaldi Sjahril <rinaldi.sjahril@gmail.com>
To: "hayati_j_biosci@cbn.net.id" <hayati_j_biosci@cbn.net.id>
Bcc: trisna <trisna.ar508@gmail.com>, Irma R Jamaluddin <jamaluddinirma@gmail.com>

Mon, Jun 8, 2020 at 11:42 AM

Terima kasih atas kepercayaannya untk memberi kesempatan kepada kami untuk menjadikan foto-foto penelitian kami sebagai kandidat sampul foto Jurnal HAYATI Vol. 27 No. 2, 2020 yang akan terbit.

Beberapa foto sudah kami lampirkan di bawah ini agar Bapak/Ibu dapat menyeleksi sendiri. Terima kasih.

Wassalam,

Rinaldi Sjahril, Ph.D.

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hayati_j_biosci@cbn.net.id <hayati_j_biosci@cbn.net.id>
To: Rinaldi Sjahril <rinaldi.sjahril@gmail.com>

Fri, Jun 12, 2020 at 3:26 PM

Dear Rinaldi Sjahril, P.hD,

Berikut kami kirimkan proofread artikel Jurnal HAYATI yang berjudul **Selection of early maturing and high yielding mutants of Toraja local red rice grown from M2-M3 population after ion beam irradiation**. Mohon dicek dan diperbaiki sesuai koemntar tersebut. Terima kasih.

Salam,
Admin HAYATI J Biosci

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Rinaldi Sjahril <rinaldi.sjahril@gmail.com>
To: trisna <trisna.ar508@gmail.com>, Yaya <yaya_nurhaya@yahoo.com>

Fri, Jun 12, 2020 at 3:51 PM

Assalaamu'alaikum wrwb.,

Alhamdulillah, sudah ada permintaan Proof Read dari HAYATI. Minta tolong Proof Read ya??? Please??? Terima kasih.

Wassalam,

Rinaldi Sjahril, Ph.D.

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Rinaldi Sjahlil <rinaldi.sjahlil@gmail.com>
To: Andi Muliarni Okasa <andimuliarniokasa10@gmail.com>

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Minta tolong di-Proof Read ya??

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To: Rinaldi Sjahlil <rinaldi.sjahlil@gmail.com>

Tue, Jun 16, 2020 at 4:24 PM

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hayati_j_biosci@cbn.net.id <hayati_j_biosci@cbn.net.id>
To: Rinaldi Sjahlil <rinaldi.sjahlil@gmail.com>

Thu, Jun 18, 2020 at 11:47 AM

Dear Rinaldi Sjahlil, P.hD,

Mengingatkan kembali proofread artikel HAYATI yang dikirimkan pada tgl 12 Juni 2020, berjudul **Selection of early maturing and high yielding mutants of Toraja local red rice grown from M2-M3 population after ion beam irradiation**. Mohon untuk dikirimkan kembali artikel yang sudah dikoreksi sesuai komentar tersebut.

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To: "hayati_j_biosci@cbn.net.id" <hayati_j_biosci@cbn.net.id>

Thu, Jun 18, 2020 at 4:10 PM

Bapak/Ibu Admin Hayati yth.,

Mohon maaf saya baru buka email saya.

InsyaAllah kalau masih bisa diberi waktu kami akan selesaikan malam ini Proof Read yang diminta.

Selain itu kami juga ingin mengetahui apakah Proof Read dari pihak ketiga tetap dibutuhkan? Terima kasih atas pengertian dan bantuannya.

Hormat kami,

Rinaldi Sjahlil, Ph.D.

Laboratory of Plant Bioscience & Reproduction Biotechnology
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Rinaldi Sjahril <rinaldi.sjahril@gmail.com>
To: "hayati_j_biosci@cbn.net.id" <hayati_j_biosci@cbn.net.id>

Fri, Jun 19, 2020 at 8:35 PM

Bapak/Ibu Admin HAYATI yth.,

Terlampir kami kirimkan hasil revisi Proof Read dalam format PDF yang telah kami revisi pada Word file dan kami ubah ke format PDF. Hasil revisi dalam Word file bentuk Track Changes juga telah kami lampirkan.

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Demikian hasil revisi dan Proof Read dari kami. Atas perhatiannya disampaikan banyak terima kasih.

Hormat kami,

Rinaldi Sjahril, Ph.D.

Laboratory of Plant Bioscience & Reproduction Biotechnology
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hayati_j_biosci@cbn.net.id <hayati_j_biosci@cbn.net.id>
To: Rinaldi Sjahril <rinaldi.sjahril@gmail.com>

Mon, Jun 22, 2020 at 2:22 PM

Dear Rinaldi Sjahril, Ph.D,

Terlampir artikel yang sudah kami edit sesuai komentar tersebut. Mohon dicek kembali, masih ada sedikit yang perlu dikoreksi, dan untuk setiap perbaikan diberikan tanda warna kuning agar memudahkan kami saat

mengedit. Terima kasih.

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hayati_j_biosci@cbn.net.id <hayati_j_biosci@cbn.net.id>
To: Rinaldi Sjahril <rinaldi.sjahril@gmail.com>

Mon, Jun 22, 2020 at 2:45 PM

Dear Rinaldi Sjahril, Ph.D,

Terlampir artikel yang sudah kami edit sesuai komentar tersebut. Mohon dicek kembali, masih ada sedikit yang perlu dikoreksi, dan untuk setiap perbaikan tolong diberikan tanda warna kuning agar memudahkan kami saat mengedit. Terima kasih.

Salam,
Admin HAYATI J Biosci
-----Original Message-----

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Sent: Friday, 19 June, 2020 19:35

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Rinaldi Sjahril <rinaldi.sjahril@gmail.com>
To: trisna <trisna.ar508@gmail.com>

Tue, Jun 23, 2020 at 5:40 AM

Rinaldi Sjahril, Ph.D.

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----- Forwarded message -----

From: **hayati_j_biosci@cbn.net.id** <hayati_j_biosci@cbn.net.id>

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[Quoted text hidden]

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hayati_j_biosci@cbn.net.id <hayati_j_biosci@cbn.net.id>
To: Rinaldi Sjahril <rinaldi.sjahril@gmail.com>

Wed, Jun 24, 2020 at 12:08 PM

Dear Rinaldi Sjahril, Ph.D

Mohon dikirimkan kembali artikel yang telah diperbaiki, untuk setiap perbaikan tolong diberikan tanda warna kuning agar memudahkan kami saat mengedit. Terima kasih.

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Rinaldi Sjahril <rinaldi.sjahril@gmail.com>
To: "hayati_j_biosci@cbn.net.id" <hayati_j_biosci@cbn.net.id>

Wed, Jun 24, 2020 at 11:41 PM

Bapak/Ibu Admin HAYATI J Biosci,

Terlampir bersama ini kami kirimkan hasil proof read terakhir yang telah kami lakukan. Mohon maaf karena ternyata banyak sekali perubahan baik pada tabel maupun text. Perubahan tersebut telah kami tandai dan berikan perbaikan untuk di-Replace dan ditambahkan. Selain itu ada beberapa telah kami delete dan ada penambahan 2 pustaka atas nama Sjahril R et al. 2018 dan Riadi M et al. 2019. Conclusion telah kami replace.

Demikian hasil proof read kami dan semoga sudah dapat diterima dengan baik tanpa ada kesalahan lagi atau perbaikan lagi.

Terima kasih kami haturkan atas perhatian dan kerjasamanya.

Wassalam

Hormat kami,


Rinaldi Sjahril, Ph.D.

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Rinaldi Sjahril <rinaldi.sjahril@gmail.com>

Fwd: Re: Invoice artikel HAYATI

1 message

rinaldi.sjahril <rinaldi.sjahril@gmail.com>
To: Irma R Jamaluddin <jamaluddinirma@gmail.com>

Thu, Jul 23, 2020 at 12:45 PM

Sent from my Samsung Galaxy smartphone.

----- Original message -----

From: hayati_j_biosci@cbn.net.id
Date: 07/23/2020 12:23 (GMT+08:00)
To: Rinaldi Sjahril <rinaldi.sjahril@gmail.com>
Subject: Re: Invoice artikel HAYATI

Terima kasih bukti pembayaran telah kami terima, berikut kami kirimkan kwitansi pembayaran artikel.

Salam,

Admin HAYATI J Biosci

-----Original Message-----

From: "Rinaldi Sjahril" <rinaldi.sjahril@gmail.com>
Sent: Tuesday, 21 July, 2020 09:57
To: "hayati_j_biosci@cbn.net.id" <hayati_j_biosci@cbn.net.id>
Subject: Re: Invoice artikel HAYATI

Assalamualaikum wrwb.

Pak Berry dan Admin HAYATI ysh.,

Terima kasih banyak kerjasama dan bantuan atas publikasi artikel kami. Bersama ini kami kirimkan bukti pembayaran proses publikasi artikel tersebut.

Wassalam,

Rinaldi Sjahril, Ph.D.

Laboratory of Plant Bioscience & Reproduction Biotechnology
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On Mon, Jul 20, 2020 at 12:53 PM hayati_j_biosci@cbn.net.id <hayati_j_biosci@cbn.net.id> wrote:

Dear Rinaldi Sjahril, P.hD

Terlampir invoice artikel yang berjudul **Selection of Early Maturing and High Yielding Mutants of Toraja Local Red Rice Grown from M2-M3 Population after Ion Beam Irradiation**, yang terbit di Jurnal HAYATI Vol. 27 No. 2 2020. Terima kasih.

Salam,

Admin HAYATI J Biosci

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Selection Of Early Maturing And High Yielding Mutants Of Toraja Local Red Rice Grown From M2-M3 Population After Ion Beam Irradiation

R. Sjahril¹, A.R. Trisnawaty^{2*}, M. Riadi¹, Rafiuddin¹, T. Sato³, K. Toriyama³, T. Abe⁴

¹ Department of Agronomy, Faculty of Agriculture, Hasanuddin University, Makassar, South Sulawesi-90245, Indonesia

² Department of Agrotechnology, Faculty of Agriculture, Animal Husbandry and Fisheries, Muhammadiyah University Sidenreng Rappang, Sidrap, South Sulawesi-91651, Indonesia

³ Graduate School of Agricultural Science, Tohoku University, Sendai, 980-8572 Japan

⁴ Radiation Biology Team, Nishina Center for Accelerator-Based Science RIKEN, Wako, Saitama 351-0198 Japan

*Corresponding author's e-mail: trisna.ar508@gmail.com

ABSTRACT

This study aims to obtain the genotype of local Toraja red rice M2 mutants that have the potential to be developed into early maturing varieties and have high yields. Research carried out with a mass selection method consisting of two irradiation treatments, namely: 1) irradiation with a Carbon ion dose of 150 Gy (PL-C); 2) irradiation with Argon ion dose of 10 Gy (PL-A) and control as a comparison. Selected strains of M2 seed population were planted in experimental strains plus 2 strains as control, each row had 50 plants. Selection is done in two stages. First stage, the selection is based on the criteria of plant growth components, to select rice plants that have early-middle age characters, dwarf-semi-dwarf, and many tillers. Second stage, selected strains from the scoring method were then grouped based on the degree of similarity with the cluster analysis method. Irradiation with ion beams produces mutants with shorter harvest times than their parents, with the harvest time 16-17 days or 10% shorter compared to their parents. This study produced 10 selected strains, 5 strains of carbon ion and 5 strains of argon ion irradiation treatment, and formed 5 groups with a 90% similarity coefficient.

Keywords: red rice, Toraja local rice, mutants, early maturity, ion beam, irradiation

1. Introduction

Based on data from the North Toraja District Agriculture Service (2013), Tana Toraja is one of the areas in South Sulawesi that has a diversity of local rice germplasm. Based on the results of a survey of local rice in Tana Toraja Regency conducted by Suhardi, et al. (2013) it was found about 29 local Toraja rice varieties, each of which had exotic characteristics such as black, red, aromatic and fluffier rice.

One of the local rice varieties used for generations as part of the tradition and culture of the Toraja community is red rice which in the local language is called Pare Lea. The use of local rice, especially red rice in traditional cultural rituals, helps to preserve this traditional rice variety amid the rapid use of introduced varieties.

Local rice, in general, has several disadvantages including long life, high plant posture and low yield (Wahdah and Langai, 2009). This character is a limiting factor that can cause farmers' interest in growing local rice to be lower, so it is feared that it will cause a lack of availability of local rice cultivars as a source of germplasm.

To optimize the potential of local rice as one of the germplasm sources for plant breeding, the first step that needs to be implemented is to improve the character of the local rice varieties. Character improvement can be done in various ways, both conventionally and mutation induction.

As time goes on, breeding techniques are now being developed using ion beams through the use of heavy ion beams. This ion beam is safer, does not damage the endosperm, because the dosage is low so the level of mutation induction is higher.

Compared to gamma rays, ion beam irradiation has several advantages including higher mutation rates, lower doses with high survival rates, high levels of mutation induction and various other variations, ion files can be focused and regulated through the embryo tissue, and does not damage the endosperm (Yazid and Muryono, 2000). Thus, mutation induction in local rice with radiation irradiation is expected to produce mutant rice that has properties that are better than the original varieties.

This research is expected to be the initial stage to create new red rice varieties early maturing, short postures, and high production. Based on these matters, it is necessary to carry out research "Early maturing Toraja Local Red Rice Mutant Selection from M2 Population from Ion Beam Irradiation".

2. Materials And Methods

2.1. Place and Time

This research was conducted in Alla Subdistrict, Enrekang Regency, South Sulawesi with an altitude of 650 m above sea level at coordinates S: 3° 19' 47.44"; E: 119° 50' 1.57" and lasted from May to October 2017.

2.2. Material

The material used is the M2 generation of local Toraja red rice (Pare Lea) seed produced by ion beam irradiation. To obtain M2 seeds, Toraja local red rice seed (M0) was irradiated using ion beam carbon 150 Gy and ion beam argon 10 Gy, then grown on agricultural land in accordance with the standard of maintenance of rice plants. Individual plants that grew (M1) then produced seeds (M2) which were collected to be used in this study. Other materials used are manure and soil for seeding media, rat poisons, pesticides, irrigation water, and label paper. The tools used are plastic pots for sowing, machetes, hoes, tractors, sprayers, nets, meters, analytical scales, Contador seed counter, cameras, and writing instruments.

2.3. Method

This study consisted of three treatments, namely irradiation with a dose of 150 Gy ion beam (PL-C), irradiation with argon ion dose 10 Gy (PL-A), and control as a comparison. M2 seeds used are seeds produced by M1 plants which produce more than 50 seeds per clump of plants. Based on these criteria there are 61 selected strains, consisting of 46 strains derived from irradiation carbon beam ion treatment and 15 strains of argon ion beam irradiation treatment. M2 seeds from selected strains are planted in rows containing 50 plants each. Thus the total seeds planted in the irradiation carbon beam ion treatment were 2300 seeds, and in the irradiation treatment, the argon beam was 750 seeds. As a control, two local Toraja red rice seeds were planted, each containing 50 plants. Each individual plant is given a line number that shows the row and position of the plant in the row. The total number of individual plants grown was 1303 plants in the carbon beam ion irradiation treatment, 108 in the argon ion beam irradiation treatment and 89 plants in the control treatment.

2.4. Data Analysis

Observation data in the form of plant height, harvest age and number of productive tillers or panicle numbers are grouped into 3 criteria classes based on the Rice Plant Characterization and Evaluation System Guide (BPPP, 2003), namely:

1. Parameters of plant height: dwarf (<110 cm), semi-dwarf (110-130 cm) and high (> 130 cm).
2. Harvest age parameters: early (<120 days), middle (120-150 days) and deep (> 150 days).
3. Parameters for the number of productive tillers: few (<8 panicles), middle (9-16 panicles) and many (≥17 panicles).

Furthermore, the selection activity was carried out to select rice plants that had early-maturity characters, short-medium canopy, and many tillers. Mutant frequency is calculated based on the number of mutants that have a certain character and the overall mutant plant population with the formula:

$$\text{Mutant Frequency (MF)} = \frac{\text{Number of mutant plants}}{\text{Number of population}} \times 100 \% \dots \dots \dots (1)$$

For the selection of strains based on the component production parameters, the scoring method is used. Each observation parameter from the production component will be given a score, and then summed to obtain the level of association. The final result of the scoring system is the classification of the level of relevance of the output parameters calculated by the equation:

$$S_n = (X_n - X_1) / SD + S_1 \dots \dots \dots (2)$$

Where :

S_n = data nth score

X_n = Value of the nth data

SD = Standard Deviation

X_1 = Lowest data value

S_1 = Lowest data score

3. Results

3.1. Number and Frequency of Mutants

Data on measuring plant height, harvest age and number of productive tillers are classified into three grouping criteria. The recapitulation of the number of individuals and the frequency of mutants based on the grouping criteria is presented in Table 1.

Table 1. Recapitulation of the number of individuals and mutant frequencies based on plant height, harvest age and the number of productive tillers.

Character	Criteria	Treatments*					
		PL-Control		PL-Carbon		PL-Argon	
		Number of Plants (No.)	Frequency (%)	Number of Plants (No.)	Frequency (%)	Number of Plants (No.)	Frequency (%)
Plant Height	Dwarf (<110 cm)	0	0.0	9	0.7	1	0.9
	Semi-dwarf (110-130 cm)	0	0.0	23	1.8	0	0.0
	High (>130 cm).	89	100.0	1271	97.5	107	99.1
Harvest Date	Early (<120 days)	0	0.0	0	0.0	0	0.0
	Middle (120-150 days)	0	0.0	113	8.7	23	21.3
	Deep (>150 days).	89	100.0	1190	91.3	85	78.7
Number of Panicle per Plant	Few (≤8 panicles)	27	30.3	340	26.1	3	2.8
	Middle (9-16 panicles)	25	28.1	579	44.4	7	6.5
	Many (≥17 panicles).	37	41.6	384	29.5	98	90.7
Total Number of Plants		89		1303		108	

* PL-Control: M2 plants without irradiation, PL-Carbon: M2 plants with carbon-ion irradiation, PL-Argon: M2 plants with argon-ion irradiation.

The data in Table I shows irradiation treatment, both by using carbon ions and argon ions to improve plant height character, by producing mutants that have dwarf and semi-dwarf plant postures, even in low frequencies. The irradiation treatment of carbon ions produced 9 plants mutant that had the character of dwarf plant postures with a frequency of 0.7% and 23 plants mutant of semi-dwarf plant postures with a frequency of 1.8%. While the argon ion irradiation treatment only produces 1 plant that has the character of dwarf plant postures with a frequency of 0.9%. In the argon ion, irradiation treatment was not obtained with semi-dwarf plant posture. In the control treatment, all plant populations have high postures.

The ion beam irradiation treatment also succeeded in producing M2 mutants with an early harvest age than the control treatment. The carbon ion irradiation treatment produced 113 plants of mutants with middle maturing (120-150 days) with a mutant frequency of 8.7%. Meanwhile, argon ion irradiation produces 23 plants middle maturing mutant (120-150 days) with a frequency of 21.3%. While in the control treatment, all plant populations were in the deep maturing (>150 days).

For the character of a large number of productive tillers (> 17), the treatment of irradiation with a carbon beam ion produced the highest number of clumps of 384 plants with a frequency of 29.5%, while the treatment of argon ion irradiation reached 98 plants with a frequency of 90.7%. In the control treatment, the number of clumps with many productive tillers characters reached 37 plants with a frequency of 41.6%.

3.2. Selection Strain Based on Growth Components

Based on the grouping of growth components that have been done previously, namely in terms of plant height, harvest age and number of productive tillers, a combination of the three criteria is then carried out. From the results of the third combination, these components, 27 types of criteria were obtained from each treatment and the number of strains included in the criteria. The combination results obtained are presented in Table 2.

Table 2. Recapitulation of population numbers by a combination of criteria of harvest age, plant height and number of productive tillers.

Criteria			Treatments		
Harvest maturing	Plant height	Productive tillers	PL-Control	PL-Carbon	PL-Argon
Early	Dwarf	Few	0	0	0
Early	Dwarf	Middle	0	0	0
Early	Dwarf	Many	0	0	0
Early	Semi-Dwarf	Few	0	0	0
Early	Semi-Dwarf	Middle	0	0	0
Early	Semi-Dwarf	Many	0	0	0
Early	High	Few	0	0	0
Early	High	Middle	0	0	0
Early	High	Many	0	0	0
Middle	Dwarf	Few	0	0	0
Middle	Dwarf	Middle	0	0	0
Middle	Dwarf	Many	0	0	0
Middle	Semi-Dwarf	Few	0	1	0
Middle	Semi-Dwarf	Middle	0	0	0
Middle	Semi-Dwarf	Many	0	0	0
Middle	High	Few	0	31	1
Middle	High	Middle	0	53	1
Middle*	High*	Many*	0	27	21
Deep	Dwarf	Few	0	8	0
Deep	Dwarf	Middle	0	1	0
Deep	Dwarf	Many	0	0	1
Deep	Semi-Dwarf	Few	0	14	0
Deep	Semi-Dwarf	Middle	0	7	0
Deep	Semi-Dwarf	Many	0	0	0
Deep	High	Few	31	360	2
Deep	High	Middle	21	445	6
Deep	High	Many	37	356	76
Total number of plants			89	1303	108

Information: * Criteria of the selected strain

Table 2 shows that there are no strains that have criteria for early maturing, dwarf postures and many tillers so that the criteria used to select are criteria for middle maturing, high posture, and many tillers. Based on these criteria, 27 plants were selected in the irradiation treatment of carbon beam ions and 21 plants in the ion beam argon irradiation treatment, a total of 48 plants were selected.

3.3. Selection Strain Based on Yield Components

Based on the table of recapitulation of the number of selected strains in terms of growth components (plant height, the age of harvest and productive tillers) in Table 2, the production component data is then calculated. From the results of these calculations, the strains are grouped according to the criteria of production components that have been predetermined. The results of the calculation and grouping are then scored in the highest order. The results of the selection and scoring of selected strains based on production component data can be seen in Table 3.

Table 3. Selection and scoring results of selected strains based on growth and yield components data on carbon and argon ion irradiation treatments.

No.	Nomor Galur	PH	HD	NP	LP	NSP	WSP	DS	WFP	W100	PBBM	PF	Total score
1	PL-C-1.7-20	4.48	2.22	2.01	1.68	1.59	1.89	1.91	2.55	6.03	4.01	2.41	30.78
2	PL-C-1.7-36	4.23	2.82	1.80	1.80	2.08	2.57	2.58	3.27	6.24	4.77	2.87	35.02
3	PL-C-2.2-29	3.80	1.00	2.11	4.58	2.69	2.34	2.04	2.82	4.59	3.89	2.76	32.62
4	PL-C-3.12-10	2.95	2.22	1.60	2.46	1.95	1.36	2.05	1.78	5.41	2.31	1.58	25.66
5	PL-C-3.12-25	5.08	2.22	1.60	2.10	1.25	1.29	1.20	1.84	4.80	3.24	1.56	26.17
6	PL-C-3.12-39	3.55	2.22	1.60	1.58	2.87	2.77	3.94	3.27	4.70	4.15	3.08	33.73
7	PL-C-4.1-17*	4.23	2.22	1.50	3.23	3.39	4.18	3.57	4.86	5.79	5.40	3.61	41.97
8	PL-C-9.1-8	4.23	2.22	1.80	1.77	2.43	2.69	3.13	3.07	6.56	3.78	2.34	34.01
9	PL-C-10.16-14	3.80	1.00	1.30	2.47	3.22	3.35	3.82	3.89	4.78	4.58	2.75	34.98
10	PL-C-17.9-11	3.12	1.00	1.00	3.86	4.06	3.65	4.04	4.23	3.52	4.86	2.61	35.95
11	PL-C-17.13-2	2.95	1.00	2.31	1.17	1.55	2.07	2.12	2.87	5.85	4.87	2.66	29.40
12	PL-C-17.13-4	3.46	1.00	1.00	1.75	1.75	2.06	2.12	2.59	5.94	3.63	1.57	26.88
13	PL-C-17.13-21	4.23	2.22	1.30	1.17	1.63	1.80	2.25	2.23	5.23	3.35	1.75	27.15
14	PL-C-17.13-43	2.53	1.00	1.40	1.16	1.58	1.87	2.18	2.53	5.38	4.21	1.73	25.58
15	PL-C-18.2-1	3.55	1.00	2.01	3.16	3.62	2.15	3.92	2.49	3.45	1.88	2.35	29.58
16	PL-C-18.2-20	1.68	1.00	1.20	3.06	3.08	2.80	3.26	3.25	5.75	3.73	2.28	31.09
17	PL-C-18.8-22	3.97	1.00	1.00	2.00	2.13	2.47	2.55	2.99	5.76	3.94	1.97	29.78
18	PL-C-18.8-40	4.23	1.00	2.01	1.52	1.62	2.11	2.04	2.77	5.12	4.64	2.36	29.41
19	PL-C-19.4-21	1.00	1.00	1.00	4.35	3.80	2.85	3.45	3.08	4.68	3.09	2.02	30.33
20	PL-C-19.10-43	4.48	2.82	2.61	1.02	1.71	2.22	2.46	2.93	5.48	4.64	3.17	33.55
21	PL-C-20.1-1*	3.89	1.00	1.80	3.50	3.50	4.17	3.55	5.02	5.55	5.61	4.14	41.74
22	PL-C-20.5-44*	2.78	1.00	1.00	3.42	4.01	4.44	4.25	5.10	6.01	4.82	3.06	39.88
23	PL-C-20.5-49*	3.80	1.00	1.20	3.34	2.88	3.99	2.85	4.96	5.98	6.04	3.27	39.32
24	PL-C-20.6-6	2.02	1.00	1.40	1.36	1.51	2.26	1.95	3.08	5.72	5.28	2.34	27.92
25	PL-C-20.6-7	1.85	1.00	1.20	1.48	1.61	2.48	2.04	3.29	7.11	5.16	2.22	29.44
26	PL-C-20.6-50	5.08	1.00	1.70	1.14	1.73	2.35	2.44	3.09	5.64	4.83	2.60	31.60
27	PL-C-20.11-44*	2.10	1.00	1.20	3.90	4.83	4.59	4.96	4.92	5.37	4.11	3.24	40.23
1	PL-A-1.1-8	2.36	1.00	1.30	2.75	1.40	1.15	1.15	1.60	3.59	2.59	1.36	20.25
2	PL-A-1.1-12	3.21	1.00	1.60	3.45	2.83	1.00	2.73	1.00	1.00	1.00	1.00	19.82
3	PL-A-1.1-20	2.10	1.00	1.60	1.69	1.00	1.44	1.00	2.24	5.23	4.83	1.86	23.99
4	PL-A-1.1-21	1.59	1.00	2.01	2.52	1.50	1.87	1.39	2.62	4.92	4.79	2.45	26.67
5	PL-A-1.1-22	2.10	1.00	1.60	1.72	1.02	1.15	1.01	1.93	5.36	4.58	1.57	23.04
6	PL-A-1.1-24	2.02	1.00	2.41	2.97	2.32	2.61	2.30	3.29	5.02	4.63	3.27	31.83
7	PL-A-1.1-35	4.14	1.00	1.91	2.44	1.85	1.70	1.91	2.21	4.30	3.42	2.07	26.95
8	PL-A-1.1-41	3.21	1.00	3.21	2.83	1.77	1.53	1.62	2.06	4.15	3.42	2.45	27.27
9	PL-A-1.1-43	4.14	1.00	2.81	2.53	1.60	1.53	1.52	2.19	4.39	3.90	2.43	28.05
10	PL-A-1.1-45	3.97	1.00	2.41	1.47	1.08	1.47	1.22	2.30	4.72	5.04	2.42	27.09
11	PL-A-1.1-47	3.97	1.00	1.70	2.62	2.35	3.04	2.52	3.76	6.27	4.89	3.08	35.20
12	PL-A-1.1-48	4.40	1.00	1.80	1.54	1.05	1.69	1.14	2.53	5.29	5.30	2.25	28.00
13	PL-A-6.5-1	4.14	1.00	1.60	2.95	2.59	2.30	2.67	2.41	5.79	2.59	2.04	30.09
14	PL-A-6.5-4*	3.21	1.00	2.61	3.86	4.32	4.62	4.36	4.74	6.45	3.99	4.98	44.13
15	PL-A-6.5-6*	3.29	1.00	2.61	3.79	3.09	3.81	2.88	4.42	6.00	5.04	4.66	40.58
16	PL-A-6.5-15*	2.70	1.00	3.82	3.82	3.20	3.29	3.01	3.68	5.87	3.87	5.12	39.36
17	PL-A-6.5-21	2.02	1.00	3.61	4.33	3.30	3.25	2.87	3.37	5.96	3.26	4.50	37.47
18	PL-A-6.5-23*	1.93	7.08	3.41	3.37	2.80	3.11	2.73	3.55	5.70	4.05	4.55	42.28
19	PL-A-6.5-29*	3.21	1.00	2.81	3.73	4.22	4.17	4.32	4.43	5.74	3.92	4.55	42.09
20	PL-A-7.2-29*	4.40	1.00	2.01	2.49	1.59	1.79	1.52	2.48	4.77	4.27	1.71	28.03
21	PL-A-7.2-35	3.55	1.00	6.83	2.67	1.94	1.63	1.92	2.06	4.83	3.11	3.65	33.19
Average		149,38	26.81	26.18	156.65	2.67	5.93	2.21	2.47	59.52	56.46	31.53	31.53
Min Value		140	17	22.06	99.64	1.31	4.18	0.49	1.56	22.38	9.80	16.00	16.00
Max Value		150	75	31.50	256.37	4.55	8.64	3.88	2.87	82.58	121.93	44.13	44.13
Standard Deviation		1,65	9.94	2.65	40.92	0.90	1.12	0.83	0.21	11.94	27.24	6.44	6.44
The best Value Limit		151,02	36.76	28.83	197.57	3.56	7.05	3.03	2.68	71.45	83.70	37.97	37.97

Information:

PH = Plant Height (cm)
 HD = Harvest Date (days)
 NP = Number of Panicle (No.)
 LP = Length of Panicle (cm)
 NSP = Number of Seed per Panicle (No.)

WSP = Weight of Seed per Panicle (g)
 DS = Density of Seed (no./cm)
 WFP = Weight of Fertile seed per Panicle (g)
 W100 = Weight of 100 seeds (g)
 PF = Percentage of Fertile seed (%)
 *= Selected strains

From the results of the scoring that has been done, strains are selected which have a total score above the best value limit of 37.97. The best value limit is obtained from the average value of the total score plus the standard deviation value. There were 10 strains that had a score above the best value, each of the 5 strains of the carbon ion irradiation treatment and 5 strains of the argon ion irradiation treatment for selection criteria, middle maturing, high posture, and many productive tillers. The ten strains are then grouped according to the level of similarity with the cluster analysis method. As for the level of similarity of these strains based on the production components are presented in Figure 1.

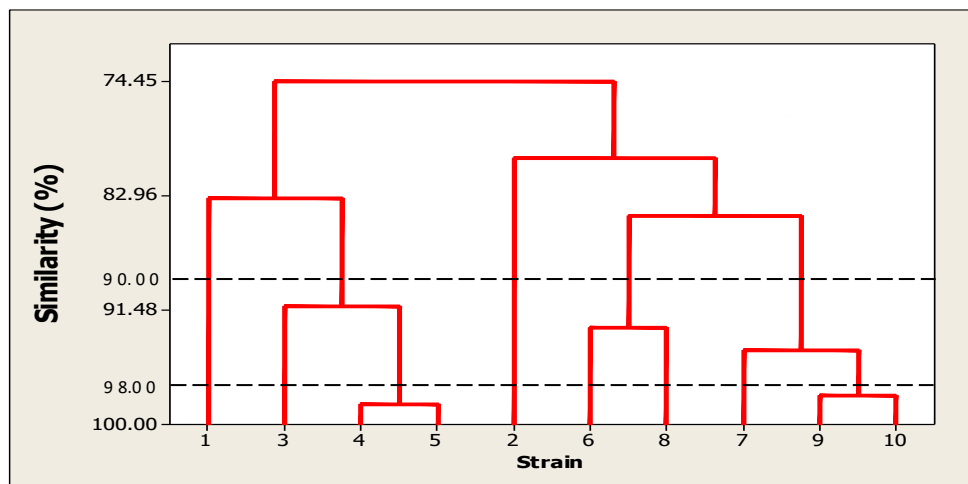


Figure 1. Dendrogram grouping of 10 selected strains of production components.

Figure 1 shows that based on the dendrogram of 10 Toraja local red rice strains selected at the 90% similarity coefficient to form 5 Groups, namely Group I consists of 1 strain, Group II consists of 2 Subgroups (3 strains), Group III consists of 1 strain, Group IV consists of 2 strains and Group V consists of 2 subgroups (3 strains). The group analysis of the production components in the two planting locations based on the dendrogram graph is presented in Table 4.

Table 4. Grouping of yield components data 10 strains selected at the planting location with a similarity rate of 90%.

Yield Components	Number of Strain
Group I	1
Group II	3, 4, 5
Group III	2
Group IV	6, 8
Group V	7, 9, 10

Information: 1= PL-C-4.1-17; 2= PL-C-20.1-1; 3= PL-C-20.5-44; 4= PL-C-20.5-49; 5= PL-C-20.11-44; 6= PL-A-6.5-4; 7= PL-A-6.5-6; 8= PL-A-6.5-15; 9= PL-A-6.5-23; 10= PL-A-6.5-29

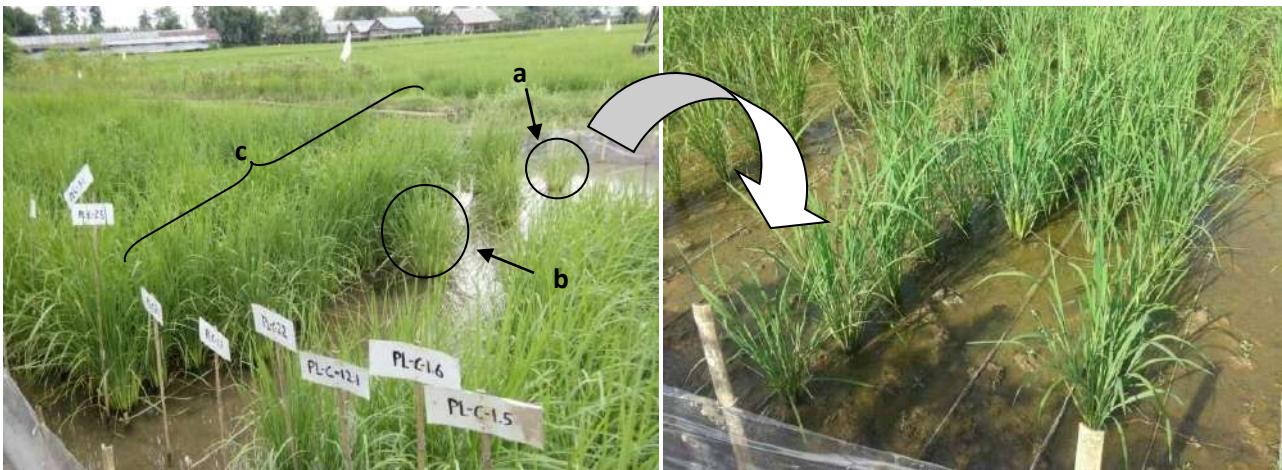


Figure 2. Variations of rice plant phenotypes at age (a. Dwarf-Type; b. Semi-Dwarf Type; c. Normal Type).

4. DISCUSSION

The results of this study indicate that irradiation of ion beams using ion beam carbon and argon can be used to improve the character of local Toraja red rice plants, particularly related to the characteristics of harvest age, posture, and crop production. The control plants were harvested 162 - 166 days after seedling, while the mutant were harvested in the range of 146 - 161 days after seedling. Irradiation with ion beams speeds up the harvest time for 16 days or about 10% of the age of its parents. The use of carbon ion irradiation to produce plants with faster harvest time was also reported by Dong and Li (2012) in sweet sorghum plants and Du et al (2014) in *Arabidopsis thaliana*.

In the areas of plant origin, namely in North Toraja Regency, the age of harvesting of red rice (Lea Pare) is included in the deepest age category of 180 days after seedling (Sahardi et al., 2013). The length of harvest time causes the low interest of the community to cultivate this type of local rice. Lea Pare cultivation is very limited, only to meet the needs of the use of these types of rice in traditional rituals. The presence of the Toraja local red rice variety with early maturing character is expected to increase public interest in cultivating this type of rice, and prevent the extinction of this species. The production of local red rice mutants with early maturity character can be the first step to produce early maturing varieties. In this study, ion beam irradiation using carbon ions produced 9 mutants with short postures. Unfortunately, these short characters are not present along with a short harvest age character and high production yield in one individual mutant, so they are not included in the selected strain group. Rice mutants with shorter postures due to ion beam irradiation were reported by Ling et al (2013) in rice cultivars MR219. According to Ling et al (2013), the increase in Carbon ion dosage is directly proportional to the inhibition of plant height growth.

Overall, in this study, ion beam irradiation produced 10 strains with shorter harvest ages and a high number of productive tillers. The character of the number of productive tillers is expected to be positively correlated with rice grain production. The use of ion beam irradiation to increase crop yields has been reported in various studies, including by Zhao et al (2006) and Xie et al (2008).

The results of the analysis and grouping of criteria on the three observation parameters, namely plant height, age of harvest and productive tillers at the planting location (Table 1) showed that carbon irradiation treatment produced higher mutant amounts compared to argon ion irradiation

treatments, but in terms of frequency, Argon ions irradiation treatment produce higher mutant frequencies.

This shows that carbon ion irradiation treatment is better at producing mutants mainly in terms of plant height criteria, harvest age and panicle number. Mutations that occur may be a mutagenic effect of ion beam irradiation which indicates that the treatment is quite effective in creating genetic diversity in population M2. High frequency of mutations caused by irradiation of carbon ions is caused by the energy fired by carbon ions greater than is 135 MeV / u compared to the energy in Argon ions which is only 95 MeV / u (Y. Hayashi, et al., 2007). This allows the amount of damage to the gene caused by the release of energy from the ions that are fired so that the diversity (mutants) generated is also increasing. Other advantages of ion beam mutagenesis include high survival rates, high levels of mutation induction and various other variations (Yazid and Muryono, 2000).

From a total of 48 strains (Table 2), after the housing was done against the score obtained and compared with the best total score (above 37.97), 10 strains were selected with the total score above the best score (Table 3), namely 5 strains in carbon treatment (PL-C-4.1-17, PL-C-20.1-1, PL-C-20.5-44, PL-C-20.5-49 and PL-C-20.11-44) and 5 in the treatment argon (PL-A-6.5-4, PL-A-6.5-6, PL-A-6.5-15, PL-A-6.5-23 and PL-A-6.5-29).

The selection of character selection in the tested population is based on components that have a close relationship with the character of production. Thus, the selection of populations of diploid rice strains can use the character of the generative phase of plant height, a number of pithy grain per panicle, total grain per panicle, weight of 1,000 rice grain and character of grain yield per clump. Aryana et al. (2011) explained the character of number of tillers, number of rice grain, total grain count per panicle, and weight of 1,000 grains can be used as character selection, whereas in Safitri et al. (2011) explained the character of the number of productive tillers, number of rice grain per panicle, and the weight of 1,000 grains can be used as character selection.

In Figure 1 and Table 1 above, it can be seen that at the 90% similarity level, based on the data of the production components of the 10 selected strains, 5 groups were obtained, namely Group I consisting of 1 strain, namely PL-C-4.1-17. Group II consisted of 2 subgroups consisting of 3 strains namely PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44. Group III consisted of 1 strain, namely PL-C-20.1-1. Group IV consists of 2 strains, namely PL-A-6.5-4 and PL-A-6.5-15. Group V consists of 2 subgroups consisting of 3 strains, namely PL-A-6.5-6, PL-A-6.5-23, and PL-A-6.5-29.

Based on the level of similarity it can be seen that the PL-C-20.5-49 and PL-C-20.11-44 and PL-A-6.5-23 and PL-A-6.5-29 strains have very close kinship relationships. The line has a 95% similarity level. This shows that strains that are closely related will have a close genetic distance. This is consistent with Lee (1998) 's opinion that closely related individuals will have a close genetic distance, whereas if they are distant relatives they will have a far genetic distance. The results of the grouping of 10 selected strains consisting of 5 groups showed that the strains in the same group generally had almost the same number of strains. Erlina (2011) suggests that the same cluster is characterized by almost the same genotype name. This shows that the genotypes are formed from the same population so that the level of kinship is closer.

5. CONCLUSIONS

The results of this study showed treatment with ion beam irradiation either by using carbon ions as well as argon ions are capable of producing Toraja local red rice mutant genotype with a shorter harvest age compared to the parental. There is an acceleration of age harvest 16-17 days or about 10% from the harvesting age of parental. Out of 48 total strains, 10 strains were

selected, 5 strains of carbon ion irradiation treatment (PL-C-4.1-17, PL-C-20.1-1, PL-C-20.5-44, PL-C-20.5-49 and PL-C-20.11-44) and 5 strains of argon ion irradiation treatment (PL-A-6.5-4, PL-A-6.5-6, PL-A-6.5-15, PL-A-6.5-23 and PL-A-6.5-29) for selection criteria of middle maturity, high posture and many productive tillers.

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Selection of Early Maturing and High Yielding Mutants of Toraja Local Red Rice Grown from M2-M3 Population after Ion Beam Irradiation

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ABSTRACT

This study aims to obtain the genotype of local Toraja red rice M2 mutants that have the potential to be developed into early maturing varieties and have high yields. Research carried out with a mass selection method consisting of two irradiation treatments, namely: (1) irradiation with a Carbon ion dose of 150 Gy (PL-C); (2) irradiation with Argon ion dose of 10 Gy (PL-A) and control as a comparison. Selected lines of M2 seed population were planted in experimental rows plus 2 rows as control, each row had 50 plants. Selection is done in two stages. The first stage, the selection was based on the criteria of plant growth components; to select rice plants that have early to middle maturity characters, dwarf to semi-dwarf, and larger number of tillers. The second stage, selected lines from the scoring method were then grouped based on the degree of similarity with the cluster analysis method. Irradiation with ion beams produced mutants with shorter harvest times than their parents, with the harvest time 16-17 days or 10% shorter compared to their parents. This study produced 10 selected lines, 5 lines of Carbon ion and 5 lines of Argon ion irradiation treatment, and formed 5 groups with a 90% similarity coefficient.

1. Introduction

Based on data from the North Toraja Regency Agriculture Service (2013), Tana Toraja is one of the areas in South Sulawesi that has a diversity of local rice germplasm. Based on the results of a Survey of local rice in Tana Toraja Regency conducted by Sahardi and Djufry (2013) it was found about 29 local Toraja rice varieties, each of which had exotic characteristics such as black, red, aromatic, and fluffier rice found in that

One of the local rice varieties used for generations as part of the tradition and culture of the Toraja community is red rice which in the local language is called Pare Lea. The use of local rice, especially red rice in traditional cultural rituals, helps to preserve this traditional rice variety amid the rapid use of introduced varieties.

Local rice, in general, has several disadvantages

including long life, high plant posture and low yield (Wahdah *et al.* 2012). This character is a limiting factor that can cause farmers' interest in growing local rice to be lower, so it is feared that it will cause a lack of availability of local rice cultivars as a source of germplasm.

To optimize the potential of local rice as one of the germplasm sources for plant breeding, the first step that needs to be implemented is to improve the character of the local rice varieties. Character improvement can be done in various ways, both conventionally and mutation induction. As time goes on, breeding techniques are now being developed using ion beams through the use of heavy-ion beams. This ion beam irradiation is safer, does not damage the endosperm, because the dosage is low so the level of mutation induction is higher.

Compared to gamma rays, ion beam irradiation has several advantages including higher mutation rates, lower doses with high survival rates, high levels of mutation induction and various other variations, ion

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files can be focused and regulated through the embryo tissue, and does not damage the endosperm (Yazid and Muryono 2000; Abe *et al.* 2012). Thus, mutation induction in local rice with radiation irradiation is expected to produce mutant rice that has properties that are better than the original varieties.

This research is expected to be the initial stage to create new red rice varieties early maturing, short postures, and high production. Based on these matters, it is necessary to carry out research "Early maturing Toraja Local Red Rice Mutant Selection from M2 Population from Ion Beam Irradiation".

2. Materials and Methods

2.1. Place and Time

This research was conducted in Alla Subdistrict, Enrekang Regency, South Sulawesi with an altitude of 650 m above sea level at coordinates S: 3° 19' 47.44"; E: 119° 50' 1.57" and lasted from May to October 2017.

2.2. Materials

The material used is the M2 generation of local Toraja red rice (Pare Lea) seed produced by ion beam irradiation. To obtain M2 seeds, Toraja local red rice seed (M0) was irradiated using ion beam Carbon 150 Gy and ion beam Argon 10 Gy, then grown on agricultural land in accordance with the standard of maintenance of rice plants. Individual plants that grew (M1) plants grew then produced seeds (M2) which were collected to be used in this study. Other materials used are manure and soil for seeding media, rat poisons, pesticides, irrigation water, and paper label paper. The tools used are plastic pots for sowing, machetes, hoes, tractors, sprayers, nets, meters, analytical scales, Contador seed counter, cameras, and writing instruments.

2.3. Methods

This study consisted of three treatments, namely irradiation with a dose of 150 Gy Carbon ion beam (PL-C), irradiation with Argon ion beam dose 10 Gy (PL-A), and control as a comparison. M2 seeds used are seeds produced by M1 plants which produce more than 50 seeds per clump of plants. Based on these criteria there are 61 selected strains consisting of 46 strains derived from irradiation Carbon ion beam ion treatment and 15 strains of Argon ion beam irradiation treatment. M2 seeds from selected strains are planted in rows containing 50 plants each. Thus the total seeds planted in the Carbon ion beam irradiation carbon beam ion treatment were 2,300 seeds, and in the irradiation treatment, the argon beam

was 750 seeds. As a control, two local Toraja red rice seeds were planted in rows, each containing 50 plants. Each individual plant is given a line number that shows the row and position of the plants in the rows. The total number of individual plants grown was 1,303 plants in the Carbon ion beam ion irradiation treatment, 108 in the Argon ion beam irradiation treatment and 89 plants in the control treatment.

2.4. Data Analysis

Observation data in the form of plant height, harvest age and number of productive tillers or panicle numbers are grouped into 3 criteria classes based on the Rice Plant Characterization and Evaluation System Guide (BPPP 2003), namely:

1. Parameters of plant height: dwarf (<110 cm), semi-dwarf (110-130 cm), and high (>130 cm).
2. Harvesting age parameters: early (<120 days), middle (120-150 days), and deep (>150 days)...
3. Parameters for the number of productive tillers: few (<8 panicles), middle (9-16 panicles), and many (≥17 panicles).

Furthermore, the selection activity was carried out to select rice plants that had early-maturity characters, short-medium canopy, and many tillers. Mutant frequency is calculated based on the number of mutants that have a certain character and the overall mutant plant population with the formula:

$$\text{Mutant frequency (MF)} = \frac{\text{Number of mutant plants}}{\text{Number of population}} \times 100\% \quad (1)$$

For the selection of strains based on the component production parameters, the scoring method is used. Each observation parameter from the production component will be given a score, and then summed to obtain the level of association. The final result of the scoring system is the classification of the level of relevance of the output parameters calculated by the equation:

$$S_n = (X_n - X_1) / SD + S_1 \quad (2)$$

Informations:

- S_n : data nth score
- SD : standard Deviation
- S_1 : lowest data score
- X_n : value of the nth data
- X_1 : lowest data value

3. Results

3.1. Number and Frequency of Mutants

Data on measuring plant height, harvest age and number of productive tillers are classified into three grouping criteria. The recapitulation of the number of individuals and the frequency of mutants based on the grouping criteria is presented in Table 1.

The data in Table 1 shows irradiation treatment, both by using Carbon ions and Argon ions to improve plant height character, by producing mutants that have dwarf and semi-dwarf plant postures, even in low frequencies. The irradiation treatment of Carbon ions produced 9 plants mutant that had the character of dwarf plant postures with a frequency of 0.7% and 23 plants mutant of semi-dwarf plant postures with a frequency of 1.8%. While the Argon-ion irradiation treatment only produces 1 plant that has the character of dwarf plant postures with a frequency of 0.9%. In the Argon-ion irradiation treatment there was not obtained with semi-dwarf plant posture. In the control treatment, all plant populations have high postures.

The ion beam irradiation treatment also succeeded in producing M2 mutants with an early

harvest age than the control treatment. The carbon ion irradiation treatment produced 113 plants of mutants with middle maturing (120-150 days) with a mutant frequency of 8.7%. Meanwhile, argon ion irradiation produces 23 plants middle maturing mutant (120-150 days) with a frequency of 21.3%. While in the control treatment, all plant populations were in the deep maturing (>150 days).

For the character of a large number of productive tillers (>17), the treatment of irradiation with a carbon beam ion produced the highest number of clumps of 384 plants with a frequency of 29.5%, while the treatment of argon ion irradiation reached 98 plants with a frequency of 90.7%. In the control treatment, the number of clumps with many productive tillers characters reached 37 plants with a frequency of 41.6%.

3.2. Selections Strain Based on Growth Components

Based on the grouping of growth components that have been done previously, namely in terms of plant height, harvest age and number of productive tillers, a combination of the three criteria is then carried out. From the results of the third combination, these

Table 1. Recapitulation of the number of individuals and mutant frequencies based on plant height, harvest age and the number of productive tillers

Character	Criteria	Treatments*					
		PL-control		PL-carbon		PL-Argon	
		Number of plants (no.)	Frequency (%)	Number of plants (no.)	Frequency (%)	Number of plants (no.)	Frequency (%)
Plant height	Dwarf (<110 cm)	0	0.0	9	0.7	1	0.9
	Semi-dwarf (110-130 cm)	0	0.0	23	1.8	0	0.0
	High (>130 cm)	89	100.0	1,271	97.5	107	99.1
Harvest date	Early (<120 days)	0	0.0	0	0.0	0	0.0
	Middle (120-150 days)	0	0.0	113	8.7	23	21.3
	Deep (>150 days)	89	100.0	1,190	91.3	85	78.7
Number of panicle per plant	Early (<120 days)	27	30.3	340	26.1	3	2.8
	Middle (120-150 days)	25	28.1	579	44.4	7	6.5
	Deep (>150 days)	37	41.6	384	29.5	98	90.7
Total number of plants		89		1,303		108	

*PL-control: M2 plants without irradiation, PL-carbon: M2 plants with carbon ion irradiation, PL-argon: M2 plants with argon ion irradiation

components, 27 types of criteria were obtained from each treatment and the number of strains included in the criteria. The combination results obtained are presented in Table 2.

Table 2 shows that there are no strains that have criteria for early maturing, dwarf postures and many tillers, so that the criteria used to select are criteria for middle maturing, high posture, and many tillers. Based on these criteria, 27 plants were selected in the irradiation treatment of carbon beam ions and 21 plants in the ion beam argon irradiation treatment, a total of 48 plants were selected.

3.3. Selection Strain Based on Yield Components

Based on the table of recapitulation of the number of selected strains in terms of growth components

(plant height, the age of harvest, and productive tillers) in Table 2, the production component data is then calculated. From the results of these calculations, the strains are grouped according to the criteria of production components that have been predetermined. The results of the calculation and grouping are then scored in the highest order. The results of the selection and scoring of selected strains based on production component data can be seen in Table 3.

From the results of the scoring that has been done, strains are selected which have a total score above the best value limit of 37.97. The best value limit is obtained from the average value of the total score plus the standard deviation value. There were 10 strains that had a score above the best value, each of the 5 strains of the carbon ion irradiation treatment

Table 2. Recapitulation of population numbers by a combination of criteria of harvest age, plant height, and number of productive tillers

Harvest maturing	Criteria		Treatments		
	Plant height	Productive tillers	PL-control	PL-carbon	PL-argon
Early	Dwarf	Few	0	0	0
Early	Dwarf	Middle	0	0	0
Early	Dwarf	Many	0	0	0
Early	Semi-Dwarf	Few	0	0	0
Early	Semi-Dwarf	Middle	0	0	0
Early	Semi-Dwarf	Many	0	0	0
Early	High	Few	0	0	0
Early	High	Middle	0	0	0
Early	High	Many	0	0	0
Middle	Dwarf	Few	0	0	0
Middle	Dwarf	Middle	0	0	0
Middle	Dwarf	Many	0	0	0
Middle	Semi-Dwarf	Few	0	1	0
Middle	Semi-Dwarf	Middle	0	0	0
Middle	Semi-Dwarf	Many	0	0	0
Middle	High	Few	0	31	1
Middle	High	Middle	0	53	1
Middle*	High*	Many*	0	27	21
Deep	Dwarf	Few	0	8	0
Deep	Dwarf	Middle	0	1	0
Deep	Dwarf	Many	0	0	1
Deep	Semi-Dwarf	Few	0	14	0
Deep	Semi-Dwarf	Middle	0	7	0
Deep	Semi-Dwarf	Many	0	0	0
Deep	High	Few	31	360	2
Deep	High	Middle	21	445	6
Deep	High	Many	37	356	76
Total number of plants			89	1,303	108

*Criteria of the selected strain

Table 3. Selection and scoring results of selected strains based on growth and yield components data on carbon and argon ion irradiation treatments

Nomor galur	PH	HD	NP	LP	NSP	WSP	DS	WFP	W100	PBBM	PF	Total score
PL-C-1.7-20	4.48	2.22	2.01	1.68	1.59	1.89	1.91	2.55	6.03	4.01	2.41	30.78
PL-C-1.7-36	4.23	2.82	1.80	1.80	2.08	2.57	2.58	3.27	6.24	4.77	2.87	35.02
PL-C-2.2-29	3.80	1.00	2.11	4.58	2.69	2.34	2.04	2.82	4.59	3.89	2.76	32.62
PL-C-3.12-10	2.95	2.22	1.60	2.46	1.95	1.36	2.05	1.78	5.41	2.31	1.58	25.66
PL-C-3.12-25	5.08	2.22	1.60	2.10	1.25	1.29	1.20	1.84	4.80	3.24	1.56	26.17
PL-C-3.12-39	3.55	2.22	1.60	1.58	2.87	2.77	3.94	3.27	4.70	4.15	3.08	33.73
PL-C-4.1-17*	4.23	2.22	1.50	3.23	3.39	4.18	3.57	4.86	5.79	5.40	3.61	41.97
PL-C-9.1-8	4.23	2.22	1.80	1.77	2.43	2.69	3.13	3.07	6.56	3.78	2.34	34.01
PL-C-10.16-14	3.80	1.00	1.30	2.47	3.22	3.35	3.82	3.89	4.78	4.58	2.75	34.98
PL-C-17.9-11	3.12	1.00	1.00	3.86	4.06	3.65	4.04	4.23	3.52	4.86	2.61	35.95
PL-C-17.13-2	2.95	1.00	2.31	1.17	1.55	2.07	2.12	2.87	5.85	4.87	2.66	29.40
PL-C-17.13-4	3.46	1.00	1.00	1.75	1.75	2.06	2.12	2.59	5.94	3.63	1.57	26.88
PL-C-17.13-21	4.23	2.22	1.30	1.17	1.63	1.80	2.25	2.23	5.23	3.35	1.75	27.15
PL-C-17.13-43	2.53	1.00	1.40	1.16	1.58	1.87	2.18	2.53	5.38	4.21	1.73	25.58
PL-C-18.2-1	3.55	1.00	2.01	3.16	3.62	2.15	3.92	2.49	3.45	1.88	2.35	29.58
PL-C-18.2-20	1.68	1.00	1.20	3.06	3.08	2.80	3.26	3.25	5.75	3.73	2.28	31.09
PL-C-18.8-22	3.97	1.00	1.00	2.00	2.13	2.47	2.55	2.99	5.76	3.94	1.97	29.78
PL-C-18.8-40	4.23	1.00	2.01	1.52	1.62	2.11	2.04	2.77	5.12	4.64	2.36	29.41
PL-C-19.4-21	1.00	1.00	1.00	4.35	3.80	2.85	3.45	3.08	4.68	3.09	2.02	30.33
PL-C-19.10-43	4.48	2.82	2.61	1.02	1.71	2.22	2.46	2.93	5.48	4.64	3.17	33.55
PL-C-20.1-1*	3.89	1.00	1.80	3.50	3.50	4.17	3.55	5.02	5.55	5.61	4.14	41.74
PL-C-20.5-44*	2.78	1.00	1.00	3.42	4.01	4.44	4.25	5.10	6.01	4.82	3.06	39.88
PL-C-20.5-49*	3.80	1.00	1.20	3.34	2.88	3.99	2.85	4.96	5.98	6.04	3.27	39.32
PL-C-20.6-6	2.02	1.00	1.40	1.36	1.51	2.26	1.95	3.08	5.72	5.28	2.34	27.92
PL-C-20.6-7	1.85	1.00	1.20	1.48	1.61	2.48	2.04	3.29	7.11	5.16	2.22	29.44
PL-C-20.6-50	5.08	1.00	1.70	1.14	1.73	2.35	2.44	3.09	5.64	4.83	2.60	31.60
PL-C-20.11-44*	2.10	1.00	1.20	3.90	4.83	4.59	4.96	4.92	5.37	4.11	3.24	40.23
PL-A-1.1-8	2.36	1.00	1.30	2.75	1.40	1.15	1.15	1.60	3.59	2.59	1.36	20.25
PL-A-1.1-12	3.21	1.00	1.60	3.45	2.83	1.00	2.73	1.00	1.00	1.00	1.00	19.82
PL-A-1.1-20	2.10	1.00	1.60	1.69	1.00	1.44	1.00	2.24	5.23	4.83	1.86	23.99
PL-A-1.1-21	1.59	1.00	2.01	2.52	1.50	1.87	1.39	2.62	4.92	4.79	2.45	26.67
PL-A-1.1-22	2.10	1.00	1.60	1.72	1.02	1.15	1.01	1.93	5.36	4.58	1.57	23.04
PL-A-1.1-24	2.02	1.00	2.41	2.97	2.32	2.61	2.30	3.29	5.02	4.63	3.27	31.83
PL-A-1.1-35	4.14	1.00	1.91	2.44	1.85	1.70	1.91	2.21	4.30	3.42	2.07	26.95
PL-A-1.1-41	3.21	1.00	3.21	2.83	1.77	1.53	1.62	2.06	4.15	3.42	2.45	27.27
PL-A-1.1-43	4.14	1.00	2.81	2.53	1.60	1.53	1.52	2.19	4.39	3.90	2.43	28.05
PL-A-1.1-45	3.97	1.00	2.41	1.47	1.08	1.47	1.22	2.30	4.72	5.04	2.42	27.09
PL-A-1.1-47	3.97	1.00	1.70	2.62	2.35	3.04	2.52	3.76	6.27	4.89	3.08	35.20
PL-A-1.1-48	4.40	1.00	1.80	1.54	1.05	1.69	1.14	2.53	5.29	5.30	2.25	28.00
PL-A-6.5-1	4.14	1.00	1.60	2.95	2.59	2.30	2.67	2.41	5.79	2.59	2.04	30.09
PL-A-6.5-4*	3.21	1.00	2.61	3.86	4.32	4.62	4.36	4.74	6.45	3.99	4.98	44.13
PL-A-6.5-6*	3.29	1.00	2.61	3.79	3.09	3.81	2.88	4.42	6.00	5.04	4.66	40.58
PL-A-6.5-15*	2.70	1.00	3.82	3.82	3.20	3.29	3.01	3.68	5.87	3.87	5.12	39.36
PL-A-6.5-21	2.02	1.00	3.61	4.33	3.30	3.25	2.87	3.37	5.96	3.26	4.50	37.47
PL-A-6.5-23*	1.93	7.08	3.41	3.37	2.80	3.11	2.73	3.55	5.70	4.05	4.55	42.28
PL-A-6.5-29*	3.21	1.00	2.81	3.73	4.22	4.17	4.32	4.43	5.74	3.92	4.55	42.09
PL-A-7.2-29*	4.40	1.00	2.01	2.49	1.59	1.79	1.52	2.48	4.77	4.27	1.71	28.03
PL-A-7.2-35	3.55	1.00	6.83	2.67	1.94	1.63	1.92	2.06	4.83	3.11	3.65	33.19
Average	149.38	26.81	26.18	156.65	2.67	5.93	2.21	2.47	59.52	56.46	31.53	31.53
Min value	140.00	17.00	22.06	99.64	1.31	4.18	0.49	1.56	22.38	9.80	16.00	16.00
Max value	150.00	75.00	31.50	256.37	4.55	8.64	3.88	2.87	82.58	121.93	44.13	44.13
Standard deviation	1.65	9.94	2.65	40.92	0.90	1.12	0.83	0.21	11.94	27.24	6.44	6.44
The best value limit	151.02	36.76	28.83	197.57	3.56	7.05	3.03	2.68	71.45	83.70	37.97	37.97

*=selected strains. PH=Plant height (cm), HD=harvest date (days), NP=number of panicle (no.), LP=length of panicle (cm), NSP=number of seed per panicle (no.), WSP=weight of seed per panicle (g), DS=density of seed (no./cm), WFP=weight of fertile seed per panicle (g), W100=weight of 100 seeds (g), PF=percentage of fertile seed (%)

and 5 strains of the argon ion irradiation treatment for selection criteria, middle maturing, high posture, and many productive tillers. The ten strains are then grouped according to the level of similarity with the cluster analysis method. As for the level of similarity of these strains based on the production, components are presented in Figure 2.

Figure 2 shows that based on the dendrogram of 10 Toraja local red rice strains selected at the

90% similarity coefficient to form 5 Groups, namely Group I consists of 1 strain, Group II consists of 2 Subgroups (3 strains), Group III consists of 1 strain, Group IV consists of 2 strains and Group V consists of 2 subgroups (3 strains). The group analysis of the production components in the two planting locations based on the dendrogram graph is presented in Table 4.

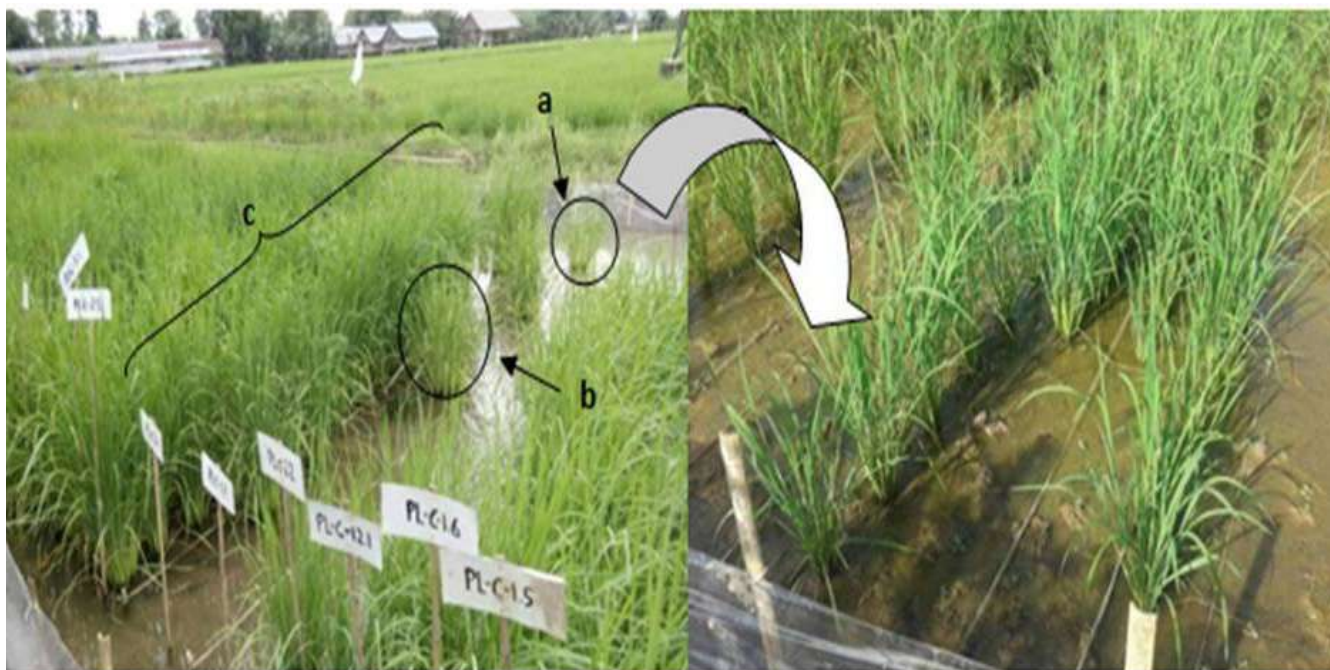


Figure 1. Variations of rice plant phenotypes at age (a) dwarf-type, (b) semi-dwarf type, (c) normal type

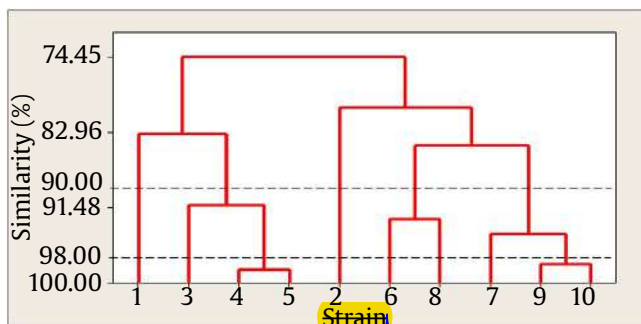


Figure 2. Dendrogram grouping of 10 selected strains of production components

Table 4. Grouping of yield components data 10 strains selected at the planting location with a similarity rate of 90%

Yield Components	Number of strain
Group I	1
Group II	3, 4, 5
Group III	2
Group IV	6, 8
Group V	7, 9, 10

1=PL-C-4.1-17, 2=PL-C-20.1-1, 3=PL-C-20.5-44, 4=PL-C-20.5-49, 5=PL-C-20.11-44, 6=PL-A-6.5-4, 7=PL-A-6.5-6, 8=PL-A-6.5-15, 9=PL-A-6.5-23, 10=PL-A-6.5-29

4. Discussion

The results of this study indicate that irradiation of ion beams using ion beam carbon and argon can be used to improve the character of local Toraja red rice plants, particularly related to the characteristics of harvest age, posture, and crop production. The control plants were harvested 162-166 days after seedling, while the mutant was harvested in the range of 146-161 days after seedling. Irradiation with ion beams speeds up the harvest time for 16 days or about 10% of the age of its parents. The use of carbon ion irradiation to produce plants with faster harvest time was also reported by Dong and Li (2012) in sweet sorghum plants and Du *et al.* (2014) in *Arabidopsis thaliana*.

In the areas of plant origin, namely in North Toraja Regency, the age of harvesting of red rice (Lea Pare) is included in the deepest age category of 180 days after seedling (Sahardi *et al.* 2013). The length of harvest time causes the low interest of the community to cultivate this type of local rice. Lea Pare cultivation is very limited, only to meet the needs of the use of these types of rice in traditional rituals. The presence of the Toraja local red rice variety with early maturing character is expected to increase public interest in cultivating this type of rice and prevent the extinction of this species. The production of local red rice mutants with early maturity character can be the first step to produce early maturing varieties. In this study, ion beam irradiation using carbon ions produced 9 mutants with short postures. Unfortunately, these short characters are not present along with a short harvest age character and high production yield in one individual mutant, so they are not included in the selected strain group. Rice mutants with shorter postures due to ion beam irradiation were reported by Ling *et al.* (2013) in rice cultivars MR219. According to Ling *et al.* (2013), the increase in Carbon ion dosage is directly proportional to the inhibition of plant height growth.

Overall, in this study, ion beam irradiation produced 10 strains with shorter harvest ages and a high number of productive tillers. The character of the number of productive tillers is expected to be positively correlated with rice grain production. The use of ion beam irradiation to increase crop yields has been reported in various studies, including by Zhao *et al.* (2006) and Xie *et al.* (2008).

The results of the analysis and grouping of criteria on the three observation parameters, namely plant height, age of harvest, and productive tillers at the planting

location (Table 1) showed that carbon irradiation treatment produced higher mutant amounts compared to argon ion irradiation treatments, but in terms of frequency, Argon ions irradiation treatment produce higher mutant frequencies.

This shows that carbon ion irradiation treatment is better at producing mutants mainly in terms of plant height criteria, harvest age and panicle number. Mutations that occur may be a mutagenic effect of ion beam irradiation which indicates that the treatment is quite effective in creating genetic diversity in population M2. High frequency of mutations caused by irradiation of carbon ions is caused by the energy fired by carbon ions greater than that is 135 MeV/u compared to the energy in Argon ions which is only 95 MeV/u (Hayashi *et al.* 2007). This allows the amount of damage to the gene caused by the release of energy from the ions that are fired so that the diversity (mutants) generated is also increasing. Other advantages of ion beam mutagenesis include high survival rates, high levels of mutation induction and various other variations (Yazid and Muryono 2000).

From a total of 48 strains (Table 2), after the housing was done against the score obtained and compared with the best total score (above 37.97), 10 strains were selected with the total score above the best score (Table 3), namely 5 strains in carbon treatment (PL-C-4.1-17, PL-C-20.1-1, PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44) and 5 in the treatment argon (PL-A-6.5-4, PL-A-6.5-6, PL-A-6.5-15, PL-A-6.5-23, and PL-A-6.5-29).

The selection of character selection in the tested population is based on components that have a close relationship with the character of production. Thus, the selection of populations of diploid rice strains can use the character of the generative phase of plant height, a number of pithy grain per panicle, total grain per panicle, weight of 1,000 rice grain and character of grain yield per clump. Aryana (2007) explained the character of number of tillers, number of rice grain, total grain count per panicle, and weight of 1,000 grains can be used as character selection, whereas in Safitri *et al.* (2011) explained the character of the number of productive tillers, number of rice grain per panicle, and the weight of 1,000 grains can be used as character selection.

In Figure 2 and Table 1 above, it can be seen that at the 90% similarity level, based on the data of the production components of the 10 selected strains, 5 groups were obtained, namely Group I consisting of 1 strain, namely PL-C-4.1-17. Group II consisted of 2

subgroups consisting of 3 strains namely PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44. Group III consisted of 1 strain, namely PL-C-20.1-1. Group IV consists of 2 strains namely PL-A-6.5-4 and PL-A-6.5-15. Group V consists of 2 subgroups consisting of 3 strains namely PL-A-6.5-6, PL-A-6.5-23, and PL-A-6.5-29.

Based on the level of similarity it can be seen that the PL-C-20.5-49 and PL-C-20.11-44 and PL-A-6.5-23 and PL-A-6.5-29 strains have very close kinship relationships. The line has a 95% similarity level. This shows that strains that are closely related will have a close genetic distance. This is consistent with Lee (1998)'s opinion that closely related individuals will have a close genetic distance, whereas if they are distant relatives they will have a far genetic distance. The results of the grouping of 10 selected strains consisting of 5 groups showed that the strains in the same group generally had almost the same number of strains. Erlina et al. (2011) suggests that the same cluster is characterized by almost the same genotype name. This shows that the genotypes are formed from the same population so that the level of kinship is closer.

5. Conclusion

The results of this study showed treatment with ion beam irradiation either by using carbon ions as well as argon ions are capable of producing Toraja local red rice mutant genotype with a shorter harvest age compared to the parental. There is an acceleration of age harvest 16-17 days or about 10% from the harvesting age of parental. Out of 48 total strains, 10 strains were selected, 5 strains of carbon ion irradiation treatment (PL-C-4.1-17, PL-C-20.1-1, PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44) and 5 strains of argon ion irradiation treatment (PL-A-6.5-4, PL-A-6.5-6, PL-A-6.5-15, PL-A-6.5-23, and PL-A-6.5-29) for selection criteria of middle maturity, high posture and many productive tillers.

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Selection of Early Maturing and High Yielding Mutants of Toraja Local Red Rice Grown from M2-M3 Population after Ion Beam Irradiation

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ABSTRACT

This study aims to obtain the genotype of local Toraja red rice M2 mutants that have the potential to be developed into early maturing varieties and have high yields. Research carried out with a mass selection method consisting of two irradiation treatments, namely: (1) irradiation with a Carbon ion dose of 150 Gy (PL-C); (2) irradiation with Argon ion dose of 10 Gy (PL-A) and control as a comparison. Selected strains of M2 seed population were planted in experimental strains plus 2 strains as control, each row had 50 plants. Selection is done in two stages. The first stage, the selection is based on the criteria of plant growth components, to select rice plants that have early-middle age characters, dwarf-semi-dwarf, and many tillers. The second stage, selected strains from the scoring method were then grouped based on the degree of similarity with the cluster analysis method. Irradiation with ion beams produces mutants with shorter harvest times than their parents, with the harvest time 16-17 days or 10% shorter compared to their parents. This study produced 10 selected strains, 5 strains of carbon ion and 5 strains of argon ion irradiation treatment, and formed 5 groups with a 90% similarity coefficient.

1. Introduction

Based on data from the North Toraja District Agriculture Service (2013), Tana Toraja is one of the areas in South Sulawesi that has a diversity of local rice germplasm. Based on the results of a survey of local rice in Tana Toraja Regency conducted by Suhardi *et al.* (2013) it was found about 29 local Toraja rice varieties, each of which had exotic characteristics such as black, red, aromatic, and fluffier rice.

One of the local rice varieties used for generations as part of the tradition and culture of the Toraja community is red rice which in the local language is called Pare Lea. The use of local rice, especially red rice in traditional cultural rituals, helps to preserve this traditional rice variety amid the rapid use of introduced varieties.

Local rice, in general, has several disadvantages including long life, high plant posture and low

yield (Wahdah and Langai 2009). This character is a limiting factor that can cause farmers' interest in growing local rice to be lower, so it is feared that it will cause a lack of availability of local rice cultivars as a source of germplasm.

To optimize the potential of local rice as one of the germplasm sources for plant breeding, the first step that needs to be implemented is to improve the character of the local rice varieties. Character improvement can be done in various ways, both conventionally and mutation induction. As time goes on, breeding techniques are now being developed using ion beams through the use of heavy-ion beams. This ion beam is safer, does not damage the endosperm, because the dosage is low so the level of mutation induction is higher.

Compared to gamma rays, ion beam irradiation has several advantages including higher mutation rates, lower doses with high survival rates, high levels of mutation induction and various other variations, ion files can be focused and regulated through the embryo tissue, and does not damage the endosperm

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(Yazid and Muryono 2000). Thus, mutation induction in local rice with radiation irradiation is expected to produce mutant rice that has properties that are better than the original varieties.

This research is expected to be the initial stage to create new red rice varieties early maturing, short postures, and high production. Based on these matters, it is necessary to carry out research "Early maturing Toraja Local Red Rice Mutant Selection from M2 Population from Ion Beam Irradiation".

2. Materials and Methods

2.1. Place and Time

This research was conducted in Alla Subdistrict, Enrekang Regency, South Sulawesi with an altitude of 650 m above sea level at coordinates S: 3° 19' 47.44"; E: 119° 50' 1.57" and lasted from May to October 2017.

2.2. Materials

The material used is the M2 generation of local Toraja red rice (Pare Lea) seed produced by ion beam irradiation. To obtain M2 seeds, Toraja local red rice seed (M0) was irradiated using ion beam carbon 150 Gy and ion beam argon 10 Gy, then grown on agricultural land in accordance with the standard of maintenance of rice plants. Individual plants that grew (M1) then produced seeds (M2) which were collected to be used in this study. Other materials used are manure and soil for seeding media, rat poisons, pesticides, irrigation water, and label paper. The tools used are plastic pots for sowing, machetes, hoes, tractors, sprayers, nets, meters, analytical scales, Contador seed counter, cameras, and writing instruments.

2.3. Methods

This study consisted of three treatments, namely irradiation with a dose of 150 Gy ion beam (PL-C), irradiation with argon ion dose 10 Gy (PL-A), and control as a comparison. M2 seeds used are seeds produced by M1 plants which produce more than 50 seeds per clump of plants. Based on these criteria there are 61 selected strains, consisting of 46 strains derived from irradiation carbon beam ion treatment and 15 strains of argon ion beam irradiation treatment. M2 seeds from selected strains are planted in rows containing 50 plants each. Thus the total seeds planted in the irradiation carbon beam ion treatment were 2,300 seeds, and in the irradiation treatment, the argon beam was 750 seeds.

As a control, two local Toraja red rice seeds were planted, each containing 50 plants. Each individual plant is given a line number that shows the row and position of the plant in the row. The total number of individual plants grown was 1,303 plants in the carbon beam ion irradiation treatment, 108 in the argon ion beam irradiation treatment and 89 plants in the control treatment.

2.4. Data Analysis

Observation data in the form of plant height, harvest age and number of productive tillers or panicle numbers are grouped into 3 criteria classes based on the Rice Plant Characterization and Evaluation System Guide (BPPP 2003), namely:

1. Parameters of plant height: dwarf (<110 cm), semi-dwarf (110-130 cm), and high (>130 cm).
2. Harvest age parameters: early (<120 days), middle (120-150 days), and deep (>150 days).
3. Parameters for the number of productive tillers: few (<8 panicles), middle (9-16 panicles), and many (≥ 17 panicles).

Furthermore, the selection activity was carried out to select rice plants that had early-maturity characters, short-medium canopy, and many tillers. Mutant frequency is calculated based on the number of mutants that have a certain character and the overall mutant plant population with the formula:

$$\text{Mutant frequency (MF)} = \frac{\text{Number of mutant plants}}{\text{Number of population}} \times 100\% \quad (1)$$

For the selection of strains based on the component production parameters, the scoring method is used. Each observation parameter from the production component will be given a score, and then summed to obtain the level of association. The final result of the scoring system is the classification of the level of relevance of the output parameters calculated by the equation:

$$S_n = (X_n - X_1) / SD + S_1 \quad (2)$$

Informations:

- S_n : data nth score
- SD : standard Deviation
- S_1 : lowest data score
- X_n : value of the nth data
- X_1 : lowest data value

3. Results

3.1. Number and Frequency of Mutants

Data on measuring plant height, harvest age and number of productive tillers are classified into three grouping criteria. The recapitulation of the number of individuals and the frequency of mutants based on the grouping criteria is presented in Table 1.

The data in Table I shows irradiation treatment, both by using carbon ions and argon ions to improve plant height character, by producing mutants that have dwarf and semi-dwarf plant postures, even in low frequencies. The irradiation treatment of carbon ions produced 9 plants mutant that had the character of dwarf plant postures with a frequency of 0.7% and 23 plants mutant of semi-dwarf plant postures with a frequency of 1.8%. While the argon-ion irradiation treatment only produces 1 plant that has the character of dwarf plant postures with a frequency of 0.9%. In the argon-ion, irradiation treatment was not obtained with semi-dwarf plant posture. In the control treatment, all plant populations have high postures.

The ion beam irradiation treatment also succeeded in producing M2 mutants with an early

harvest age than the control treatment. The carbon ion irradiation treatment produced 113 plants of mutants with middle maturing (120-150 days) with a mutant frequency of 8.7%. Meanwhile, argon ion irradiation produces 23 plants middle maturing mutant (120-150 days) with a frequency of 21.3%. While in the control treatment, all plant populations were in the deep maturing (>150 days).

For the character of a large number of productive tillers (>17), the treatment of irradiation with a carbon beam ion produced the highest number of clumps of 384 plants with a frequency of 29.5%, while the treatment of argon ion irradiation reached 98 plants with a frequency of 90.7%. In the control treatment, the number of clumps with many productive tillers characters reached 37 plants with a frequency of 41.6%.

3.2. Selections Strain Based on Growth Components

Based on the grouping of growth components that have been done previously, namely in terms of plant height, harvest age and number of productive tillers, a combination of the three criteria is then carried out. From the results of the third combination, these

Table 1. Recapitulation of the number of individuals and mutant frequencies based on plant height, harvest age and the number of productive tillers

Character	Criteria	Treatments*					
		PL-control		PL-carbon		PL-Argon	
		Number of plants (no.)	Frequency (%)	Number of plants (no.)	Frequency (%)	Number of plants (no.)	Frequency (%)
Plant height	Dwarf (<110 cm)	0	0.0	9	0.7	1	0.9
	Semi-dwarf (110-130 cm)	0	0.0	23	1.8	0	0.0
	High (>130 cm)	89	100.0	1,271	97.5	107	99.1
Harvest date	Early (<120 days)	0	0.0	0	0.0	0	0.0
	Middle (120-150 days)	0	0.0	113	8.7	23	21.3
	Deep (>150 days)	89	100.0	1,190	91.3	85	78.7
Number of panicle per plant	Early (<120 days)	27	30.3	340	26.1	3	2.8
	Middle (120-150 days)	25	28.1	579	44.4	7	6.5
	Deep (>150 days)	37	41.6	384	29.5	98	90.7
Total number of plants		89		1,303		108	

*PL-control: M2 plants without irradiation, PL-carbon: M2 plants with carbon-ion irradiation, PL-argon: M2 plants with argon-ion irradiation

components, 27 types of criteria were obtained from each treatment and the number of strains included in the criteria. The combination results obtained are presented in Table 2.

Table 2 shows that there are no strains that have criteria for early maturing, dwarf postures and many tillers so that the criteria used to select are criteria for middle maturing, high posture, and many tillers. Based on these criteria, 27 plants were selected in the irradiation treatment of carbon beam ions and 21 plants in the ion beam argon irradiation treatment, a total of 48 plants were selected.

3.3. Selection Strain Based on Yield Components

Based on the table of recapitulation of the number of selected strains in terms of growth components

(plant height, the age of harvest, and productive tillers) in Table 2, the production component data is then calculated. From the results of these calculations, the strains are grouped according to the criteria of production components that have been predetermined. The results of the calculation and grouping are then scored in the highest order. The results of the selection and scoring of selected strains based on production component data can be seen in Table 3.

From the results of the scoring that has been done, strains are selected which have a total score above the best value limit of 37.97. The best value limit is obtained from the average value of the total score plus the standard deviation value. There were 10 strains that had a score above the best value, each of the 5 strains of the carbon ion irradiation treatment

Table 2. Recapitulation of population numbers by a combination of criteria of harvest age, plant height and number of productive tillers

Harvest maturing	Criteria		Treatments		
	Plant height	Productive tillers	PL-control	PL-carbon	PL-argon
Early	Dwarf	Few	0	0	0
Early	Dwarf	Middle	0	0	0
Early	Dwarf	Many	0	0	0
Early	Semi-Dwarf	Few	0	0	0
Early	Semi-Dwarf	Middle	0	0	0
Early	Semi-Dwarf	Many	0	0	0
Early	High	Few	0	0	0
Early	High	Middle	0	0	0
Early	High	Many	0	0	0
Middle	Dwarf	Few	0	0	0
Middle	Dwarf	Middle	0	0	0
Middle	Dwarf	Many	0	0	0
Middle	Semi-Dwarf	Few	0	1	0
Middle	Semi-Dwarf	Middle	0	0	0
Middle	Semi-Dwarf	Many	0	0	0
Middle	High	Few	0	31	1
Middle	High	Middle	0	53	1
Middle*	High*	Many*	0	27	21
Deep	Dwarf	Few	0	8	0
Deep	Dwarf	Middle	0	1	0
Deep	Dwarf	Many	0	0	1
Deep	Semi-Dwarf	Few	0	14	0
Deep	Semi-Dwarf	Middle	0	7	0
Deep	Semi-Dwarf	Many	0	0	0
Deep	High	Few	31	360	2
Deep	High	Middle	21	445	6
Deep	High	Many	37	356	76
Total number of plants			89	1,303	108

*Criteria of the selected strain

Table 3. Selection and scoring results of selected strains based on growth and yield components data on carbon and argon ion irradiation treatments

Nomor galur	PH	HD	NP	LP	NSP	WSP	DS	WFP	W100	PBBM	PF	Total score
PL-C-1.7-20	4.48	2.22	2.01	1.68	1.59	1.89	1.91	2.55	6.03	4.01	2.41	30.78
PL-C-1.7-36	4.23	2.82	1.80	1.80	2.08	2.57	2.58	3.27	6.24	4.77	2.87	35.02
PL-C-2.2-29	3.80	1.00	2.11	4.58	2.69	2.34	2.04	2.82	4.59	3.89	2.76	32.62
PL-C-3.12-10	2.95	2.22	1.60	2.46	1.95	1.36	2.05	1.78	5.41	2.31	1.58	25.66
PL-C-3.12-25	5.08	2.22	1.60	2.10	1.25	1.29	1.20	1.84	4.80	3.24	1.56	26.17
PL-C-3.12-39	3.55	2.22	1.60	1.58	2.87	2.77	3.94	3.27	4.70	4.15	3.08	33.73
PL-C-4.1-17*	4.23	2.22	1.50	3.23	3.39	4.18	3.57	4.86	5.79	5.40	3.61	41.97
PL-C-9.1-8	4.23	2.22	1.80	1.77	2.43	2.69	3.13	3.07	6.56	3.78	2.34	34.01
PL-C-10.16-14	3.80	1.00	1.30	2.47	3.22	3.35	3.82	3.89	4.78	4.58	2.75	34.98
PL-C-17.9-11	3.12	1.00	1.00	3.86	4.06	3.65	4.04	4.23	3.52	4.86	2.61	35.95
PL-C-17.13-2	2.95	1.00	2.31	1.17	1.55	2.07	2.12	2.87	5.85	4.87	2.66	29.40
PL-C-17.13-4	3.46	1.00	1.00	1.75	1.75	2.06	2.12	2.59	5.94	3.63	1.57	26.88
PL-C-17.13-21	4.23	2.22	1.30	1.17	1.63	1.80	2.25	2.23	5.23	3.35	1.75	27.15
PL-C-17.13-43	2.53	1.00	1.40	1.16	1.58	1.87	2.18	2.53	5.38	4.21	1.73	25.58
PL-C-18.2-1	3.55	1.00	2.01	3.16	3.62	2.15	3.92	2.49	3.45	1.88	2.35	29.58
PL-C-18.2-20	1.68	1.00	1.20	3.06	3.08	2.80	3.26	3.25	5.75	3.73	2.28	31.09
PL-C-18.8-22	3.97	1.00	1.00	2.00	2.13	2.47	2.55	2.99	5.76	3.94	1.97	29.78
PL-C-18.8-40	4.23	1.00	2.01	1.52	1.62	2.11	2.04	2.77	5.12	4.64	2.36	29.41
PL-C-19.4-21	1.00	1.00	1.00	4.35	3.80	2.85	3.45	3.08	4.68	3.09	2.02	30.33
PL-C-19.10-43	4.48	2.82	2.61	1.02	1.71	2.22	2.46	2.93	5.48	4.64	3.17	33.55
PL-C-20.1-1*	3.89	1.00	1.80	3.50	3.50	4.17	3.55	5.02	5.55	5.61	4.14	41.74
PL-C-20.5-44*	2.78	1.00	1.00	3.42	4.01	4.44	4.25	5.10	6.01	4.82	3.06	39.88
PL-C-20.5-49*	3.80	1.00	1.20	3.34	2.88	3.99	2.85	4.96	5.98	6.04	3.27	39.32
PL-C-20.6-6	2.02	1.00	1.40	1.36	1.51	2.26	1.95	3.08	5.72	5.28	2.34	27.92
PL-C-20.6-7	1.85	1.00	1.20	1.48	1.61	2.48	2.04	3.29	7.11	5.16	2.22	29.44
PL-C-20.6-50	5.08	1.00	1.70	1.14	1.73	2.35	2.44	3.09	5.64	4.83	2.60	31.60
PL-C-20.11-44*	2.10	1.00	1.20	3.90	4.83	4.59	4.96	4.92	5.37	4.11	3.24	40.23
PL-A-1.1-8	2.36	1.00	1.30	2.75	1.40	1.15	1.15	1.60	3.59	2.59	1.36	20.25
PL-A-1.1-12	3.21	1.00	1.60	3.45	2.83	1.00	2.73	1.00	1.00	1.00	1.00	19.82
PL-A-1.1-20	2.10	1.00	1.60	1.69	1.00	1.44	1.00	2.24	5.23	4.83	1.86	23.99
PL-A-1.1-21	1.59	1.00	2.01	2.52	1.50	1.87	1.39	2.62	4.92	4.79	2.45	26.67
PL-A-1.1-22	2.10	1.00	1.60	1.72	1.02	1.15	1.01	1.93	5.36	4.58	1.57	23.04
PL-A-1.1-24	2.02	1.00	2.41	2.97	2.32	2.61	2.30	3.29	5.02	4.63	3.27	31.83
PL-A-1.1-35	4.14	1.00	1.91	2.44	1.85	1.70	1.91	2.21	4.30	3.42	2.07	26.95
PL-A-1.1-41	3.21	1.00	3.21	2.83	1.77	1.53	1.62	2.06	4.15	3.42	2.45	27.27
PL-A-1.1-43	4.14	1.00	2.81	2.53	1.60	1.53	1.52	2.19	4.39	3.90	2.43	28.05
PL-A-1.1-45	3.97	1.00	2.41	1.47	1.08	1.47	1.22	2.30	4.72	5.04	2.42	27.09
PL-A-1.1-47	3.97	1.00	1.70	2.62	2.35	3.04	2.52	3.76	6.27	4.89	3.08	35.20
PL-A-1.1-48	4.40	1.00	1.80	1.54	1.05	1.69	1.14	2.53	5.29	5.30	2.25	28.00
PL-A-6.5-1	4.14	1.00	1.60	2.95	2.59	2.30	2.67	2.41	5.79	2.59	2.04	30.09
PL-A-6.5-4*	3.21	1.00	2.61	3.86	4.32	4.62	4.36	4.74	6.45	3.99	4.98	44.13
PL-A-6.5-6*	3.29	1.00	2.61	3.79	3.09	3.81	2.88	4.42	6.00	5.04	4.66	40.58
PL-A-6.5-15*	2.70	1.00	3.82	3.82	3.20	3.29	3.01	3.68	5.87	3.87	5.12	39.36
PL-A-6.5-21	2.02	1.00	3.61	4.33	3.30	3.25	2.87	3.37	5.96	3.26	4.50	37.47
PL-A-6.5-23*	1.93	7.08	3.41	3.37	2.80	3.11	2.73	3.55	5.70	4.05	4.55	42.28
PL-A-6.5-29*	3.21	1.00	2.81	3.73	4.22	4.17	4.32	4.43	5.74	3.92	4.55	42.09
PL-A-7.2-29*	4.40	1.00	2.01	2.49	1.59	1.79	1.52	2.48	4.77	4.27	1.71	28.03
PL-A-7.2-35	3.55	1.00	6.83	2.67	1.94	1.63	1.92	2.06	4.83	3.11	3.65	33.19
Average	149.38	26.81	26.18	156.65	2.67	5.93	2.21	2.47	59.52	56.46	31.53	31.53
Min value	140.00	17.00	22.06	99.64	1.31	4.18	0.49	1.56	22.38	9.80	16.00	16.00
Max value	150.00	75.00	31.50	256.37	4.55	8.64	3.88	2.87	82.58	121.93	44.13	44.13
Standard deviation	1.65	9.94	2.65	40.92	0.90	1.12	0.83	0.21	11.94	27.24	6.44	6.44
The best value limit	151.02	36.76	28.83	197.57	3.56	7.05	3.03	2.68	71.45	83.70	37.97	37.97

*=selected strains, PH=Plant height (cm), HD=harvest date (days), NP=number of panicle (no.), LP=length of panicle (cm), NSP=number of seed per panicle (no.), WSP=weight of seed per panicle (g), DS=density of seed (no./cm), WFP=weight of fertile seed per panicle (g), W100=weight of 100 seeds (g), PF=percentage of fertile seed (%)

and 5 strains of the argon-ion irradiation treatment for selection criteria, middle maturing, high posture, and many productive tillers. The ten strains are then grouped according to the level of similarity with the cluster analysis method. As for the level of similarity of these strains based on the production, components are presented in Figure 1.

Figure 1 shows that based on the dendrogram of 10 Toraja local red rice strains selected at the

90% similarity coefficient to form 5 Groups, namely Group I consists of 1 strain, Group II consists of 2 Subgroups (3 strains), Group III consists of 1 strain, Group IV consists of 2 strains and Group V consists of 2 subgroups (3 strains). The group analysis of the production components in the two planting locations based on the dendrogram graph is presented in Table 4.

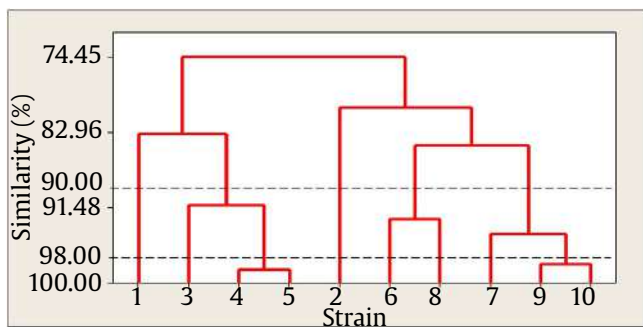


Figure 1. Dendrogram grouping of 10 selected strains of production components

Table 4. Grouping of yield components data 10 strains selected at the planting location with a similarity rate of 90%

Yield Components	Number of strain
Group I	1
Group II	3, 4, 5
Group III	2
Group IV	6, 8
Group V	7, 9, 10

1=PL-C-4.1-17, 2=PL-C-20.1-1, 3=PL-C-20.5-44, 4=PL-C-20.5-49, 5=PL-C-20.11-44, 6=PL-A-6.5-4, 7=PL-A-6.5-6, 8=PL-A-6.5-15, 9=PL-A-6.5-23, 10=PL-A-6.5-29

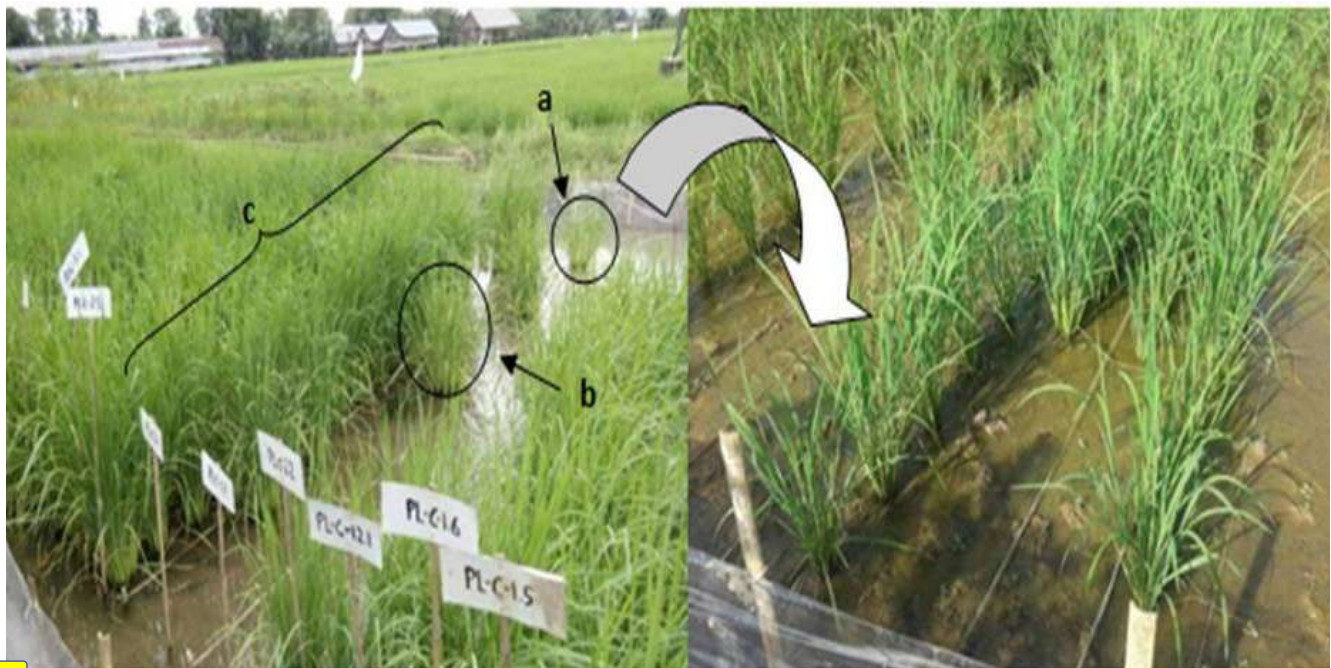


Figure 2. Variations of rice plant phenotypes at age (a) dwarf-type, (b) semi-dwarf type, (c) normal type

4. Discussion

The results of this study indicate that irradiation of ion beams using ion beam carbon and argon can be used to improve the character of local Toraja red rice plants, particularly related to the characteristics of harvest age, posture, and crop production. The control plants were harvested 162-166 days after seedling, while the mutant was harvested in the range of 146-161 days after seedling. Irradiation with ion beams speeds up the harvest time for 16 days or about 10% of the age of its parents. The use of carbon ion irradiation to produce plants with faster harvest time was also reported by Dong and Li (2012) in sweet sorghum plants and Du *et al.* (2014) in *Arabidopsis thaliana*.

In the areas of plant origin, namely in North Toraja Regency, the age of harvesting of red rice (Lea Pare) is included in the deepest age category of 180 days after seedling (Sahardi *et al.* 2013). The length of harvest time causes the low interest of the community to cultivate this type of local rice. Lea Pare cultivation is very limited, only to meet the needs of the use of these types of rice in traditional rituals. The presence of the Toraja local red rice variety with early maturing character is expected to increase public interest in cultivating this type of rice and prevent the extinction of this species. The production of local red rice mutants with early maturity character can be the first step to produce early maturing varieties. In this study, ion beam irradiation using carbon ions produced 9 mutants with short postures. Unfortunately, these short characters are not present along with a short harvest age character and high production yield in one individual mutant, so they are not included in the selected strain group. Rice mutants with shorter postures due to ion beam irradiation were reported by Ling *et al.* (2013) in rice cultivars MR219. According to Ling *et al.* (2013), the increase in Carbon ion dosage is directly proportional to the inhibition of plant height growth.

Overall, in this study, ion beam irradiation produced 10 strains with shorter harvest ages and a high number of productive tillers. The character of the number of productive tillers is expected to be positively correlated with rice grain production. The use of ion beam irradiation to increase crop yields has been reported in various studies, including by Zhao *et al.* (2006) and Xie *et al.* (2008).

The results of the analysis and grouping of criteria on the three observation parameters, namely plant height, age of harvest and productive tillers at the planting

location (Table 1) showed that carbon irradiation treatment produced higher mutant amounts compared to argon ion irradiation treatments, but in terms of frequency, Argon ions irradiation treatment produce higher mutant frequencies.

This shows that carbon ion irradiation treatment is better at producing mutants mainly in terms of plant height criteria, harvest age and panicle number. Mutations that occur may be a mutagenic effect of ion beam irradiation which indicates that the treatment is quite effective in creating genetic diversity in population M2. High frequency of mutations caused by irradiation of carbon ions is caused by the energy fired by carbon ions greater than that is 135 MeV/u compared to the energy in Argon ions which is only 95 MeV/u (Y. Hayashi *et al.* 2007). This allows the amount of damage to the gene caused by the release of energy from the ions that are fired so that the diversity (mutants) generated is also increasing. Other advantages of ion beam mutagenesis include high survival rates, high levels of mutation induction and various other variations (Yazid and Muryono 2000).

From a total of 48 strains (Table 2), after the housing was done against the score obtained and compared with the best total score (above 37.97), 10 strains were selected with the total score above the best score (Table 3), namely 5 strains in carbon treatment (PL-C-4.1-17, PL-C-20.1-1, PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44) and 5 in the treatment argon (PL-A-6.5-4, PL-A-6.5-6, PL-A-6.5-15, PL-A-6.5-23, and PL-A-6.5-29).

The selection of character selection in the tested population is based on components that have a close relationship with the character of production. Thus, the selection of populations of diploid rice strains can use the character of the generative phase of plant height, a number of pithy grain per panicle, total grain per panicle, weight of 1,000 rice grain and character of grain yield per clump. Aryana *et al.* (2011) explained the character of number of tillers, number of rice grain, total grain count per panicle, and weight of 1,000 grains can be used as character selection, whereas in Safitri *et al.* (2011) explained the character of the number of productive tillers, number of rice grain per panicle, and the weight of 1,000 grains can be used as character selection.

In Figure 1 and Table 1 above, it can be seen that at the 90% similarity level, based on the data of the production components of the 10 selected strains, 5 groups were obtained, namely Group I consisting of 1 strain, namely PL-C-4.1-17. Group II consisted of 2

subgroups consisting of 3 strains namely PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44. Group III consisted of 1 strain, namely PL-C-20.1-1. Group IV consists of 2 strains, namely PL-A-6.5-4 and PL-A-6.5-15. Group V consists of 2 subgroups consisting of 3 strains, namely PL-A-6.5-6, PL-A-6.5-23, and PL-A-6.5-29.

Based on the level of similarity it can be seen that the PL-C-20.5-49 and PL-C-20.11-44 and PL-A-6.5-23 and PL-A-6.5-29 strains have very close kinship relationships. The line has a 95% similarity level. This shows that strains that are closely related will have a close genetic distance. This is consistent with Lee (1998)'s opinion that closely related individuals will have a close genetic distance, whereas if they are distant relatives they will have a far genetic distance. The results of the grouping of 10 selected strains consisting of 5 groups showed that the strains in the same group generally had almost the same number of strains. Erlina (2011) suggests that the same cluster is characterized by almost the same genotype name. This shows that the genotypes are formed from the same population so that the level of kinship is closer.

5. Conclusion

The results of this study showed treatment with ion beam irradiation either by using carbon ions as well as argon ions are capable of producing Toraja local red rice mutant genotype with a shorter harvest age compared to the parental. There is an acceleration of age harvest 16-17 days or about 10% from the harvesting age of parental. Out of 48 total strains, 10 strains were selected, 5 strains of carbon ion irradiation treatment (PL-C-4.1-17, PL-C-20.1-1, PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44) and 5 strains of argon ion irradiation treatment (PL-A-6.5-4, PL-A-6.5-6, PL-A-6.5-15, PL-A-6.5-23, and PL-A-6.5-29) for selection criteria of middle maturity, high posture and many productive tillers.

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Selection of Early Maturing and High Yielding Mutants of Toraja Local Red Rice Grown from M2-M3 Population after Ion Beam Irradiation

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ABSTRACT

This study aims to obtain the genotype of local Toraja red rice mutants that have the potential to be developed into early maturing varieties and have high yields. Research carried out with a mass selection method consisting of two irradiation treatments, namely: (1) irradiation with a Carbon ion dose of 150 Gy (PL-C); (2) irradiation with Argon ion dose of 10 Gy (PL-A) and control as a comparison. Selected lines of M2 seed population were planted in experimental rows plus 2 rows as control, each row had 50 plants. Selection is done in two stages. The first stage, the selection was based on the criteria of plant growth components; to select rice plants that have early mid-early maturity characters, dwarf to semi-dwarf, and larger number of tillers. The second stage, selected lines from the scoring method were then grouped based on the degree of similarity with the cluster analysis method. Irradiation with ion beams produced mutants with shorter harvesting age than their. This study produced 10 selected lines, 5 lines of Carbon ion and 5 lines of Argon ion irradiation treatment, and formed 5 groups with a 90% similarity coefficient.

1. Introduction

Based on data from the North Toraja Regency Agriculture Service (2013), Tana Toraja is one of the areas in South Sulawesi that has a diversity of local rice germplasm. Survey of local rice in Tana Toraja Regency conducted by Sahardi and Djufry (2013) reported that there were about 29 local Toraja rice varieties, each of which had exotic characteristics such as black, red, aromatic, and fluffier rice found in that regency alone. However according to Riadi *et al.* (2019) there are about a total of 27 local Toraja varieties found in Tana Toraja Regency and 22 local varieties found in North Toraja Regency.

One of the local rice varieties used for generations as part of the tradition and culture of the Toraja community is red rice which in the local language is called Pare Lea.

The use of local rice, especially red rice in traditional cultural rituals, helps to preserve this traditional rice variety amid the rapid use of introduced varieties.

Local rice, in general, has several disadvantages including long life, high plant posture and low yield (Wahdah *et al.* 2012). This character is a limiting factor that can cause farmers' interest in growing local rice to be lower, so it is feared that it will cause a lack of availability of local rice cultivars as a source of germplasm.

To optimize the potential of local rice as one of the germplasm sources for plant breeding, the first step that needs to be implemented is to improve the character of the local rice varieties. Character improvement can be done in various ways, both conventionally and mutation induction. As time goes on, breeding techniques are now being developed using ion beams through the use of heavy-ion beams. This ion beam irradiation is safer, does not damage the endosperm, because the dosage is low so the level of mutation induction is higher.

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Compared to gamma rays, ion beam irradiation has several advantages including higher mutation rates, lower doses with high survival rates, high levels of mutation induction and various other variations, ion files can be focused and regulated through the embryo tissue, and does not damage the endosperm (Yazid and Muryono 2000; Abe *et al.* 2012). Thus, mutation induction in local rice with radiation irradiation is expected to produce mutant rice that has properties that are better than the original varieties.

This research is expected to be the initial stage to create new red rice varieties early maturing, short postures, and high production. Based on these matters, it is necessary to carry out research "Early maturing Toraja Local Red Rice Mutant Selection from M2 Population from Ion Beam Irradiation".

2. Materials and Methods

2.1. Place and Time

This research was conducted in Alla Subdistrict, Enrekang Regency, South Sulawesi with an altitude of 650 m above sea level at coordinates S: 3° 19' 47.44"; E: 119° 50' 1.57" and lasted from May to October 2017.

2.2. Materials

The material used was the M2 generation of local Toraja red rice (Pare Lea) seed produced by ion beam irradiation (Sjahril *et al.* 2018). Toraja local red rice seed (M0) was irradiated using 150 Gy Carbon ion beam and 10 Gy Argon ion beam, then grown on agricultural land in accordance with the standard of maintenance of rice plants. Individual M1 plants grew then produced seeds (M2) which were collected to be used in this study. Other materials used were manure and soil for seeding media, rat poisons, pesticides, irrigation water, and paper label. The tools used were plastic pots for sowing, machetes, hoes, tractors, sprayers, nets, meters, analytical scales, Contador seed counter, cameras, and writing instruments.

2.3. Methods

This study consisted of three treatments, namely irradiation with a dose of 150 Gy Carbon ion beam (PL-C), irradiation with Argon ion dose 10 Gy (PL-A), and control as a comparison. M2 seeds used are seeds produced by M1 plants which produce more than 50 seeds per hill of plants. Based on these criteria there are 61 selected lines, consisting of 46 lines derived from irradiation Carbon ion beam treatment and

15 lines of Argon ion beam irradiation treatment. M2 seeds from selected lines were planted in rows containing 50 plants each. Thus the total seeds planted from the Carbon ion beam irradiation were 2,300 seeds, and 750 seeds from Argon ion beam irradiation treatment. As a control, non-irradiated local Toraja red rice were planted in rows, each containing 50 plants. Each individual plant was given a line number that shows the row and position of the plants in the rows. The total number of individual plants grown were 1,303 plants from the Carbon ion beam irradiation, 108 from the Argon ion beam irradiation and 89 plants from control treatment.

2.4. Data Analysis

Observation data in the form of plant height, harvest age and number of productive tillers counted as panicle numbers are grouped into 3 criteria classes based on the Rice Plant Characterization and Evaluation System Guide (BPPP 2003), namely:

1. Parameters of plant height: dwarf (<110 cm), semi-dwarf (110-130 cm), and high (>130 cm).
2. Harvesting age parameters: early (<120 days), middle (120-150 days), and deep (>150 days). Harvesting age is defined here as additional days of flowering growth of average 30 days from heading.
3. Parameters for the number of productive tillers: few (<8 panicles), middle (9-16 panicles), and many (≥17 panicles).

Furthermore, selection was carried out to select rice plants that had early-maturity characters, short-medium canopy, and many tillers. Mutant frequency was calculated based on the number of mutants that have a certain character and the overall mutant plant population with the formula:

$$\text{Mutant frequency (MF)} = \frac{\text{Number of mutant plants}}{\text{Number of population}} \times 100\% \quad (1)$$

For the selection of mutant lines based on the component production parameters, the scoring method was used. Each observation parameter from the production component was given a score, and then summed to obtain the level of association. The final result of the scoring system was the classification of the level of relevance of the output parameters calculated by the equation:

$$S_n = (X_n - X_1) / SD + S_1 \quad (2)$$

Informations:

- S_n : data nth score
 SD : standard Deviation
 S_1 : lowest data score
 X_n : value of the nth data
 X_1 : lowest data value

3. Results

3.1. Number and Frequency of Mutants

Data on plant height (cm), harvesting age (days), and number of productive tillers (panicle) are classified into three grouping criteria. The recapitulation of the number of individuals and the frequency of mutants based on the grouping criteria is presented in Table 1.

The data in Table 1 shows irradiation treatment, both by using Carbon ions and Argon ions to improve plant height character, by producing mutants that have dwarf and semi-dwarf plant postures, even in low frequencies. The irradiation treatment of Carbon ions produced 9 plants mutant that had the character of dwarf plant postures with a frequency of 0.7% and 23 plants mutant of semi-dwarf plant postures with a frequency of 1.8%. While the Argon ion irradiation treatment only produces only one plant that has the character of dwarf plant postures with a frequency of 0.9%. In the Argon ion irradiation treatment was none obtained with semi-dwarf plant posture. Whereas, in the control treatment, all plant populations have high postures. Growth in the field is shown in Figure 1. Figure

1 shows field planting condition of M2 normal plants, dwarf, and semi dwarf plants.

The ion beam irradiation treatment also succeeded in producing M2-M3 mutants with a mid-early maturity harvesting age (147-148 days) than the control treatment (deep maturity, 162-165 days). In average we obtained 16-17 days earlier harvesting age (data not shown). Carbon ion irradiation treatment produced 113 mutants plants with mid-early maturing harvesting age mutant (120-150 days) with a mutant frequency of 8.7%. Meanwhile, Argon ion irradiation produces 23 plants middle maturing mutant (120-150 days) with a frequency of 21.3%. While in control treatment, all plant populations were in the deep maturing (>150 days).

The highest number of productive tillers (≥ 17 panicles per hill) per line was found in the treatment of irradiation with Carbon ion beam (i.e. 384 hills) with a frequency of 29.5%, while the treatment of Argon ion irradiation reached 98 hills with a frequency of 90.7%. In the control treatment, the number of hills with many productive tillers characters reached 37 plants with a frequency of 41.6%.

3.2. Lines Selection Based on Growth Components

Based on the grouping of growth components that have been done previously, in terms of plant height, harvesting age and number of productive tillers, a combination of the three criteria is then carried out. From the results of the third combination, these components, 27 types of criteria were obtained from each treatment and the number of lines included in the criteria. The combination results obtained are presented in Table 2.

Table 1. Recapitulation of the number of individuals and mutant frequencies based on plant height (cm), harvesting age (days), and the number of productive tillers (panicles)

Character	Criteria	Treatments*					
		PL-control		PL-Carbon		PL-Argon	
		Number of plants	Frequency (%)	Number of plants	Frequency (%)	Number of plants	Frequency (%)
Plant height	Dwarf (<110 cm)	0	0.0	9	0.7	1	0.9
	Semi-dwarf (110-130 cm)	0	0.0	23	1.8	0	0.0
	High (>130 cm)	89	100.0	1,271	97.5	107	99.1
Harvesting age	Early (<120 days)	0	0.0	0	0.0	0	0.0
	Middle (120-150 days)	0	0.0	113	8.7	23	21.3
	Deep (>150 days)	89	100.0	1,190	91.3	85	78.7
Number of productive tillers per hill	Few (≤ 8 panicles)	27	30.3	340	26.1	3	2.8
	Middle (9-16 panicles)	25	28.1	579	44.4	7	6.5
	Many (≥ 17 panicles)	37	41.6	384	29.5	98	90.7
Total number of plants		89		1,303		108	

*PL-control: M2 plants without irradiation, PL-Carbon: M2 plants with Carbon ion irradiation, PL-Argon: M2 plants with Argon ion irradiation

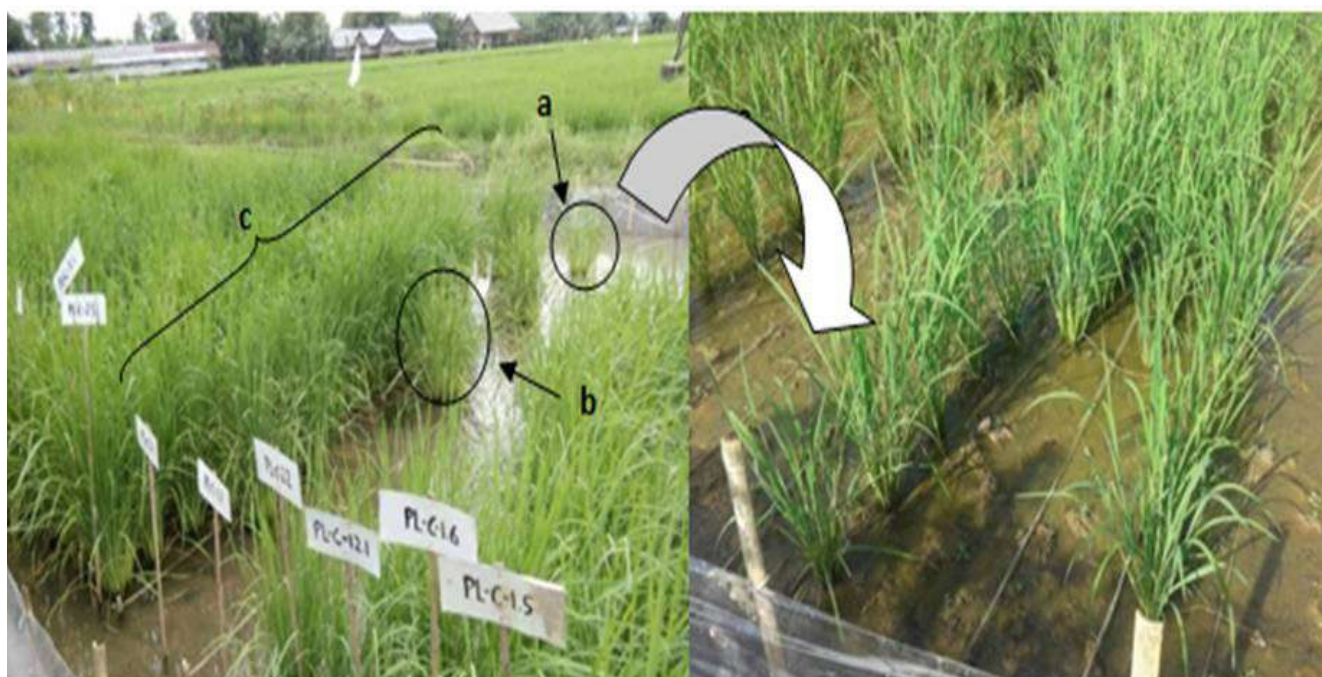


Figure 1. Variations of rice plant phenotypes at 40 days after planting (a) dwarf-type, (b) semi-dwarf type, (c) normal type

Table 2. Recapitulation of population numbers by a combination of qualitative criteria of harvesting age, plant height and number of productive tillers (panicles)

Harvesting age*	Criteria		Treatments		
	Plant height	Productive tillers	PL-control	PL-Carbon	PL-Argon
Early	Dwarf	Few	0	0	0
Early	Dwarf	Middle	0	0	0
Early	Dwarf	Many	0	0	0
Early	Semi-Dwarf	Few	0	0	0
Early	Semi-Dwarf	Middle	0	0	0
Early	Semi-Dwarf	Many	0	0	0
Early	High	Few	0	0	0
Early	High	Middle	0	0	0
Early	High	Many	0	0	0
Mid-early	Dwarf	Few	0	0	0
Mid-early	Dwarf	Middle	0	0	0
Mid-early	Dwarf	Many	0	0	0
Mid-early	Semi-Dwarf	Few	0	1	0
Mid-early	Semi-Dwarf	Middle	0	0	0
Mid-early	Semi-Dwarf	Many	0	0	0
Mid-early	High	Few	0	31	1
Mid-early	High	Middle	0	53	1
Mid-early**	High**	Many**	0	27	21
Deep	Dwarf	Few	0	8	0
Deep	Dwarf	Middle	0	1	0
Deep	Dwarf	Many	0	0	1
Deep	Semi-Dwarf	Few	0	14	0
Deep	Semi-Dwarf	Middle	0	7	0
Deep	Semi-Dwarf	Many	0	0	0
Deep	High	Few	31	360	2
Deep	High	Middle	21	445	6
Deep	High	Many	37	356	76
Total number of plants			89	1,303	108

*Early maturity (146-153 days), Mid-early maturity (154-161 days), Deep maturity (162-168 days)

**Criteria of the selected line

Table 2 shows that there are no lines that meet the criteria for early maturing, dwarf postures and many tillers numbers, so that the criteria used to select are criteria for middle maturing, high posture, and many tillers. Based on these criteria, 27 plants were selected from the irradiation treatment of Carbon ion beam and 21 plants from Argon ion beam irradiation treatment. A total of 48 plants were selected.

3.3. Lines Selection Based on Yield Components

Based on the table of recapitulation of the number of selected lines in terms of growth components (plant height, harvesting age, and productive tillers) in Table 2, the production component data is then calculated. From the results of these calculations, the lines are grouped according to the criteria of production components that have been predetermined. The results of the calculation and grouping are then scored in the highest order. The results of the selection and scoring of selected lines based on production component data can be seen in Table 3.

From the results of the scoring, some lines have a total score above the best value limit of 37.97 were selected. The best value limit is obtained from the average value of the total score plus the standard deviation value. There were 10 lines that had a score above the best value, each of which 5 lines were from Carbon ion irradiation treatment and 5 lines were from the Argon ion irradiation treatment for selection criteria, middle maturing, high posture, and many productive tillers. The 10 lines are then grouped according to the level of similarity with the cluster analysis method. The level of similarity of these lines based on the production, components are presented in Figure 2.

Figure 2 shows that based on the dendrogram of 10 Toraja local red rice lines selected at the 90% similarity coefficient formed five groups. Group I consisted of 1 line, Group II consisted of 2 subgroups (3 lines), Group III of 1 line, Group IV of 2 lines, and Group V of 2 subgroups (3 lines). The group analysis of the production components based on the dendrogram graph is presented in Table 4.

Table 3. Selection and scoring results of selected lines based on growth and yield components data on Carbon and Argon ion irradiation treatments

Nomor galur	PH	HD	NP	LP	NSP	WSP	DS	WFP	W100	PBBM	PF	Total score
PL-C-1.7-20	4.48	2.22	2.01	1.68	1.59	1.89	1.91	2.55	6.03	4.01	2.41	30.78
PL-C-1.7-36	4.23	2.82	1.80	1.80	2.08	2.57	2.58	3.27	6.24	4.77	2.87	35.02
PL-C-2.2-29	3.80	1.00	2.11	4.58	2.69	2.34	2.04	2.82	4.59	3.89	2.76	32.62
PL-C-3.12-10	2.95	2.22	1.60	2.46	1.95	1.36	2.05	1.78	5.41	2.31	1.58	25.66
PL-C-3.12-25	5.08	2.22	1.60	2.10	1.25	1.29	1.20	1.84	4.80	3.24	1.56	26.17
PL-C-3.12-39	3.55	2.22	1.60	1.58	2.87	2.77	3.94	3.27	4.70	4.15	3.08	33.73
PL-C-4.1-17*	4.23	2.22	1.50	3.23	3.39	4.18	3.57	4.86	5.79	5.40	3.61	41.97
PL-C-9.1-8	4.23	2.22	1.80	1.77	2.43	2.69	3.13	3.07	6.56	3.78	2.34	34.01
PL-C-10.16-14	3.80	1.00	1.30	2.47	3.22	3.35	3.82	3.89	4.78	4.58	2.75	34.98
PL-C-17.9-11	3.12	1.00	1.00	3.86	4.06	3.65	4.04	4.23	3.52	4.86	2.61	35.95
PL-C-17.13-2	2.95	1.00	2.31	1.17	1.55	2.07	2.12	2.87	5.85	4.87	2.66	29.40
PL-C-17.13-4	3.46	1.00	1.00	1.75	1.75	2.06	2.12	2.59	5.94	3.63	1.57	26.88
PL-C-17.13-21	4.23	2.22	1.30	1.17	1.63	1.80	2.25	2.23	5.23	3.35	1.75	27.15
PL-C-17.13-43	2.53	1.00	1.40	1.16	1.58	1.87	2.18	2.53	5.38	4.21	1.73	25.58
PL-C-18.2-1	3.55	1.00	2.01	3.16	3.62	2.15	3.92	2.49	3.45	1.88	2.35	29.58
PL-C-18.2-20	1.68	1.00	1.20	3.06	3.08	2.80	3.26	3.25	5.75	3.73	2.28	31.09
PL-C-18.8-22	3.97	1.00	1.00	2.00	2.13	2.47	2.55	2.99	5.76	3.94	1.97	29.78
PL-C-18.8-40	4.23	1.00	2.01	1.52	1.62	2.11	2.04	2.77	5.12	4.64	2.36	29.41
PL-C-19.4-21	1.00	1.00	1.00	4.35	3.80	2.85	3.45	3.08	4.68	3.09	2.02	30.33
PL-C-19.10-43	4.48	2.82	2.61	1.02	1.71	2.22	2.46	2.93	5.48	4.64	3.17	33.55
PL-C-20.1-1*	3.89	1.00	1.80	3.50	3.50	4.17	3.55	5.02	5.55	5.61	4.14	41.74
PL-C-20.5-44*	2.78	1.00	1.00	3.42	4.01	4.44	4.25	5.10	6.01	4.82	3.06	39.88
PL-C-20.5-49*	3.80	1.00	1.20	3.34	2.88	3.99	2.85	4.96	5.98	6.04	3.27	39.32
PL-C-20.6-6	2.02	1.00	1.40	1.36	1.51	2.26	1.95	3.08	5.72	5.28	2.34	27.92
PL-C-20.6-7	1.85	1.00	1.20	1.48	1.61	2.48	2.04	3.29	7.11	5.16	2.22	29.44
PL-C-20.6-50	5.08	1.00	1.70	1.14	1.73	2.35	2.44	3.09	5.64	4.83	2.60	31.60
PL-C-20.11-44*	2.10	1.00	1.20	3.90	4.83	4.59	4.96	4.92	5.37	4.11	3.24	40.23
PL-A-1.1-8	2.36	1.00	1.30	2.75	1.40	1.15	1.15	1.60	3.59	2.59	1.36	20.25

Table 3. Continued

Nomor galur	PH	HD	NP	LP	NSP	WSP	DS	WFP	W100	PBBM	PF	Total score
PL-A-1.1-12	3.21	1.00	1.60	3.45	2.83	1.00	2.73	1.00	1.00	1.00	1.00	19.82
PL-A-1.1-20	2.10	1.00	1.60	1.69	1.00	1.44	1.00	2.24	5.23	4.83	1.86	23.99
PL-A-1.1-21	1.59	1.00	2.01	2.52	1.50	1.87	1.39	2.62	4.92	4.79	2.45	26.67
PL-A-1.1-22	2.10	1.00	1.60	1.72	1.02	1.15	1.01	1.93	5.36	4.58	1.57	23.04
PL-A-1.1-24	2.02	1.00	2.41	2.97	2.32	2.61	2.30	3.29	5.02	4.63	3.27	31.83
PL-A-1.1-35	4.14	1.00	1.91	2.44	1.85	1.70	1.91	2.21	4.30	3.42	2.07	26.95
PL-A-1.1-41	3.21	1.00	3.21	2.83	1.77	1.53	1.62	2.06	4.15	3.42	2.45	27.27
PL-A-1.1-43	4.14	1.00	2.81	2.53	1.60	1.53	1.52	2.19	4.39	3.90	2.43	28.05
PL-A-1.1-45	3.97	1.00	2.41	1.47	1.08	1.47	1.22	2.30	4.72	5.04	2.42	27.09
PL-A-1.1-47	3.97	1.00	1.70	2.62	2.35	3.04	2.52	3.76	6.27	4.89	3.08	35.20
PL-A-1.1-48	4.40	1.00	1.80	1.54	1.05	1.69	1.14	2.53	5.29	5.30	2.25	28.00
PL-A-6.5-1	4.14	1.00	1.60	2.95	2.59	2.30	2.67	2.41	5.79	2.59	2.04	30.09
PL-A-6.5-4*	3.21	1.00	2.61	3.86	4.32	4.62	4.36	4.74	6.45	3.99	4.98	44.13
PL-A-6.5-6*	3.29	1.00	2.61	3.79	3.09	3.81	2.88	4.42	6.00	5.04	4.66	40.58
PL-A-6.5-15*	2.70	1.00	3.82	3.82	3.20	3.29	3.01	3.68	5.87	3.87	5.12	39.36
PL-A-6.5-21	2.02	1.00	3.61	4.33	3.30	3.25	2.87	3.37	5.96	3.26	4.50	37.47
PL-A-6.5-23*	1.93	7.08	3.41	3.37	2.80	3.11	2.73	3.55	5.70	4.05	4.55	42.28
PL-A-6.5-29*	3.21	1.00	2.81	3.73	4.22	4.17	4.32	4.43	5.74	3.92	4.55	42.09
PL-A-7.2-29*	4.40	1.00	2.01	2.49	1.59	1.79	1.52	2.48	4.77	4.27	1.71	28.03
PL-A-7.2-35	3.55	1.00	6.83	2.67	1.94	1.63	1.92	2.06	4.83	3.11	3.65	33.19
Average	149.38	26.81	26.18	156.65	2.67	5.93	2.21	2.47	59.52	56.46	31.53	31.53
Min value	140.00	17.00	22.06	99.64	1.31	4.18	0.49	1.56	22.38	9.80	16.00	16.00
Max value	150.00	75.00	31.50	256.37	4.55	8.64	3.88	2.87	82.58	121.93	44.13	44.13
Standard deviation	1.65	9.94	2.65	40.92	0.90	1.12	0.83	0.21	11.94	27.24	6.44	6.44
The best value limit	151.02	36.76	28.83	197.57	3.56	7.05	3.03	2.68	71.45	83.70	37.97	37.97

*=selected lines, PH=Plant height (cm), HD=harvest date (days), NP=number of panicle (no.), LP=length of panicle (cm), NSP=number of seed per panicle (no.), WSP=weight of seed per panicle (g), DS=density of seed (no./cm), WFP=weight of fertile seed per panicle (g), W100=weight of 100 seeds (g), PF=percentage of fertile seed (%)

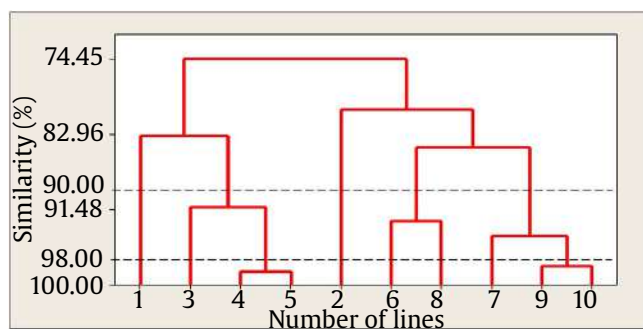


Figure 2. Dendrogram grouping of 10 selected lines of production components

4. Discussion

The results of this study indicated that irradiation of ion beams using Carbon and Argon ion beam can be used to improve the character of local Toraja red rice plants, particularly related to the characteristics of harvesting age, posture, and crop production. The control plants were harvested 162-166 days after seedling, while the mutant was harvested in the range of 146-161 days after seedling. Irradiation with ion beams speeds up the harvest time for 16 days or about 10% of the age of its parents. The use of Carbon

Table 4. Grouping of yield components data 10 lines selected at the planting location with a similarity rate of 90%

Yield Components	Number of line
Group I	1
Group II	3, 4, 5
Group III	2
Group IV	6, 8
Group V	7, 9, 10

1=PL-C-4.1-17, 2=PL-C-20.1-1, 3=PL-C-20.5-44, 4=PL-C-20.5-49, 5=PL-C-20.11-44, 6=PL-A-6.5-4, 7=PL-A-6.5-6, 8=PL-A-6.5-15, 9=PL-A-6.5-23, 10=PL-A-6.5-29

ion beam irradiation to produce plants with faster harvesting age was also reported by Dong and Li (2012) in sweet sorghum plants and Du *et al.* (2014) in *Arabidopsis thaliana*.

In the of the plant origin, namely in North Toraja Regency, the age of harvesting of red rice (Pare Lea) is categorized as deepest age category of 180 days after seedling (Sahardi *et al.* 2013). Usually it takes 21 to 30 days from seedling to planting. The long harvesting age causes low interest of the community to cultivate this type of local rice. Local Toraja rice (Pare Lea) cultivation is very limited, only to meet the needs of the use of

these types of rice in traditional ceremonies and rituals. The presence of the Toraja local red rice variety with early maturing character is expected to increase public interest in cultivating this type of rice and prevent the extinction of this species. The production of local red rice mutants with early maturity character could be the first step to produce early maturing varieties. In this study, ion beam irradiation using Carbon ions produced 9 mutants with short postures. Unfortunately, these short characters are not present along with a short harvesting age character (early maturing) and high production yield in one individual mutant, so they are not included in the selected line group. Rice mutants with shorter postures due to ion beam irradiation were reported by Ling *et al.* (2013) in rice cultivars MR219. According to Ling *et al.* (2013), the increase in Carbon ion dosage is directly proportional to the inhibition of plant height growth.

Overall, in this study, ion beam irradiation produced 10 lines with shorter harvest ages and a high number of productive tillers. The character of the number of productive tillers is expected to be positively correlated with rice grain production. The use of ion beam irradiation to increase crop yields has been reported in various studies, including by Zhao *et al.* (2006) and Xie *et al.* (2008).

The results of the analysis and grouping of criteria on the three observation parameters, namely plant height, harvesting age and productive tillers at the planting location (Table 1) showed that Carbon irradiation treatment produced higher mutant amounts compared to Argon ion irradiation treatments, but in terms of frequency, Argon ions irradiation treatment produce higher mutant frequencies.

This shows that Carbon ion irradiation treatment is better at producing mutants mainly in terms of plant height criteria, harvesting age and number of productive tillers. Mutations that occur may be a mutagenic effect of ion beam irradiation which indicates that the treatment is quite effective in creating genetic diversity in population M2-M3. High frequency of mutations caused by irradiation of Carbon ions is caused by the energy fired by carbon ions greater than 135 MeV/u compared to the energy in Argon ions which is only 95 MeV/u (Hayashi *et al.* 2007). This allows the amount of damage to the gene caused by the release of energy from the ions that are fired so that the diversity (mutants) generated is also increasing. Other advantages of ion beam mutagenesis include high survival rates, high levels of mutation induction and various other variations (Yazid and Muryono 2000).

After addition of the scores obtained from a total of 48 lines selected i.e. 27 lines from Carbon ion beam irradiation and 21 lines from Argon ion beam irradiation (Table 2), and compared with the best total score (above 37.97), 10 lines were selected with the total score above the best score (Table 3), namely 5 lines in Carbon treatment (PL-C-4.1-17, PL-C-20.1-1, PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44) and 5 in the Argon ion beam irradiation treatment (PL-A-6.5-4, PL-A-6.5-6, PL-A-6.5-15, PL-A-6.5-23, and PL-A-6.5-29).

The selection of character selection in the tested population is based on components that have a close relationship with the character of production. Thus, the selection of populations of diploid rice lines can use the character of the generative phase of plant height, a number of pithy grain per panicle, total grain per panicle, weight of 1,000 rice grain and character of grain yield per hill. Aryana (2007) explained the character of number of tillers, number of rice grain, total grain count per panicle, and weight of 1,000 grains can be used as character selection, whereas Safitri *et al.* (2011) explained the character of the number of productive tillers, number of rice grain per panicle, and the weight of 1,000 grains can be used as selection.

In Figure 2 and Table 1 above, it can be seen that at the 90% similarity level, based on the data of the production components of the 10 selected lines, 5 groups were obtained, namely Group I consisting of 1 strain, namely PL-C-4.1-17. Group II consisted of 2 subgroups consisting of 3 lines namely PL-C-20.5-44, PL-C-20.5-49, and PL-C-20.11-44. Group III consisted of 1 strain, namely PL-C-20.1-1. Group IV consists of 2 lines, namely PL-A-6.5-4 and PL-A-6.5-15. Group V consists of 2 subgroups consisting of 3 lines, namely PL-A-6.5-6, PL-A-6.5-23, and PL-A-6.5-29.

Based on the level of similarity it can be seen that the PL-C-20.5-49 and PL-C-20.11-44 and PL-A-6.5-23 and PL-A-6.5-29 lines have very close kinship relationships. The line has a 95% similarity level. This shows that lines that are closely related will have a close genetic distance. This is consistent with Lee's opinion (Lee 1998), who said that closely related individuals will have a close genetic distance, whereas if they are distant relatives they will have a far genetic distance. The results of the grouping of 10 selected lines consisting of 5 groups showed that the lines in the same group generally had almost the same number of lines. Erlina *et al.* (2011) suggests that the same cluster is characterized by almost the same genotype name. This shows that the genotypes are formed from the same population so that the level of kinship is closer.

5. Conclusion

The result of this study showed that treatments with ion beam irradiation either by using Carbon ion as well as as Argon ion are capable of producing Toraja local red rice mutant genotype with a shorter harvesting age (mid-early maturity) compared to their parent control. There is an acceleration of 16-17 days or about 10% from the harvesting age of the parents. Out of 48 total selected lines, 10 lines (five of each Carbon and Argon ion beam irradiated mutants lines) meet the selection criteria of mid-early maturity, high posture and many productive tillers.

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