Selection of high-yield rice mutant induced by heavy-ion beam irradiation

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Improvement in productivity is the main objective of rice breeding. Rice grain yield is determined based on three components: the number of panicles per plant, number of grains per panicle, and grain weight. Among them, the grain weight is a stable varietal character and effective target trait for the mutation breeding of highyield rice.

We performed mutant selection for achieving highyield rice from M_2 populations obtained by heavy-ion beams irradiation. M_2 seeds used in this study were derived from irradiated dry rice seeds of 'Nipponbare' (*Oriza Sativa* L.) with C-ion, Ar-ion, and Fe-ion. Liner energy transfer (LET) and irradiation doses are summarized in Table 1. These conditions were the most efficient with the highest mutation rate at each LET in our previous study.^{1,2})

There were 790 M_1 lines in total and ten plants from each M_1 line were planted in the paddy field. In our previous study on rice, we isolated various mutants of the flowering period, plant height, leaf color, tillering, and weakness by visual evaluation in the paddy field. However, the identification of grain size in the field for mutant selection is not easy and is inefficient. Panicle weight reflects the grain size and fertile grain number, which are two major components of the yield. We adopted the method of measuring the weight of the main panicle from each M_1 plant to select the candidates of the mutant. First, some lines were excluded from panicle weight selection when segregated mutations were observed in the field. We selected 18 candidates based on panicle weight from 462 M_1 lines (Table 2).

Next year, we conducted a yield survey on a small scale to confirm their traits and narrow down the candidates. Thirty M_3 seedlings of each candidate line were planted in a plot (0.9 m wide and 3 m long). 'Nipponbare' was used as control. To avoid the border effect, 8 plants from the inner plot (2.4 m²) were collected. The panicle weight, spikelet number, seed fertility, and grain weight were investigated on the main culms. The grains from the line with the higher weight were dehulled; the

Table 1. Irradiation conditions of heavy-ion beams.

Ion	Energy	LET	Dose
	(MeV/nucleon)	$({ m keV}/{ m \mu m})$	(Gy)
С	135	30	175
Ar (184)	160	184	20
Ar (290)	95	290	10
Fe	90	650	20

Table 2. Number of lines selected in each generation.

Ion	Planted	Surveyed	Candidates	
1011	M_1 line	M_1 line	M_2	M_3
С	200	113	5	2
Ar (184)	200	121	4	0
Ar (290)	200	122	4	1
Fe	190	106	5	2
Total	790	462	18	5

grain width, grain length, and grain thickness were measured using a rice analyzer (RGQ1 10B, SATAKE). However, some lines which had low fertility or low number of grains were not included in the grain size measurement even though their grain weight were high, because the trade-off relationship between the single grain size and grain number should be considered. In addition, lines with large individual differences were excluded. Finally, five lines were selected based on their large grain size. There was a trend for long grains in all 5 candidates (Table 3). Among them, Ar-39 achieved the best results in most components. Our previous study succeeded in discovering novel gene: LIN1 using the long grain mutant induced by Ar-ion irradiation.³⁾ The genetic analysis of these candidates to reveal relevant genes is in progress while continuing repeated yield surveys on a large scale.

References

- 1) Y. Hayashi et al., RIKEN Accel. Prog. Rep. 50, 27 (2017).
- 2) Y. Hayashi et al., RIKEN Accel. Prog. Rep. 51, 238 (2018).
- 3) R. Morita *et al.*, Molecular Breeding **39**, 135 (2019).

Table 3. Mean \pm standard error of yield-related characters and grain size for selected M₃ lines.

Line	Panicle weight	Grain number	Seed fertility	Grain weight	Grain size (mm)		
	(g/one panicle)	(/panicle)	(%)	(g/1,000 grain)	Length	Width	Thickness
'Nipponbare	2.95 ± 0.07	130.4 ± 5.58	87.3 ± 1.86	24.2 ± 0.26	5.02 ± 0.03	2.81 ± 0.02	1.88 ± 0.01
C-98	3.32 ± 0.14	135.6 ± 5.19	85.1 ± 1.44	25.0 ± 0.21	5.08 ± 0.02	2.77 ± 0.02	1.86 ± 0.01
C-190	2.80 ± 0.06	118.0 ± 2.78	83.2 ± 1.70	25.8 ± 0.13	5.18 ± 0.02	2.85 ± 0.02	1.89 ± 0.01
Ar-39	3.63 ± 0.14	148.8 ± 5.70	85.4 ± 0.89	25.7 ± 0.23	5.17 ± 0.02	2.89 ± 0.02	1.89 ± 0.01
Fe-134	3.10 ± 0.11	129.0 ± 3.91	83.5 ± 1.69	25.5 ± 0.17	5.12 ± 0.02	2.85 ± 0.02	1.88 ± 0.01
Fe-153	3.03 ± 0.10	126.6 ± 4.50	83.5 ± 1.48	25.2 ± 0.27	5.08 ± 0.02	2.85 ± 0.02	1.89 ± 0.01

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