## Approach toward isolating early-heading mutants from Tana Toraja local aromatic rice "Pare Bau" irradiated with argonand carbon-ion beams

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Tana Toraja is a mountainous area in the northern part of South Sulawesi, Indonesia. The area has been known as an aromatic-rice-producing area.<sup>1)</sup> The aromatic rice cultivar "Pare Bau" has good grain quality and fragrant aroma, making it relatively expensive in the market, but it has several disadvantages including late heading dates and long stems for modern farming systems. The heading date can be determined by the basic vegetative phase (BVP) and photoperiod sensitive phase (PSP).<sup>2)</sup> Indonesian rice cultivated in mountain areas has a relatively short PSP and relatively long BVP. This study aims to isolate early heading mutant lines induced by heavy-ion-beam irradiation.

Dry seeds of Tana Toraja local aromatic rice cultivar (*Oryza sativa* L. "Pare Bau") were irradiated with carbon ions (30 keV/ $\mu$ m) and argon ions (300 keV/ $\mu$ m) in the RI-beam factory, RIKEN Nishina Center, Japan. Owing to a limited range of Ar-ion beams, seeds for Ar-ion irradiation needed to be packed in a single layer. For this purpose, a sponge was bedded as a bottom plate in a plastic rectangular box (75 × 50 × 18 mm). The doses of Ar-ion and C-ion irradiation were 10 and 150 Gy, respectively.<sup>3,4)</sup> We investigated the heading dates of Pare Bau lines irradiated with Ar and C ions (PB-A and PB-C, respectively).

In May 2016,  $M_1$  seeds (1000 seeds of PB-A and 1000 seeds of PB-C) were germinated and sown in seedling pots and grown in a greenhouse for three weeks. The germination rates of  $M_1$  seeds were 53% for PB-C and 49% for PB-A, while that of the wild-type control was 70%. The seedlings (94 PB-A and 79 PB-C plants) were transplanted in the paddy fields of Hasanuddin University (S: 5°7′53.50″ E: 119°28′57.97″; 20 m above sea level) in June 2016. Owing to poor establishment of seedlings, only small amounts of  $M_1$  plants (13 PB-A and 13 PB-C plants) set more than 50 seeds in November 2016. In June 2017, 50 M<sub>2</sub> seedlings per line were transplanted in the paddy field of mountainous Enrekang Regency (S: 3°19'47.44" E: 119°50'1.57"; 650 m above sea level) neighboring Tana Toraja Regency. Based on heading dates at least one week earlier (<119 days) than those of the wild-type control (>125 days), we selected 82 PB-A and 26 PB-C plants as mutant candidates. From these M<sub>2</sub> plants, we sampled M<sub>3</sub> seeds with relatively large grain weights per



Fig. 1. Phenotypes of early-heading mutants induced by heavy-ion-beam irradiation in the paddy field of Enrekang Regency.

panicle from 18 lines of PB-A and one line of PB-C in October 2017. In May 2018, 50  $M_3$  seedlings per line were transplanted in the paddy field of Enrekang Regency. Heading dates of the M<sub>3</sub> individual plants and wild-type control plants were recorded for the first 25 plants showing relatively early heading. The days to heading of the wild-type control was 126 to 130 days with an average of 128 days, while those of the following nine lines were 108 to 126 days with an average of 117 to 121 days, which were over one week less than that of the wild-type control (Fig. 1). These lines are PB-A.5.3.45, PB-A.6.1.9, PB-A.7.1.30, PB-A.12.2.4, PB-A.12.2.12, PB-A.14.2.14, PB-A.14.3.1, PB-A.14.4.3, and PB-C.20.1.49. We expect these lines to contain mutant candidates. Genetic analysis will be performed for  $M_4$  lines and their hybrids with the wild type to investigate the mutated genes responsible for early heading.

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