## Production of mutant line with early flowering at a low temperature in spray-type chrysanthemum cultivar induced by C-ion beam irradiation

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Chrysanthemum (Dendranthema grandiflorum) is one of the major cut flowers. Saga Prefecture has developed 5 original cultivars of spray-type chrysanthemum by crossbreeding in 2011. However, the white-flower cultivar 'Saga SK3' showed delayed flowering under a low temperature. Recently, heavy-ion beam irradiation was reported as an effective method for breeding chrysanthemum with early flowering at a low temperature<sup>1</sup>). Therefore, in this study, we aimed to develop earlyflowering mutants of 'Saga SK3' by heavy-ion beam irradiation and selected an earlier-flowering mutant line.

In 2012, 6-cm-long cuttings without leaves of 'Saga SK3' were irradiated with C-ion beams (LET 22.6 keV/ $\mu$ m) at doses of 3 and 6 Gy. After irradiation, these cuttings were cultured in cell trays for rooting in a greenhouse. The survival rates of irradiated plants were all 100% (Table 1). The terminal buds of the cuttings were pinched off, and new shoots were allowed to grow from the axillary buds. This procedure was repeated twice or thrice for isolating chimeric structures. Then, shoots elongated after pinching were planted in a greenhouse, and these plants were grown in the January flowering cropping system in 2013 under low temperature controlled at a minimum of 13°C. 'Saga SK3' flowered 63 days after lighting stop. Individual selections from these plants flowered until 56 days after lighting stop by the standard method in Saga Prefecture. We found that 9 individuals flowered until 56 days after lighting stop from the 464 plants obtained after 6-Gy irradiation (Table 1), and we selected 5 individuals from them. On the other hand, no plant flowered until 56 days after lighting stop at a dose of 3 Gy. These results suggest that the minimum effective dose of C-ion beam irradiation using cuttings in chrysanthemum for inducing early-flowering mutation is more than 3 Gy.

In 2014 and 2015, we planted each line in a greenhouse for line selection. In 2014, we planted the 5 lines in the February flowering cropping system under low temperature controlled at a minimum of  $12^{\circ}$ C. Although 'Saga SK3' flowered 92 days after lighting stop, we selected two lines that had flowering dates earlier by 32 and 37 days compared to that of 'Saga SK3'. In 2015, we selected one line, 24-SK3-i6-4, from the two lines in the March flowering cropping system under low temperature controlled at a minimum of  $13^{\circ}$ C. The flowering date of this line was 7 days earlier than that of 'Saga SK3' (Table 2, Fig. 1). Furthermore, this line was almost not different from 'Saga SK3' in terms of the flower size, petal color, and plant height (Table 2, Fig. 2).

As a result, we selected one suitable early-flowering mutant line, 24-SK3-i6-4, which seems to retain the original flower characteristics of 'Saga SK3' after 6-Gy irradiation with C-ion beams. We are planning to investigate the characteristics of the mutant line further for commercial production in our prefecture.

Table 1. Ef	Table 1. Effects of C-ion beam irradiation on induction of early-flowering mutation <sup>z</sup> .										
Beam dose (Gy)	No. of cuttings irradiated	Survival rate (%)	No. of plants investigated	No. of flowering plants <sup>y</sup>	No. of early- flowering mutants <sup>x</sup>	No. of selected plants					
0	20	100	30	24	0	-					
3	60	100	489	271	0	-					
6	60	100	464	271	9	5					

<sup>z</sup> Planting date: 2012/10/24. Lighting stop date: 2012/12/5. Minimum temperature controlled at 13°C.

<sup>y</sup> No. of plants flowered until 70 days after lighting stop.

<sup>x</sup>No. of plants flowered until 56 days after lighting stop.

Table 2. Characteristics of selected line in March flowering croping system in 201	15 <sup>z</sup> .
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Line or cultivar	Flowering date	Days to flowering in short days	Plant height (cm)	Weight of 85 cm cut flower (g)	Number of flowers
24-SK3-i6-4	Mar. 23	56	$119.7\pm1.0^{\rm y}$	$92.7\pm12.1$	$18.3\pm2.8$
Saga SK3	Mar. 30	63	$106.4 \pm 1.6$	83.6 ± 7.1	15.6 ± 1.7

 $^z$  Planting date: 2014/11/21. Lighting stop date: 2015/1/26. Minimum temperature controlled at 13°C .  $^y$  Mean  $\pm$  SE

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Saga SK3

24-SK3-i6-4

Fig. 1. Photographs of original cultivar 'Saga SK3' and selected line, 24-SK3-i6-4, on 23 Mar. in 2015.





Saga SK3

Fig. 2. Flower of 'Saga SK3' and selected line, 24-SK3-i6-4.

## Reference

1) Ueno et al., Hort. Res. (Japan) 12, 245 (2013).

<sup>\*2</sup> Saga Prefectural Higashimatsuura Agriculture Extention Center