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Effects of Cultivation Methods on Yield and Essential Oils of *Chrysanthemum indicum* L. (Gamgug)

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ABSTRACT: ***Chrysanthemum indicum* L. (Gamgug) has been examined to study their flowering habits, yields and bioactive compounds under different planting densities and mowing dates. The planting density experiment revealed a significantly increasing stem diameter, number of flowers and branches with decreasing plant density in the 100 cm x 30 cm and 130 cm x 30 cm treatments as compared to 70 cm x 30 cm treatments, but not plant height, leaf and flower width. On the other hand, the mowing date experiment showed that growth characteristics of plants were similar to the control plants (not mowing) and June 20 treatment, but July 20 treatments had significantly smaller than the control. The weights (g plant^{-1}) of dry flowers were affected by the planting density and mowing date. The flower yield of 586 kg ha^{-1} obtained at 100 cm × 30 cm density was 11% and 22% higher than that of 120 cm × 30 cm and 70 cm x 30 cm treatments, respectively. The yield of dry flowers in the control and June 20 mowing date ranged 495-508 kg ha^{-1}, which is 40-42% higher than the yield in the July 20 treatments. The amount of essential oil (g plant^{-1}) in medically valuable flowerheads of *C. indicum* L. was statistically different between mowing dates but not among planting densities. The study showed that planting density and the mowing date could increase yields of flowerheads. An optimum planting density of 100 cm x 30 cm and mowing date of on or before June 20 is recommended for *C. indicum* L.**

Key Words: *C. indicum* L., growth, yield, planting density, mowing date, essential oil

INTRODUCTION

Wild Chrysanthemum has been used for natural medicine for food as well as a gardening flower since the ancient times[1)]. *Chrysanthemum* spp., a perennial herb which belongs to the Compositae, blooms from October to November throughout Korea with yellow flowered capitulums[2)]. In particular, *Chrysanthemum indicum* L. (Gamgug) (hereafter, *C. indicum* L.) is known to have several biological actions, such as headache and dizziness medicine peripheral analgesic properties, lowering blood pressure[3)], anticancer and antibacterial activities[4)] as well as anti-inflammatory activities[6)]. These are due to significant amount of terpene compounds, especially sesquiterpenes, and their derivates[4)]. These immediate constituents were well known as phenolic compounds; luteolin, apigenin, luteolin 7-O-β-D-glucoside, and as sesquiterpene lactone; cumambrin A, cumambrin B, arteglasin A and angeloyljadin[5)]. Several studies on the volatile flavor components have been reported[7-9)]. Chang and Kim[7)] reported that the yield of *C. indicum* L. flower oil contained 2.0% (w/w) of golden yellow color, and constituted 63 volatile flavor components, which make up 89.28% of the total aroma composition. In recent years, demands for flowerheads of *C. indicum* L. as medicinal plants have increased with growing concerns for health-improving foods. But these amounts, which are taken from the wild in mountainous areas, are not enough to satisfy such demands until now. However, the wild *C. indicum* L. supply should be supplemented by cultivating the crop employing different methods to fulfill the

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increasing demand for a high quality flower. Although some cultivation system studies in *Chrysanthemum* spp. had been reported, until recently, the results are limited. In cultivation methods, planting densities and mowing dates have a profound effect on factors affecting crop productivity such as absorption of light, water, and nutrients. Although Song et al.[10] has reported that pinching on May 20 increased the number of flowers per plant in *C. indicum* L. as compared with June, July and August pinching, until now, very few studies on plant densities and mowing dates in *C. indicum* L. were conducted. Therefore, various studies on cultivation methods in flowerheads of *C. indicum* L. are needed. This paper investigated the effects of planting densities and mowing dates on the growth, yield and essential oil contents to develop cultivation methods for high quality of flowerheads of *C. indicum* L.

MATERIALS AND METHODS

Field experiments

A field experiment was conducted in 2001 at the Gyeongsang National University. The experimental methods reported similar with *C. boreale* M.[12]. Soil texture was silt loam (SL) with 3.2 g kg^{-1} of OM, 3.2 and 0.97 $cmol^{(+)}$ kg^{-1} of exchangeable Ca and K, and a pH of 5.3. The same levels of 150-80-80 kg $N\text{-}P_2O_5\text{-}K_2O$ ha^{-1} chemical fertilizers were applied to all treatments. The fertilizers were broadcasted in the field one week before the seedlings. All phosphate was applied as basal fertilizer; nitrogen and potassium were splitted, 70% as basal and 30% applied around 25 days before flowering. Plots were arranged in a completely randomized design and each treatment was carried out in triplicate. The seedlings of *C. Indicum* L were transplanted on May 25, 2001, and the field was mulched with black vinyl for early weed control. The three examined planting densities were 70 cm × 30 cm, 100 cm × 30 cm and 130 cm × 30 cm. Plants in mowing dates experiment were mowed at 15 cm above soil surface considering three dates-treatments: no mowing (control), June 20, and July 20. The flower yield was determined at full bloom stage on October 20, 2001 and the picked fresh flowers before full blooming stage (October 10-15) were put immediately into deep-freezer (-70℃) for determination of essential oils contents. The growth and yield characteristics were investigated by the RDA methods[13].

Analysis of essential oils

Essential oil contents of *C. indicim* L. flowerheads were determined with simultaneous distillation extraction (SDE) apparatus, using a modification of various methods[12,14-15]. Briefly, diethyl ether extracts were separated with a Supelcowax 10 fused silica capillary column (length 60 m x inside diameter 0.32 mm x film thickness 0.25 um on a Hewlett-Packard 5880 gas chromatograph equipped with a FID detector. The operating conditions of GC were as follows: oven temperature was held at 50℃ for 5 min, than programmed to increase at 3℃ min^{-1} to 225℃ where it was maintained for 30 min. The injector and ion detector temperatures were 225 and 230℃, respectively. The flow rate of the carrier gas, nitrogen, was 1.86 ml min^{-1}. Peak areas were measured by electronic integration. The relative amounts of the individual components are based on the peak areas. MS spectra were determined by GC-MSD (HP5890 and HP5970, Hewlett-Packard, USA), with the electron impact source operating at 70 eV and 240℃. The injector and ion source detector temperatures were the same as above. Terpene compounds were identified by computer matching of the mass spectra and confirmed by GC retention times.

Statistical analysis

All data were analyzed statistically by the analysis of variance using CoStat software (CoHort Software, Monterey, USA). Yields and essential oil contents were tested in the experiment using a randomized complete block model with three replications. Mean comparisons were determined using an ANOVA protected least significant difference (LSD) ($P<0.05$) test.

RESULTS AND DISCUSSION

Planting density

The effects of planting density on growth characteristics and dry matter of *C. indicum* L. are presented in Table 1. The number of 1^{st} and 2^{nd} lateral branches in the 100 cm x 30 cm and 130 cm x 30 cm plating densities increases with decreasing planting density

Table 1. Effect of planting density on growth characteristics and biomass of *C. indicum* L.

Treatment	Plant height (cm)	No. of lateral branches		Leaf width (cm)	Stem diameter (cm)	No. flower branch	Flower width (cm)	Number of flower	Blooming time	Dry weight of flower (g plant^{-1})
		1st	2nd							
70 cm x 30 cm	125	18.8	99. 7	43.6	1.47	12.6	114	1989	10.22	9.5
100 cm x 30 cm	131	24.0	137.3	51.7	1.75	15.7	127	2503	10.22	17.6
130 cm x 30 cm	128	25.6	146.0	55.3	1.81	16.5	125	2846	10.22	17.9
$LSD_{0.05}$	ns[1)]	2.45	15.6	ns	0.26	3.11	ns	268	ns	1.48

ns[1)]: not significant different
Means separation within columns by LSD (p=0.05, n=3)

compared to the 70 cm x 30 cm treatment. This result is similar to those of Lee et al.[16)] who reported that the planting density in *C. boreale* M. had significant effect on the number of lateral branches. Furthermore, stem diameter, number of flower number and branches also significantly increased with decreasing planting density compared to 70 cm x 30 cm treatment, but not plant height, leaf width and flower width. These results are similar with those of *C. morifolium* plant investigated earlier[17)]. On the other hand, plant spacing significantly influenced the number of branches and number of flowers per plant. Blooming time in all treatments was mainly on October 22. The weights (g per plant) of dry flowers were affected by planting density. Less dense planting density tends to increase the weight of dry matter of shoot parts. The dry weight (17.6 g per plant) of flower in the 100 cm x 30 cm treatment was similar to 17.9 g per plant in the 130 cm x 30 cm treatment, but significantly higher than the dry weight of flower in the 70 cm x 30 cm. This means that 70 cm x 30 cm treatment has inhibited air flow by the high planting density. As a result, the dry weight of flower heads and leaves was seriously decreased caused by plant diseases such as powder and downy mildew, which were observed to be more prevalent in the densely spaced crop. As shown in Fig. 1, the yield (kg ha^{-1}) of dry flowers in the 100 cm × 30 cm density was greater than that in the 70 cm x 30 cm or 130 cm x 30 cm treatments. The weight of dry flowers at the 100 cm × 30 cm density was 586 kg ha^{-1}, which is 11 and 22% higher as compared with those of the 120 cm x 30 cm and 70 cm x 30 cm treatments, respectively. The yield advantage of a 100 cm × 30 cm planting density over the other can be primarily attributed to the greater number of branches per plant. Therefore, the optimum planting

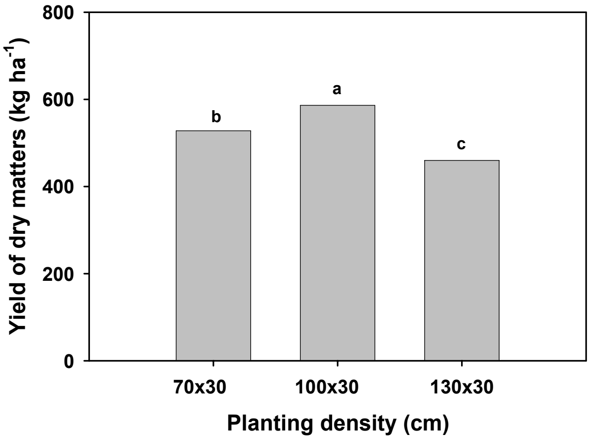

Fig. 1. Dry matter yield of *C. indicum* L. flowerheads as affected by planting density. Means with the same letter are not significantly different by LSD (p=0.05, n=3)

density of *C. indicum* L. plant was considered to be a 100 cm × 30 cm. *C. indicum* L. flower heads oil have physicochemical and bio-functional properties which are useful and important materials for industrial and medicinal purposes. The yield of essential oils of *C. indicum* L. flowerheads, as shown in Fig. 2, was affected by planting density. The 100 cm × 30 cm density gave an essential oils yield of 6.5 kg ha^{-1}, which is 15 and 27% more as compared with that of the 70 cm × 30 cm and 120 cm × 30 cm treatments. The yield of essential oil contents in medically valuable flowerheads of *C. indicum* L., along with their relative percentage, as affected by planting density, are given in Table 2. The major components of the essential oils isolated were; α-pinene, 1,8-cineol, chrysanthenone, campor, borneol, β-elemene, germacrene -D, α-curcumene, zingiberene and β-sisabolene, and occupied approximately 47% of peak area by GC-MS analyzer. Similar results were reported by Chang and Kim[7)] that essential oils of *C. indicum* L. flower constituted 63 volatile flavor components, which made up 89.28% of the total aroma composition. The total peak area of all treatments was not statistically different.

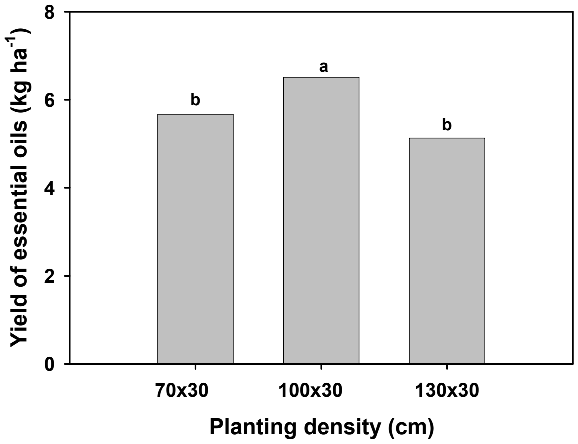

Fig. 2. Yield of essential oils of *C. indicum* L. flowerheads as affected by planting density. Means with the same letter are not significantly different by LSD (*p*=0.05, *n*=3)

Mowing date

The mean values of growth characteristics and dry weight (g per plant) of *C. indicum* L. flowerheads as affected by mowing date are shown in Table 3. Results showed that mowing date has significant influenced on plant height, number of lateral branches, leaf width, stem diameter, number of flower branches, flower width, number of flowers, and blooming time. Similar growth characteristics were observed between the control plants (not mowed) and the mowing at June 20. However, mowing at July 20 had significantly smaller plant height, stem diameter and number of lateral branches than the control. These results are similar to that Lee et al.[16] who reported that mowing date in *C. boreale* M. had

Table 2. Major components (%) of essential oil of *C. indicum* L. flowerheads as affected by planting density.

Compounds	60 cm x 30 cm		100 cm x 30 cm		120 cm x 30 cm	
	Mean	SD[1)]	Mean	SD	Mean	SD
α-pinene	8.13	0.089	8.23	0.086	8.12	0.077
1,8-cineol	7.31	0.088	6.16	0.074	6.75	0.075
chrysanthenone	10.80	0.096	10.00	0.087	11.26	0.082
campor	1.64	0.039	1.66	0.032	1.02	0.022
borneol	2.33	0.045	2.54	0.034	2.55	0.031
β-elemene	2.75	0.039	2.26	0.028	2.13	0.035
germacrene D	5.05	0.065	7.02	0.058	6.09	0.069
α-curcumene	3.85	0.032	3.56	0.036	3.78	0.022
zingiberene	2.70	0.036	2.14	0.029	2.09	0.025
β-sisabolene	3.25	0.037	3.71	0.041	4.40	0.021
Total (%)	47.8a	-	47.3a	-	48.2a	-

SD[1)]: standard deviation (*n*=3)
Total values (%) followed by the same letter are not significantly different using $LSD_{0.05}$ protected ANOVA

Table 3. Effect of mowing date on growth characteristics and biomass of *C. indicum* L.

Treatment	Plant height (cm)	No. of lateral branches		Leaf width (cm)	Stem diameter (cm)	No. of flower branch	Flower width (cm)	No. of flower	Blooming time	Dry weight of flower (g plant^{-1})
		1st	2nd							
Control[1)]	117	21.0	124.7	52.0	1.63	14.6	118	2636	10.22	14.9
June 20	107	18.7	121.3	49.7	1.55	13.7	107	2569	10.22	15.2
July 20	74	10.3	81.0	41.0	1.03	9.4	83	1800	10.23	8.8
$LSD_{0.05}$	17.2	4.3	19.1	4.8	0.19	2.9	18.7	522	ns[2)]	2.4

Control[1)]: Not mowed
ns[2)]: not significant
Means separation within columns by LSD (*p*=0.05, *n*=3)

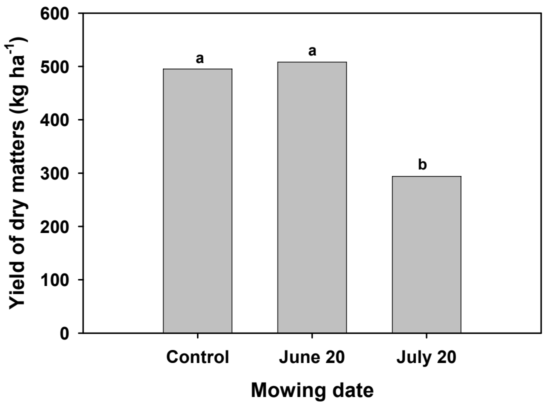

Fig. 3. Dry matter yield of *C. indicum* L. flowerheads as affected by mowing date. Means with the same letter are not significantly different by LSD (*p*=0.05, *n*=3)

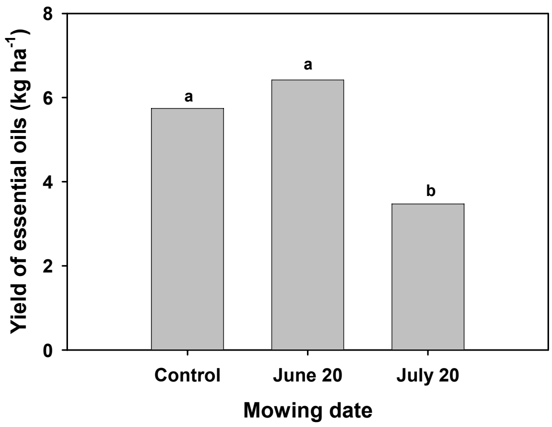

Fig. 4. Yield of essential oils of *C. indicum* L. flowerheads as affected by mowing date. Means with the same letter are not significantly different by LSD (*p*=0.05, *n*=3)

Table 4. Major component contents (%) of essential oil of *C. indicum* L. flowerheads as affected by mowing date

Compounds	Control (not mowed)		June 20		July 20	
	Mean	SD[1)]	Mean	SD	Mean	SD
α-pinene	8.28	0.068	8.06	0.074	7.56	0.079
1,8-cineol	6.88	0.072	6.21	0.075	5.39	0.074
chrysanthenone	10.50	0.075	11.42	0.086	10.88	0.098
campor	1.81	0.012	2.01	0.034	2.47	0.038
borneol	2.39	0.025	2.33	0.022	2.64	0.031
β-elemene	2.02	0.026	1.98	0.015	2.09	0.035
germacrene D	7.55	0.053	6.68	0.056	6.25	0.023
α-curcumene	2.87	0.036	3.51	0.026	2.15	0.025
zingiberene	2.65	0.021	3.06	0.032	2.94	0.029
β-sisabolene	2.20	0.018	2.17	0.038	1.86	0.016
Total (%)	47.2a	-	47.4a	-	44.2b	-

SD[1)]: standard deviation (*n*=3)
Total values (%) followed by the same letter are not significantly different using $LSD_{0.05}$ protected ANOVA

significant effect on all of treatments. Weights (g plant^{-1}) of dry flowers were affected by the mowing date. Especially, plants mowed on July 20 treatments are significantly smaller than those in the control and mowing at June 20. The accumulated dry weights of shoot were similar among all treatments. The weight of dry flowers in the control and mowing at June 20 reached 14.9 and 15.2 kg ha^{-1}, respectively, a significant increase of approximately 42% as compared with the mowing at July 20. As shown in Fig. 3, the 495-508 kg ha^{-1} yield of dry flowers in the control and mowing at June 20 was 40-42% higher than that of the mowing at July 20. Therefore, the optimum range of mowing date for *C. indicum* L. plant was on or before June 20. The yield of essential oils of *C. indicum* L. flowerheads as affected by mowing date was presented in Fig. 4. The none-mowing and mowing at June 20 gave statistically identical yield of essential oils in the range of 5.74-6.42 kg ha^{-1}, which were about 53-71% higher than those obtained in the mowing at July 20. Table 4 showed that the changes in essential oil contents of the medically valuable flowerhead of *C. indicum* L. were significantly affected by mowing date.

CONCLUSION

This study investigated the effects of planting densities and mowing dates on the growth, yield and essential oil contents of *C. indicum* L. in order to develop the cultivation methods for producing high quality flowerheads. Planting density and mowing date can significantly influence the yield of flowerheads. An optimum planting density of 100 cm x 30 cm and mowing on or before June 20 is recommended for the commercial cultivation of high quality *C. indicum* L.

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