

Improvement of White Clover (*Trifolium repens* L.) Seed Production in Japan by Transplanting and Mulching

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ABSTRACT

The effect of establishment method on the seed yield of white clover was investigated using direct sowing in autumn, transplanting in autumn and transplanting in spring. The effects of polyethylene mulching were also tested for the direct sowing and transplanting in autumn treatments. The percentage of clover plants which survived over the winter and seed yield were higher in the autumn transplanting treatment than in the autumn direct sowing treatment. However the greatest seed yield (28 g m⁻²) was obtained in the spring transplanting treatment, because the number of flower heads per plant was greater and no plants had been killed during the winter. Seedling growth was improved by mulching for the direct sowing treatment, but for transplanting, stolons were inhibited in their growth due to shading, and seed yield was therefore not improved by mulching.

Additional index words: polyethylene mulching, seed yield, direct sowing, transplanting, stolon growth, flower heads

INTRODUCTION

To encourage the use of a new herbage cultivar and to have a constant supply of seed, it is very important to obtain a stable seed yield of that cultivar. White clover, however, has been regarded as one of the most difficult herbage species from which to obtain a good seed yield. Its yield is apt to be small in Japan and unstable even in traditional seed production countries (Rolston, 1996).

White clover seeds are smaller than those of many other herbage species and their growth during the seedling stage can be slow. Therefore, white clover seedlings can be damaged by competition from weeds or, when sowing is late, by chilling during winter which in turn results in low seed yield (Pasumarty, Higuchi and Murata, 1996). In Nagano, where if snow cover is absent, the winter cold is very severe (Table 1), a delay in sowing causes low seedling survival during winter.

Flower heads of white clover are formed with the extension of stolons. High plant density will reduce the number of flower heads per stolon and seed production is decreased (Zaleski, 1961, 1964; Sugita, Fukuoka, Wakamatsu and Miura, 1982; Clifford, 1985). But stands established from low sowing rates are difficult to keep in good condition, especially in their early stages (Sugita, Fukuoka, Wakamatsu and Miura, 1984) and become uneven if death of seedlings occurs.

Under these conditions, raising of seedlings in pots under intensive care and transplanting them to the field after they have acquired resistance against stresses may be an effective method to solve these problems. Acceleration of growth during the seedling stage could also be achieved by raising soil temperature with mulching. In this study, we aimed to clarify the effectiveness of transplanting and/or mulching on white clover seed yield.

MATERIALS AND METHODS

White clover cultivar Makibashiro, bred at the Tohoku National Agricultural Experiment Station, was used for the experiment. The five treatments were autumn direct sown without mulching (A-DS) and with mulching (A-DSM), autumn transplanting without mulching (A-TP) and with mulching (A-TPM), and spring transplanting without mulching (S-TP). Sowing and transplanting dates are given in Table 2. The trial was conducted at the Nagano Station of the National Livestock Breeding Center (lat. 36°N, long. 139°E, alt. 700m), where mean temperature and annual rainfall is 10.2°C and 870 mm respectively. The soil type is a volcanic acid soil.

The seeds were sown in 3 cm diameter paper pots for A-TP and A-TPM and in 7.5 cm diameter vinyl pots for S-TP. The seedlings were grown in a glasshouse which was heated for S-TP. The mean temperature while raising the seedlings was about 25°C for A-TP and A-TPM and about 15°C for S-TP. The seedlings were watered regularly. Day length was about 13 hours for A-TP and A-TPM and 11.5 hours for S-TP under natural daylight conditions.

For direct sowing, about ten seeds were sown in one spot and seedlings were thinned to one plant per spot after emergence.

Chemical fertiliser (15%N, 15%P₂O₅, 15%K₂O) was applied at the rate of 2g plant⁻¹ except for S-TP on 29 September 1993. Superphosphate (18% P), fused magnesium phosphate (20% P, 13%Mg and 0.5%B) and KCl (60%K) were applied at the rate of 1.48, 1.48 and 1.0 kg ha⁻¹ respectively for all treatments on 11 April 1994. Honey bees were released as pollinators from 13 May to the time of harvest.

Plot size was 13.5m² (4.5mx3m) for each treatment. For all treatments, 30 plants per plot were grown as spaced plants (75cm x 50cm apart in a rectangular pattern). Measurements

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Table 1: Weather data for each month over the experimental period and 10 year average.

Month	Mean temperature (°C)		Rainfall (mm)		Sunshine (h)	
	1993/94	10 yr av	1993/94	10 yr av	1993/94	10 yr av
Oct '93	10.9	11.9	93	88	157	125
Nov	7.9	5.8	80	34	164	162
Dec	1.1	0.8	67	21	175	178
Jan '94	-2.5	-2.0	23	22	183	161
Feb	-1.5	-2.0	8	31	177	148
Mar	1.1	1.8	30	55	186	170
Apr	10.1	8.3	24	55	199	180
May	14.5	13.6	50	81	161	150
June	17.9	17.5	81	129	97	96
July	24.8	21.4	25	164	154	98

Table 2: Design of the experiment.

Treatment	Abbreviation	Time of Sowing	Time of Transplanting
Autumn direct sowing without mulching	A-DS	30 Sept '93	-
Autumn direct sowing with mulching	A-DSM	30 Sept '93	-
Autumn transplanting without mulching	A-TP	10 Aug '93	30 Sept '93
Autumn transplanting with mulching	A-TPM	10 Aug '93	30 Sept '93
Spring transplanting without mulching	S-TP	7 Feb '94	5 Apr '94

Table 3. Effect of establishment method and mulching on the size of individual plants and time of flowering.

Treatment ¹	Size of plants ² (cm)		Time of flowering ³
	3 Dec '93	3 June '94	
A-DS	4	3.0 ^{d5}	24 June
A-DSM	4	28.4 ^c	1 June
A-TP	13.7 ^{a5}	60.1 ^a	16 May
A-TPM	9.1 ^b	49.4 ^b	16 May
S-TP	-	35.6 ^c	1 June

¹ Refer to abbreviations in Table 2.

² Diameters of individual plants were measured diagonally and averaged.

³ The time of flowering for each treatment was determined when half of the surviving plants had opened 3 or more florets on at least one head.

⁴ Could not be measured because no stolons present.

⁵ Means within a column followed by the same letter are not significantly different at P=0.01.

Table 4. Clover survival (%) in early winter and in spring

Treatment ¹	3 December 1993	16 May 1994
A-DS	56	6
A-DSM	92	50
A-TP	100	89
A-TPM	100	86
S-TP	-	100

¹ Refer to abbreviations in Table 2.

Table 5: Effect of establishment method and mulching on the number of white clover flower heads per plant and seed yield in each treatment.

Treatment ¹	No of flower heads per plant	Yield per head (g)	Yield per plant (g)	Yield (g m ⁻²)
A-DS	79 ^{c2}	0.030 ^{b2}	2.3 ^{c2}	0.1 ^{d2}
A-DSM	250 ^a	0.052 ^a	11.7 ^b	12.3 ^c
A-TP	237 ^b	0.051 ^a	12.0 ^b	23.7 ^b
A-TPM	213 ^b	0.052 ^a	10.6 ^b	20.3 ^b
S-TP	265 ^a	0.049 ^a	12.5 ^a	27.8 ^a

¹ Refer to abbreviations in Table 2.

² Means within a column followed by the same letter are not significantly different at $P=0.01$.

were carried out at twelve planting spots per plot excluding those in the peripheral area. Each treatment was replicated three times.

For the mulching treatments, black polyethylene film (0.02 mm thick, light transmission <1%) was placed on the plots at the time of direct sowing/transplanting and removed on 26-28 March 1994.

The number of surviving plants was examined for each plot on 3 December 1993 and 16 May 1994. The size of individual plants (average of diagonal diameters) was measured on 3 December 1993 and 3 June 1994 for each plot. The time of flowering for each treatment was determined when half of the surviving plants had opened 3 or more florets on at least one head.

The flower heads were harvested by hand on 1 August and dried. Then the seeds were hand cleaned using sieves and a seed blower. The number of flower heads (flower heads showing white corollas through to those which were dark brown) at the time of harvest, and seed weight were recorded for each plot.

RESULTS

Weather data during the experiment are shown in Table 1. The temperature soon after the spring transplanting (from April to early May) was higher than average by about 2°C. In July, rainfall was far less than normal, because this month was drier, warmer and sunnier than an average year.

For the two direct sowing treatments, the time of emergence was earlier in A-DSM than in A-DS by about three days. Seedlings reached the 1-3 leaf stage for A-DS and the 3-4 leaf stage for A-DSM one month after sowing. No stolons appeared before winter in either treatment.

For the autumn transplanting treatment, seedlings were transplanted at the 5-6 leaf stage and stolons began to appear in the middle of October. All plants had produced stolons before winter, but in mulched plots, some stolons grew in the space between the ground and mulching film and were inhibited in their growth by shading. The size of plants on 3 December was larger in A-TP than A-TPM (Table 3).

Cold injury occurred from the end of November when the minimum temperature constantly went below 0°C. Survival of plants in early winter (3 December) was 100% for A-TP and A-TPM and approximately 90% for A-DSM (Table 4). About half of the plants were killed for A-DS before winter. The survival rate in spring (16 May) decreased to about 90% for A-TP and A-TPM, 50% for A-DSM and 6% for A-DS.

The size of individual plants on 3 June was largest in A-TP where seedlings were the largest in autumn (Table 3). Some plants in this treatment had grown into their neighbouring plants by 3 June. The size of A-TPM was the second highest. In A-DS and A-DSM, growth in spring was low because of the damage caused by the chilling during winter.

For S-TP, seedlings were at the 12-15 leaf stage and some stolons were beginning to grow when transplanted. The weather after transplanting was mild and the seedlings grew well. Plant size was larger than that of autumn direct sowing treatments (A-DS and A-DSM) and smaller than that of autumn transplanting treatments (A-TP and A-TPM). All plots of the three transplanting treatments had reached 100% ground coverage before harvest.

Flower head emergence and flowering were earliest in the two autumn transplanting treatments, where individual plants were the largest (Table 3). For A-DSM and S-TP, flower head emergence and flowering occurred a little later. However, the number of flower heads per plant at the time of harvest was higher in A-DSM and S-TP than in A-TP and A-TPM (Table 5).

The seed yields per flower head were almost the same for all treatments except for A-DS, where the value was about 60% of the other treatments (Table 5). The seed yield per plant was highest in S-TP but the difference between S-TP and the other three treatments was small. No plants were killed until harvest, and the highest seed yield of 27.8g m⁻² was obtained in S-TP.

DISCUSSION

Sowing at an optimum time is important for obtaining good stand establishment and high seed yield. Sugita *et al* (1984) pointed out that a delay in sowing of white clover leads to insufficient growth of seedlings and therefore low seed yield, especially for low density sowings.

At Nagano Station, sowing of white clover for seed production is apt to be delayed until the middle or the end of September because of a dry August and the monsoon in September. However, seed for transplanting can be sown independent of weather, so that larger seedlings are therefore available at the beginning of winter.

Survival in spring was higher when seedlings were larger at the beginning of winter. This suggests that larger seedlings have higher resistance to cold. In the Saku area, mean temperature goes below 5°C toward the end of November and the growth of white clover stops. The growth period

before winter for direct sowing treatments was therefore about two months and this period might not be sufficient for white clover seedlings to become large enough to gain sufficient cold tolerance.

Growth in spring and seed yield per plant were also better in plots where seedlings were larger in autumn. Based on these results, it can be concluded that sowing at an optimum time and then transplanting can assure sufficient growth before winter, improve overwintering and growth in spring, and thus lead to higher seed yield.

The growth of seedlings was also accelerated by mulching. In the direct sown plots, plant growth before winter was improved by mulching and this led to a higher survival rate in spring and also better growth in spring. Seed yield was much higher in A-DSM than in A-DS. In transplanted plots, however, stolons were inhibited in their growth by shading and this led to decreased seed yield.

The highest yield of 27.8 g m⁻² was obtained in S-TP. In this treatment, the growth of seedlings was good as they had been raised in a glasshouse. The number of flower heads per plant was larger while plant size was smaller in S-TP than in A-TP and A-ATM. This suggests that overmatured flower heads in autumn transplanting treatments might have been lost by collapsing before harvest.

It should be noted that temperature after transplanting happened to be higher by about 2°C than usual, and this condition was favourable to the growth of S-TP plants. To clarify whether autumn transplanting or spring transplanting would be most effective to improve seed production in white clover, further research might be necessary.

The results in this study strongly suggest the advantage of the transplanting method in Japanese white clover basic seed production, and the possibility of obtaining seed yields of several kilograms per 100 m².

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