Effect of the Plant Growth Regulator Parlay on the Seed Production of the White Clover cvs. Menna and Olwen
I. Stolon Growth and Development

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ABSTRACT

Two white clover cultivars, Olwen (large-leaved) and Menna (medium-leaved) were sown in field plots at a seed rate of 3 kg ha^{-1}. At two dates in spring, corresponding to distinct phases of reproductive growth, plots were mechanically defoliated or left uncut, followed by a subsequent application of the plant growth regulator Parlay (Paclobutrazol) at a rate of 1.0 kg a.i. ha^{-1}. Controls received no growth regulator. Measurements of primary stolon growth, stolon growth per unit area and petiole and peduncle height were made to determine the effect of Parlay on the stolon growth and development of white clover under different managements.

Parlay significantly reduced primary stolon length and the number of axillary buds per stolon in both years but in 1987 only, significantly reduced the number of nodes per stolon and increased the number of reproductive nodes. Parlay also increased the overall number of axillary buds, growing points, total nodes m^{-2} and percentage of nodes forming axillary buds in both years but had no effect on stolon density. Petiole height was reduced but there was little effect on peduncle height. The effectiveness of Parlay was influenced by application date which differed between years. Defoliation had little effect on these growth parameters apart from reducing petiole height. Cultivar differences resulted primarily from the fact that cv. Menna had a significantly higher stolon density, more nodes m^{-2} and longer primary stolons than cv. Olwen in both years. Cv. Menna also had more nodes but fewer reproductive nodes than cv. Olwen in 1988. Generally cultivars responded similarly to Parlay and there was little difference between Parlay application to cut and uncut plots. The implications of these differences in stolon growth and development and the relative heights of the petioles and peduncles are discussed in relation to their likely effect on seed production.

Additional index words: **white clover, *Trifolium repens*, seed production, plant growth regulator, Parlay, stolon growth.**

INTRODUCTION

White clover seed production in the United Kingdom is limited because of the difficulty in obtaining consistent economic seed yields (Hides, Lewis and Marshall, 1984). This inconsistency results primarily from variable weather conditions during critical periods of crop growth and lack of suitable seed crop management practices for optimising seed yield. Low seed yields can also be attributed to the characteristic growth pattern of white clover during seed production. The intermittent inflorescence production along the stolon (Thomas, 1981) and indeterminate flowering pattern dictate that inflorescences, the main component of seed yield (Evans, Williams and Davies, 1986; Hollington, Marshall and Hides, 1989) appear over an extended period of time. The resulting range of inflorescence ripeness within the crop makes it difficult to optimise harvest date, early harvesting resulting in a high proportion of immature inflorescences whilst late harvesting may miss the larger earlier formed inflorescences (Hollington *et al.*, 1989).

Techniques which can reduce the spread of flowering, increase inflorescence production, or improve harvestability and increase seed yield potential are therefore essential. One approach has been to examine whether plant growth regulators could be used to manipulate plant growth and increase potential seed yields. A preliminary field experiment (Marshall and Hides, 1986) identified the plant growth regulator Parlay (PP333, Paclobutrazol) as a potentially useful chemical. Subsequent glasshouse studies (Marshall and Hides, 1987) have shown that Parlay applied at a rate of 1.0 kg a.i. ha^{-1} at certain stages of inflorescence development can successfully modify stolon growth and development in ways that may increase potential seed yield. The effects of these application dates and rates have however not yet been tested in the field.

Techniques developed to improve seed yield potential should be applicable to as wide a range of white clover cultivars as possible. It is essential therefore that they be evaluated on cultivars which may differ in their pattern of inflorescence production (Evans *et al.*, 1986) and in stolon morphology (Marshall and James, 1988).

The effect of Parlay on the seed yield potential of white clover may also be linked with other seed crop managements and since Parlay is soil acting (Froggatt, Thomas and Batch, 1982), it is essential that when applied,

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it reaches the soil surface. As seed crops are also normally defoliated in spring (Clifford, 1979; Marshall, Hollington and Hides, 1989) this may be ensured by applying Parlay directly after cutting.

This paper reports the results of a field experiment carried out over two harvest years, designed to assess the effect of Parlay applied at two dates to uncut and cut plots, on stolon growth and development of the medium leaved cv. Menna and the large leaved cv. Olwen. The effect of these treatments on seed yield components will be reported in a later paper.

MATERIALS AND METHODS

Plots (1.5 m x 3 m) of cv. Menna and cv. Olwen were sown at a seed rate of 3 kg ha^{-1} and 15 cm drill width into soil of the Clwyd series at the Welsh Plant Breeding Station, Plas Gogerddan, Aberystwyth on 28 August 1986 and 1 July 1987. Details of experimental management are given in Table 1.

Table 1. Experimental details.

	1986	1987
Plot size	1.5 x 3 m	1.5 x 3 m
Sowing date	28 August	1 July
Sowing rate	3 kg ha^{-1}	3 kg ha^{-1}
Row spacing	15 cm	15 cm
Herbicide	Legumex Extra 4 litre ha^{-1}, 21 Aug. Carbetamix 3 kg ha^{-1}, 15 Oct. Carbetamix 4 kg ha^{-1}, 13 Feb. '87	Carbetamix 4 kg ha^{-1}, 2 March '88
Fertiliser	40 kg ha^{-1} 15:15:21[1], 26 Sept. 158 kg ha^{-1} 0:40:0, 0:0:60, 16 Feb. '87	40 kg ha^{-1} 9:24:24, 5 Oct. 60 kg ha^{-1} 9:24:24, 24 Feb. '88

[1] $N:P_2O_5:K_2O$

Development of the plants was monitored by collecting 10 stolon growing points of each cultivar at random from within the experiment at approximately weekly intervals in the spring of 1987 and 1988. At two stages of development of the growing point of each cultivar (Table 2), plots were cut to a height of approximately 3 cm or left uncut. On each cutting date, the growth regulator Parlay was applied by knapsack sprayer to half of the cut and uncut plots at a rate of 1.0 kg ai. ha^{-1} in 400 litres water, the remaining half (controls) receiving no growth regulator.

The experiment was a split, split, split plot design with cultivars as main plots, cuts as sub plots, Parlay as subsub plots and application dates as subsubsub plots.

Table 2. Stages of inflorescence development of the first flowering bud of white clover cvs. Menna and Olwen at the two application dates.

Stage of development of the first flowering bud	Date of defoliation and application of Parlay	
	1987	1988
Appearance of flower bud on stolon	30 April	21 April
Peduncle height 3 cm	26 May	24 May

Biological Measurements

Stolon growth and development

A stolon core (17 cm diameter) was removed at random from each plot at harvest (on 13 July 1987 and 8 August 1988). This corresponded to the date of maximum number of ripe inflorescences. The following measurements were made: total length of stolon, number of nodes, axillary buds and growing points.

Five intact stolons were collected on the same date

from each plot, care being taken to ensure that each stolon came from a separate plant. On each stolon the nodes immediately distal the shortest internodes were assumed to be the first to have emerged from the apical bud in the spring. The following measurements were made on each stolon: length (cm) (measured from the first elongating internode), total number of nodes, nodes producing axillary buds and reproductive nodes. Petiole and peduncle height at peak flowering (27 June 1987 and 30 June 1988) were measured on five fully emerged petioles and peduncles collected at random from each plot.

Weather

Weather conditions at Gogerddan between March and August 1987 and 1988 are shown in Table 3. Early spring temperatures in March and in May and June were higher in 1988 than in 1987, while temperatures in April, July and August were higher in 1987. Total rainfall in 1987 was less than in 1988 due primarily to lower rainfall in July with a considerably drier August. In 1987 April and June were wetter but May was drier. Sunshine hours in 1987 were higher in March, July and August but 1988 had more hours of sunshine in April and June.

Table 3. Weather data (monthly means) during the course of the experiment in 1987 and 1988.

		March	April	May	June	July	August	Total
Min. temperature	1987	2.3	5.9	5.9	9.2	11.6	11.9	-
(°C)	1988	4.5	4.4	6.9	10.2	12.1	11.3	-
Max. temperature	1987	7.8	14.4	13.7	15.3	18.5	18.2	-
(°C)	1988	9.3	13.0	16.0	18.7	16.5	17.8	-
Rainfall	1987	129.4	61.1	41.3	95.4	93.1	68.4	458.7
(mm per month)	1988	146.7	39.2	73.9	27.2	146.1	134.0	567.1
Sunshine	1987	98.9	155.0	227.6	138.6	181.1	167.2	-
(hours/month)	1988	75.9	184.8	224.6	210.9	114.9	137.7	-

RESULTS

Stolon growth and development per unit area

In both harvest years Parlay significantly increased the numbers of axillary buds, growing points, total nodes m^{-2} and the percentage of nodes forming axillary buds but had no effect on stolon density (Table 4). Cultivars also differed significantly in their stolon growth and development. Cv. Menna had a higher ($P < 0.05$) stolon density and more nodes m^{-2} than cv. Olwen in both years but more axillary buds and growing points m^{-2} in 1987 only (Table 4).

Defoliation had little effect on stolon growth but, in 1987 only, significantly increased the number and percentage of nodes forming axillary buds (Table 4).

There were no significant Parley x cultivar interactions in any stolon growth parameters and few significant Parlay x defoliation interactions. In 1988, however, the number of nodes forming axillary buds was increased ($P < 0.05$) when Parlay was applied to uncut plots (2400) compared to the cut plots (1450) but there was no difference between cutting treatments in the absence of Parlay. The number of growing points m^{-2} was also increased ($P < 0.05$) when Parlay was applied to cut plots (4300) compared to uncut plots (3250) in 1987 but greater when applied to uncut plots (5900) than to cut plots (4400) in 1988. There was no difference in the absence of Parlay.

There were also significant Parlay x application date interactions. Stolon density, in 1988 only, was reduced ($P < 0.05$) when Parlay was applied in April but increased when applied in May (Fig. 1). The number of axillary buds and growing points m^{-2} in 1987 and the number and percentage of nodes forming axillary buds in 1988 were however higher ($P < 0.05$) when Parlay was applied in May than when applied in April.

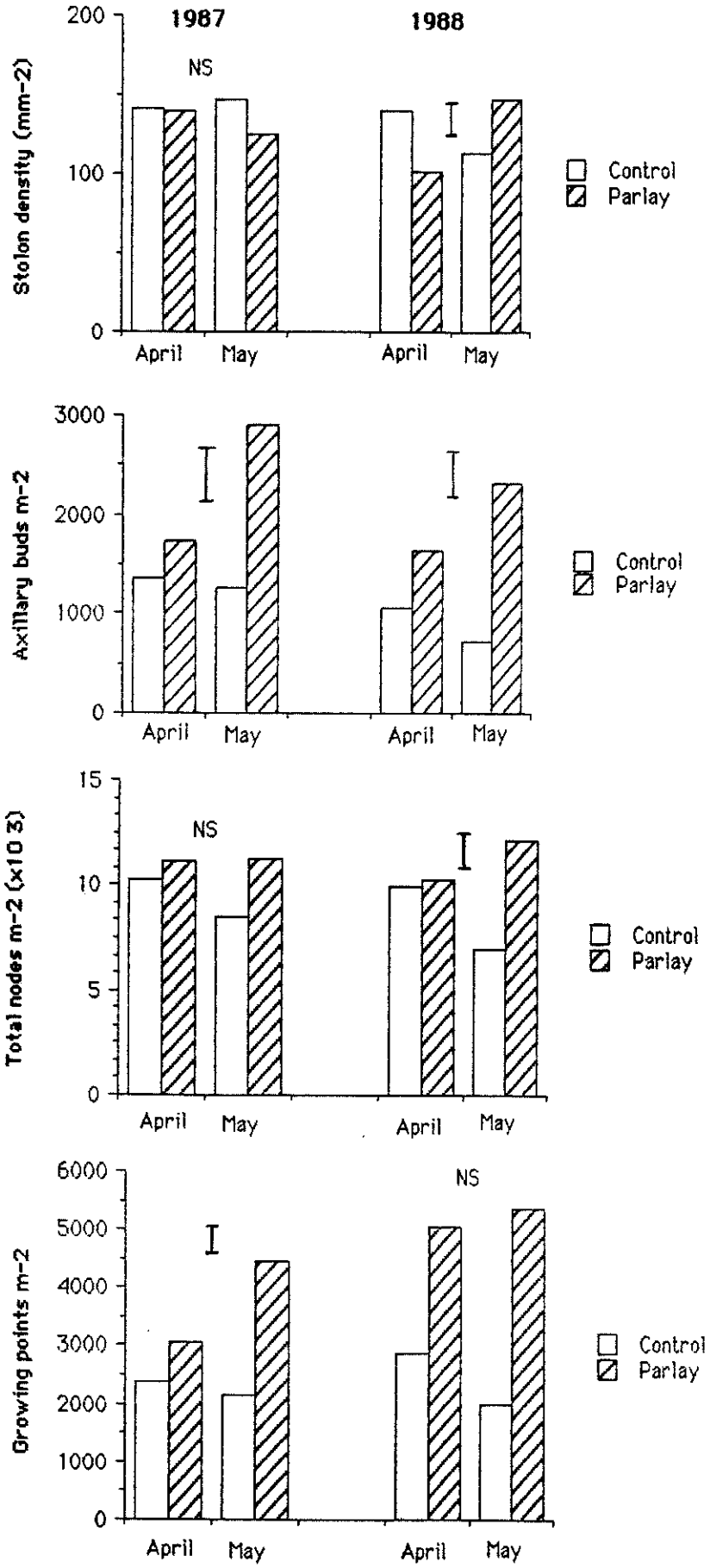


Figure 1. Effect of Parlay (shaded column) applied at 1.0 kg a.i. ha^{-1} in April and May in 1987 and 1988 on stolon growth. Controls received no growth regulator. Data are a mean of two cultivars and two cutting treatments. Vertical bars represent LSD at P = 0.05.

Table 4. Effect of Parlay (chemical), cultivar and defoliation on stolon growth of white clover.

	Stolon density (metres m^{-2})		Total nodes m^{-2}		Axillary buds m^{-2}		% nodes forming axillary buds		Growing points m^{-2}	
	1987	1988	1987	1988	1987	1988	1987	1988	1987	1988
Chemical										
No chemical	114.1	121.2	9429	8341	1328	878	14.0	10.7	2294	2431
Chemical	132.6	130.5	11217	11124	2328	1971	20.8	17.5	2756	5217
LSD (P = 0.05)	NS	NS	1358	1307	373	362	3.6	3.1	288	713
Cultivar										
Menna	162.8	152.3	12298	11200	2293	1571	18.6	13.3	3753	4207
Olwen	109.5	99.4	8350	8265	1363	1277	16.2	14.9	2297	3441
LSD (P = 0.05)	21.9	14.4	1646	1310	363	NS	NS	NS	492	NS
Defoliation										
No cut	138.1	133.2	10045	10142	1577	1569	15.8	14.7	2786	4044
Cut	134.3	118.5	10592	9323	2099	1281	19.1	13.4	3264	3605
LSD (P = 0.05)	NS	NS	NS	NS	318	NS	1.8	NS	NS	NS

Primary stolon growth and development

Parlay reduced ($P < 0.05$) primary stolon length and increased the number of axillary buds per stolon in both harvest years. In 1987 Parlay also reduced ($P < 0.05$) the number of nodes per stolon and increased the number of reproductive nodes per stolon (Table 5). Defoliation had no significant effect on any of these stolon growth parameters.

There were significant cultivar differences, cv. Menna having longer primary stolons in both harvest years and more nodes, but fewer reproductive nodes than cv. Olwen in 1988 (Table 5).

There were no significant Parlay x defoliation interactions and no significant Parlay x cultivar interactions in 1987. However, in 1988 Parlay reduced ($P < 0.05$) stolon length (23.5 cm) compared to the control (31.0 cm) and increased the number of axillary buds of cv. Menna from 0.8 to 1.0 per stolon but had no significant effect on cv. Olwen. There were also significant Parley x date interactions, Parlay increasing ($P < 0.05$) the number of axillary buds (0.45) compared to the control (0.17) when applied in April but not in May in 1987 and of reproductive buds (2.8) when compared to the control (2.0) in April but not in May in 1988.

Petiole and peduncle height

Parlay reduced petiole height more than peduncle height and therefore increased the relative height difference between the two. This led to a significant elevation of inflorescences above petiole height, particularly in 1988 (Table 6). There were also cultivar differences, cv. Olwen having longer ($P < 0.05$) petioles and peduncles than cv. Menna in 1987 but cv. Menna longer ($P < 0.05$) peduncles in 1988. There was no significant difference in 1987 but in 1988, cv. Menna had a significantly larger height difference between petioles and peduncles than cv. Olwen.

Defoliation reduced ($P < 0.05$) petiole height in both years and peduncle height in 1987 only but did lead to a significant elevation of inflorescences above petioles in both years (Table 6).

There was no significant Parlay x cultivar interaction. There was, however, a significant Parlay x defoliation interaction in 1987 only as Parlay reduced ($P < 0.05$) both petiole and peduncle height and reduced the height difference when applied to the cut plots but not to the uncut plots (Table 7). Petiole and peduncle height but not height difference were also influenced by a significant Parlay x date interaction. In 1988 petiole height was reduced ($P < 0.05$) more by Parlay compared to the controls when applied in May (Control, 23.7 cm; Parlay, 13.1 cm), than in April (Control, 28.1 cm; Parlay, 23.7 cm) when Parlay had less effect compared to the controls. In 1987 and 1988, peduncle height following Parlay application (28.6 cm in 1987 and 21.5 cm in 1988) was lower ($P < 0.05$) than the control (31.3 cm in 1987 and 31.8 cm in 1988). There were no other significant interactions.

Table 5. Effect of Parlay (chemical) and cultivar on stolon growth.

	Primary stolon length (cm)		Nodes per stolon		Axillary buds per stolon		Reproductive buds per stolon	
	1987	1988	1987	1988	1987	1988	1987	1988
Chemical								
No chemical	21.8	27.5	15.4	13.8	0.3	0.9	1.2	2.2
Chemical	17.9	22.3	14.6	13.6	0.4	1.9	1.6	2.6
LSD (P = 0.05)	1.3	2.7	0.4	NS	0.1	0.6	0.2	NS
Cultivar								
Menna	20.7	27.0	15.1	14.0	0.3	1.6	1.3	1.8
Olwen	19.1	23.1	14.9	13.5	0.4	1.3	1.4	2.9
LSD (P = 0.05)	1.3	2.0	NS	0.5	NS	NS	NS	0.3

Table 6. Effect of Parlay (chemical), cultivar and defoliation on petiole and peduncle height.

	Petiole height (cm)		Peduncle height (cm)		Height difference	
	1987	1988	1987	1988	1987	1988
Chemical						
No chemical	26.6	28.5	30.9	31.2	4.3	2.8
Chemical	23.7	18.6	29.0	27.4	5.3	8.8
LSD (P = 0.05)	0.7	1.7	0.7	1.4	0.5	1.2
Cultivar						
Menna	24.1	23.6	28.9	30.4	4.8	6.8
Olwen	26.2	23.5	31.0	28.3	4.7	4.8
LSD (P = 0.05)	0.7	NS	0.7	1.3	NS	1.1
Defoliation						
No cut	26.5	25.3	30.9	29.7	4.4	4.4
Cut	23.8	21.8	29.0	29.0	5.2	7.2
LSD (P = 0.05)	0.7	1.7	1.0	NS	0.5	1.2

DISCUSSION

This experiment has shown how the plant growth regulator Parlay can modify the stolon growth and development of white clover in ways that could increase white clover seed yields. This confirms evidence from a previous glasshouse experiment (Marshall and Hides, 1987). Generally, Parlay increased axillary bud development and the number of nodes per primary stolon forming axillary buds. Individual stolon length was also significantly reduced, as observed in previous glasshouse studies (Marshall and Hides, 1987). At harvest, stolon density was not significantly influenced by Parlay as reduced stolon length was compensated for by more branching. Parlay therefore significantly increased the number of nodes per unit area without a corresponding effect on stolon density. Since inflorescences are produced at nodes on the stolon, any increase in the number of nodes per unit area should increase inflorescence production.

Table 7. Effect of Parlay applied at 1.0 kg a.i. ha^{-1} to cut and uncut plots on petiole and peduncle height and height difference (1987). Controls received no growth regulator.

	Height (cm)								
	Petiole			Peduncle			Difference		
	Cut	Uncut	mean	Cut	Uncut	mean	Cut	Uncut	mean
No chemical	26.1	27.2	26.7	30.5	31.4	31.0	4.4	4.2	4.3
Chemical	21.4	25.9	23.7	27.6	30.5	29.1	6.2	4.6	5.4
Mean	23.8	26.9		29.1	31.0		5.3	4.4	
LSD (P = 0.05)									
Cut		0.7			0.7			0.5	
Chemical		0.7			0.7			0.5	
Cut x chemical		1.1			1.0			0.7	

Although the cultivars used in this study differed in length of primary stolons and growth per unit area, in general they responded similarly to Parlay. These differences between cultivars in stolon morphology have been shown in previous field experiments (Evans *et al.*, 1986; Marshall and James, 1988) and reflect the different agricultural systems for which they were bred. Cv. Olwen, the large leaved cultivar, was bred for conservation and rotational grazing systems where a high stolon density is unnecessary, whilst cv. Menna was bred for a wider range of management systems, including more severe grazing systems where the prevention of stolon removal is important. In these systems, a dense mat of branched stolons and better anchorage of stolons is essential.

Cultivar differences were very small, there being only one significant effect in response to Parlay. In 1988 Parlay reduced primary stolon length and increased the number of axillary buds of cv. Menna but not of cv. Olwen. The reason for these differences between cultivars is not clear, and they were not reflected in differences in inflorescence production (Marshall and Hides, 1991). Whilst this experiment has evaluated a medium and a large leaved cultivar, other field experiments have shown that Parlay applied to small leaved cultivars such as cv. Gwenda can similarly reduce primary stolon length and increase branching (Hollington, pers. comm.). It would appear therefore that in general, recommendations concerning the use of Parlay apply to cultivars in all leaf size categories.

Defoliation had little effect on stolon growth per unit area or on individual stolon growth, although in 1987 it significantly increased the number and percentage of nodes forming axillary buds. It was also clear that Parlay in general had similar effects if applied to uncut or cut plots irrespective of application date. Given that foliar uptake is regarded as being of little importance (Shearing and Batch, 1982) these results suggest that irrespective of the height and density of the leaf canopy, Parlay was able to reach the soil surface.

Although Parlay significantly reduced both petiole and peduncle height, it reduced petiole height significantly more so, which increased the elevation of the inflorescences above canopy height. Both cultivars responded similarly, confirming evidence from previous field (Marshall and Hides, 1986) and glasshouse (Marshall and Hides, 1987) experiments. The implications of these results in relation to pollination and harvesting are discussed further (Marshall and Hides, 1991).

There were significant differences in the effect of Parlay between years. This variability in effectiveness has also been demonstrated in experiments where Parlay was applied to perennial ryegrass seed crops (Hampton and Hebblethwaite, 1985) and may be attributed to the fact that Parlay is soil acting (Froggatt *et al.*, 1982) and as such requires adequate soil moisture to be taken up by the roots. In 1987, rainfall was lower in May than in April or June whilst in 1988 it was higher in May than in either April or June. Such climatic differences are likely to have an effect on soil moisture levels and consequently uptake of Parlay from the soil. Although no clear cut effect of a delay in Parlay uptake can be identified, precise application dates may be inappropriate if rainfall and soil moisture is such an important factor in the effectiveness of this particular plant growth regulator.

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