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Preliminary Observations on the Induction of Precocious Flowering in Hevea Brasiliensis *by PP333*

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The growth retardant PP333, applied as a soil drench, induced precocious flowering in young ungirdled Hevea *planted in polybags or in the field. Viable pollen and seeds were obtained from the treated plants. Some effects of PP333 on floral and vegetative morphology are described.*

Hevea is a perennial crop that has a juvenile phase of four to five years during which the trees do not flower. This vegetative phase imposes a limitation to the extent by which the breeding cycle can be shortened. Precocious flowering in *Hevea* would enable a reduction in the duration between generations which would in turn enable an acceleration of the advancement in breeding and selection of the crop. Various benefits relating to hand-pollination practice may also be derived from early flowering in young rubber plants. Among these are the convenience of hand-pollination from the ground, more effective pathogen control leading to better fruit-set[1-3] and an extended duration of flowering which may help to overcome non-synchrony in flowering between clones[1,2].

Girdling of the trunk or branches (ring barking) has generally been found to be the most effective means of inducing early flowering in *Hevea*[1,2,4-7]. Chemical treatments have occasionally been reported to be effective, but often, their effectiveness have been evident only when complemented by girdling[1,2,6,7].

Flowering has been reported to be enhanced in the apple tree after treatment with the growth retardant PP333 (paclobutrazol)[8]. This paper describes preliminary findings on the effect of PP333 on the induction of precocious flowering in young ungirdled *Hevea* plants.

MATERIALS AND METHODS

Experiment 1a. Eighteen-month-old RRIM 600 plants grown in 36 × 62 cm polybags were untreated (control), repeatedly girdled or received a single dose in varying strengths of (2RS 3RS)-1-(-4-chlorophenyl)-4,4-dimethyl-2-(1H-1,2,4-triazol-1-yl) penton-3-ol, also known as paclobutrazol or PP333 (Imperial Chemical Industries, Plant Protection Division, UK). Ten plants were allotted to each treatment.

Experiment 1b. Eighteen-month-old PB 5/51 plants in 36 × 62 cm polybags were treated as in *Experiment 1a,* but the PP333-treated plants received a second application six weeks after the first dose. Ten plants were allotted to each treatment.

Experiment 2a. In the field, treatments were assigned to fourteen-month-old RRIM 600 plants in three randomised blocks of sixteen trees per treatment. The plants were untreated (control) or given a single dose of PP333 of varying strengths.

Experiment 2b. Fourteen-month-old GT 1 plants were laid out in two blocks of twelve plants per treatment in the field. Treatments similar to those in *Experiment 2a* were applied but the PP333-treated plants received an additional second dose three months after the initial application. A repeated girdling treatment was also included.

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In the above experiments, repeated girdling was carried out by removing a 2 mm ring of bark from the stem at approximately monthly intervals. Application of PP333 was by soil drench and quantities of PP333 cited in the text refer to the active ingredient (a.i.). The compound was made up to volume with water and 100 ml were applied per polybag plant and 1 litre per field plant.

Recordings on flowering were made once a month and scores of flowering plants and flowering intensity (number of flower panicles per flowering tree) pertained to the day of observation. As such, they do not denote the total number of flowering trees or flower panicles present during the entire month but reflect the relative intensities of flowering between the different treatments on the day of observation.

To germinate pollen, staminal columns were collected in a glass vial and allowed to dehisce in the laboratory. The pollen grains were suspended in a hanging drop of 0.01% boric acid containing 15% sucrose. Pollen germination was scored the following morning under a microscope.

RESULTS

Flowering in Polybag Plants

Experiment 1a. Flowering was first observed in RRIM 600 polybag plants five months after they were treated with PP333 at a rate of 0.75 g per plant. By the eleventh month after treatment, five out of ten treated plants had flowered *(Table 1)*. A lower rate of application of 0.5 g was also effective but the effectiveness diminished when only 0.25 g was applied. Repeated girdling induced flowering in only one out of ten plants and a high incidence of dieback was observed. Flowering was not observed in any of the untreated control plants.

Experiment 1b. With PB 5/51 polybag plants, flowering was first observed four months after their first treatment. No difference was observed between the three rates of application used (2×0.75 g, 2×0.5 g, 2×0.25 g per plant). In each instance, three plants out of ten were induced to flower. As with RRIM 600, plants subjected to repeated girdling were prone to dieback. Flowering was observed in two plants while none of the untreated control plants flowered.

TABLE 1. CUMULATIVE INCIDENCE[a] OF FLOWERING IN POLYBAG PLANTS THAT WERE REPEATEDLY GIRDLED OR TREATED WITH PP333

Treatment	Incidence of flowering	
	RRIM 600 (Experiment 1a)	PB 5/51 (Experiment 1b)
Control	0	0
Repeated girdling	1	2
PP333[b]		
0.25 g	2	
0.50 g	4	
0.75 g	5	
2×0.25 g		3
2×0.50 g		3
2×0.75 g		3

[a]Number of flowering plants out of ten plants per clone
Observations were made 11 months after commencement of the treatments.

[b]RRIM 600 plants received a single application of PP333 while PB 5/51 plants received a second application after six weeks.

In both *Experiments 1a* and *1b,* plants that were treated with PP333 appeared to be in a state of stress. Leaf abscission was observed a few weeks after treatment while retarded growth was sustained over a year of observation. The new growth occurring after treatment in some of the treated plants had short internodes while leaf petioles were similarly short and closely inserted within the flush. There was however no clear relationship between this growth habit and the incidence of flowering. Flowering was also induced in plants not markedly exhibiting the trait. As in the case of the foliage, the PP333-induced inflorescences appeared similarly clumped and compact with the flowers packed very close together *(Figure 1).* Premature flower drop was frequently observed.

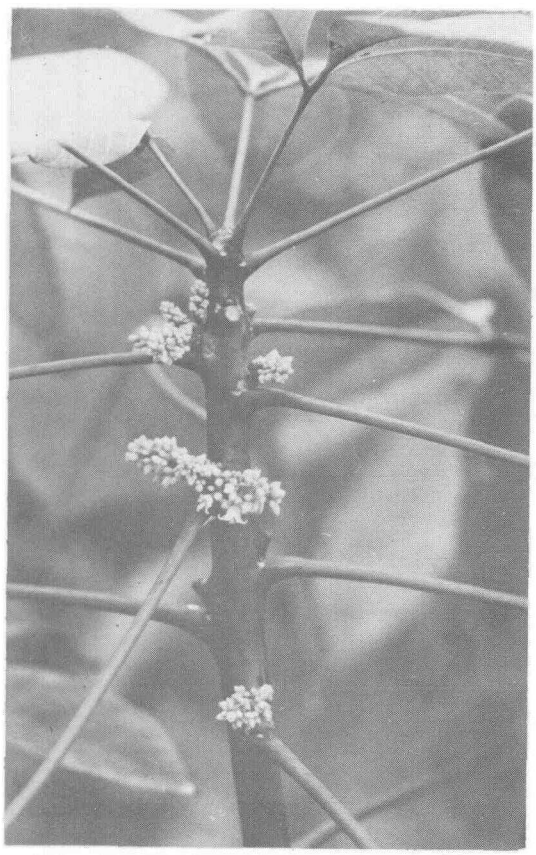

Figure 1. Compact panicles on a two-year-old PB 5/51 polybag plant treated with PP333.

Flowering in Field Planting

Experiment 2a. Following PP333 treatment of the plants in the field in July, flowering in RRIM 600 was first observed six months later, coinciding with the main flowering season that followed wintering. The chemical enhanced wintering (which is normally mild or absent in plants of this age). The most effective among the dosages tested was 10 g and this induced flowering in twenty-six out of forty-eight plants (54%) in the duration of a year from the time of first flowering *(Figure 2).* Lower rates of 0.5 g to 5 g were relatively less effective. A small number of untreated trees flowered towards the later part of the study *(Figure 2).* In terms of the intensity of flowering (number of flower panicles per flowering tree), an optimum dosage was not readily discernible from the limited observations *(Table 2)* but flowering plants treated with 2 g and 10 g PP333 bore the largest number of inflorescences.

Inflorescences of PP333-treated plants appeared normal in their development with no obvious signs of growth inhibition *(Figure 3).*

Experiment 2b. Compared with RRIM 600, GT 1 plants treated with PP333 in the field showed markedly more severe wintering with branches completely bare in many instances. Inflorescences subsequently developed on the almost bare branches of this clone. They consisted of compact panicles with closely packed flowers borne on very short pedicels *(Figure 4).* Flowering tended to be relatively sparse in PP333-treated plants that had copious refoliation.

Flower-induction in GT 1 was most effective with 2 × 5 g PP333, flowering being first observed six months from the initial treatment. Within a year from the first observation of flowering, seventeen out of twenty-four plants (71%) had flowered *(Figure 5).* Repeated trunk girdling was almost as effective, while rates higher or lower than 2 × 5 g were less effective. Only one untreated tree flowered during the period of observation *(Figure 5).* With GT 1, plants treated with 2 × 2 g and 2 × 5 g PP333 had the largest number of panicles per flowering plant *(Table 2).* On refoliation,

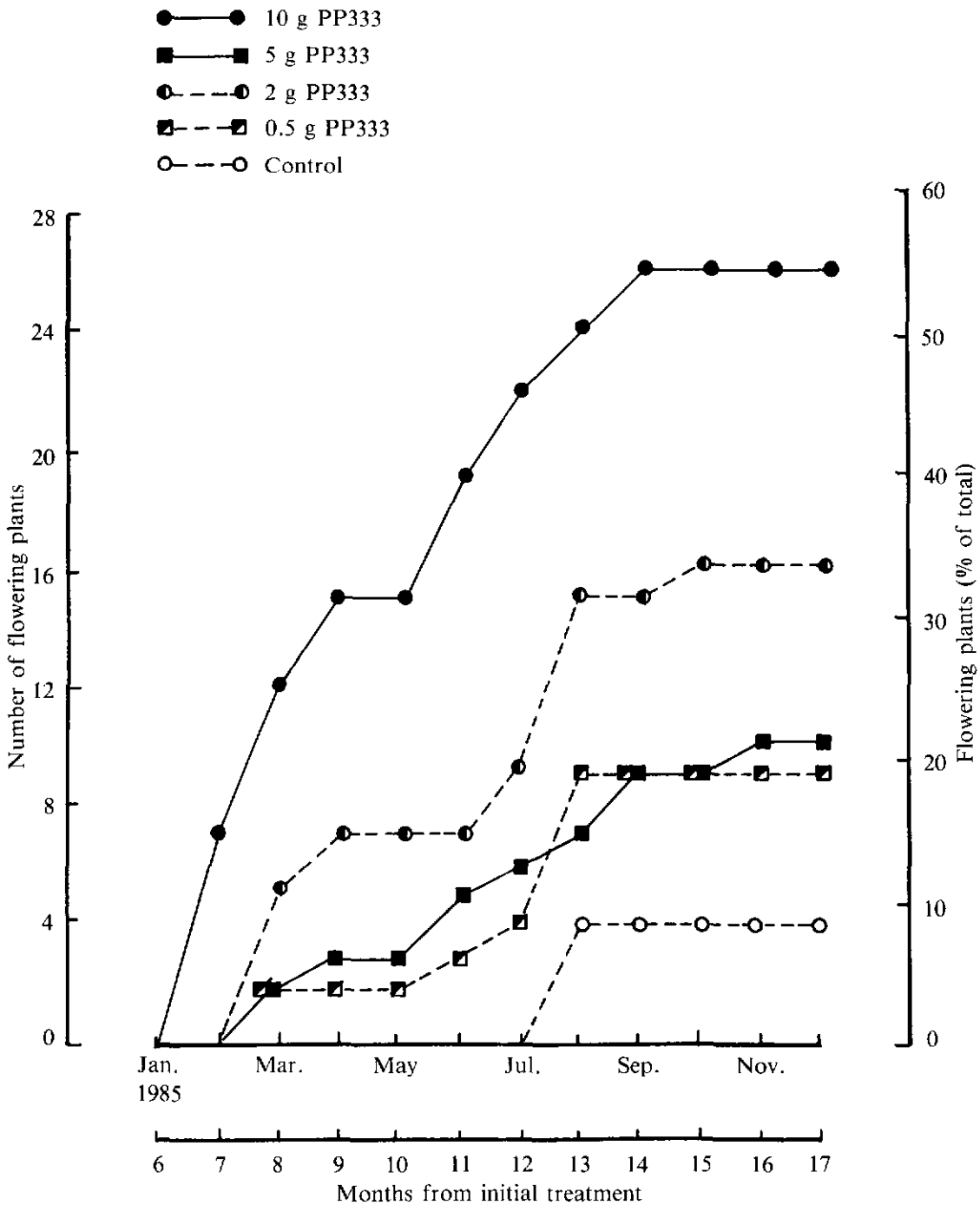

Figure 2. Cumulative incidence of flowering in RRIM 600 plants in the field. Fourteen-month-old plants (forty-eight per treatment) were given a single application of PP333 in July 1984.

TABLE 2. INTENSITY OF FLOWERING[a] IN THREE-YEAR-OLD PLANTS THAT WERE REPEATEDLY GIRDLED[b] OR TREATED WITH PP333

Treatment	Intensity of flowering	
	RRIM 600 (Experiment 2a)	GT 1 (Experiment 2b)
Control	4.3	1
Repeated girdling	—	7.8
PP333[c]		
0.5 g	6.4	
2 g	15.1	
5 g	5.1	
10 g	9.2	
2 × 0.5 g		2.7
2 × 2 g		21.0
2 × 5 g		19.2
2 × 10 g		8.2

[a]Number of flowering panicles per flowering plant; mean of one observation each in July, August and September 1985

[b]GT 1 only

[c]RRIM 600 plants received a single application of PP333 while GT 1 plants received a second application after three months.

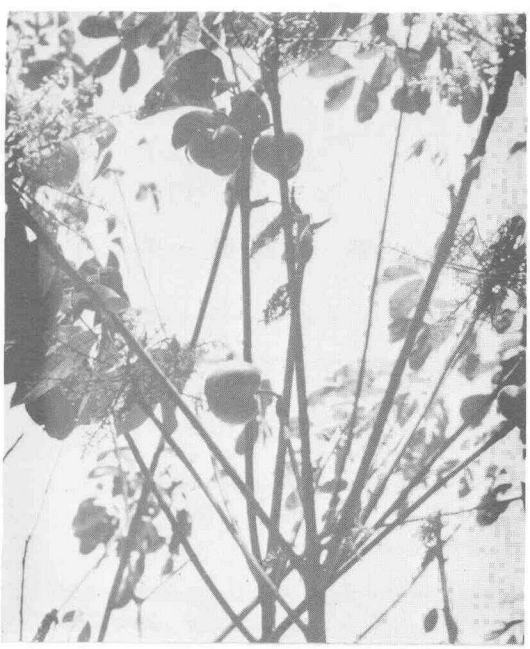

Figure 3. Flowers and fruits on a two-year-old RRIM 600 plant in the field treated with PP333.

Figure 4. Compact panicles on a two-year-old GT 1 plant in the field treated with PP333.

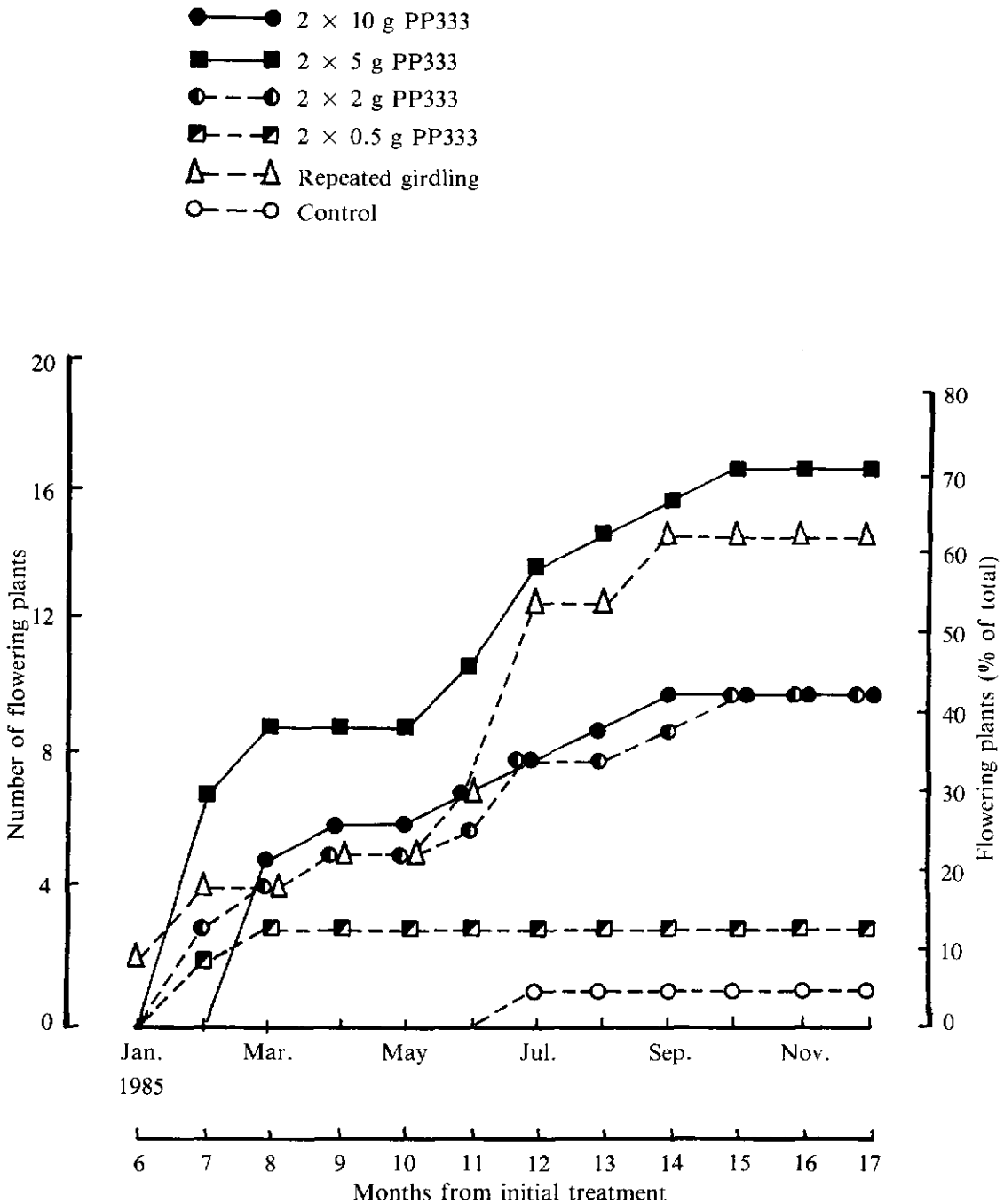

Figure 5. Cumulative incidence of flowering in GT 1 plants in the field. Fourteen-month-old plants (twenty-four per treatment) were treated with PP333 in July and October 1984. The repeated girdling treatment commenced from July 1984.

growth retarding effects on the foliage similar to those seen in polybag plants were observed. Lateral shoots failed to extend and closely packed clumps of leaves with short petioles were held very close to the main stems *(Figure 6).*

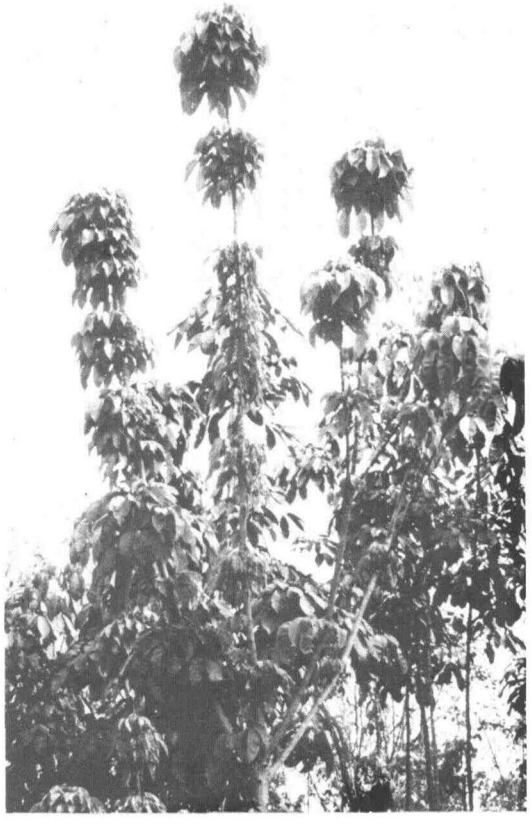

Figure 6. Growth habit of foliage in a two-year-old GT 1 plant in the field treated with PP333 showing the clumping effect on the leaves.

As in flowering induced by girdling[2], flowering induced by PP333 in both the field experiments appeared to be synchronised to the seasonal cycle of mature trees. Flowering was most evident during the two annual flowering seasons in February/March and July/August *(Figure 7).* Flowers were also observed at other times of the year, but in smaller numbers.

Pollen and Seed Germination

Over 6000 pollen grains from RRIM 600 flowers induced by PP333 were scored for germination and matched with a similar number of pollen grains from untreated mature trees of the same clone which served as controls. Pollen germination was generally poor, possibly due to the uncharacteristically wet weather. The rate of germination ranged from 0% to 33% for control pollen and 0% to 84% for pollen derived from induced flowers. The mean germination rates were 8% and 15% respectively.

Hand-pollination using the PP333-induced flowers was not attempted but fruit-set from free pollination was readily observed, particularly in the RRIM 600 plants *(Figure 3).* Out of seventy-two seeds harvested from RRIM 600 plants treated with PP333, thirty-two germinated giving a germination rate of 44.4%.

DISCUSSION

The results of this preliminary study show that PP333 induces precocious flowering in ungirdled young rubber plants. The chemical compound was effective both with polybag plants (which do not respond well to girdling) and plants in the field. In the latter, an application of 10 g a.i. — applied as a single dose (in RRIM 600) or two split doses of 5 g (in GT 1) — was most effective in bringing young rubber plants into flowering. Susceptibility to the growth retarding effect of PP333 in vegetative and floral development differred between the clones used in the study, but there was insufficient evidence to indicate if this was a clonal characteristic. The severity of retardation exhibited did not, however, necessarily reflect the effectiveness in the induction of flowering. The seasonality of response to floral induction suggested an interaction between stimulus by PP333 and climatic factors. Other factors (soil types, rootstocks, shade, *etc.)* might also have affected PP333 response and this could have given rise to the situation in RRIM 600 *(Figure 2)* where consistent correspondence between flowering responses and the chemical dosages applied was lacking.

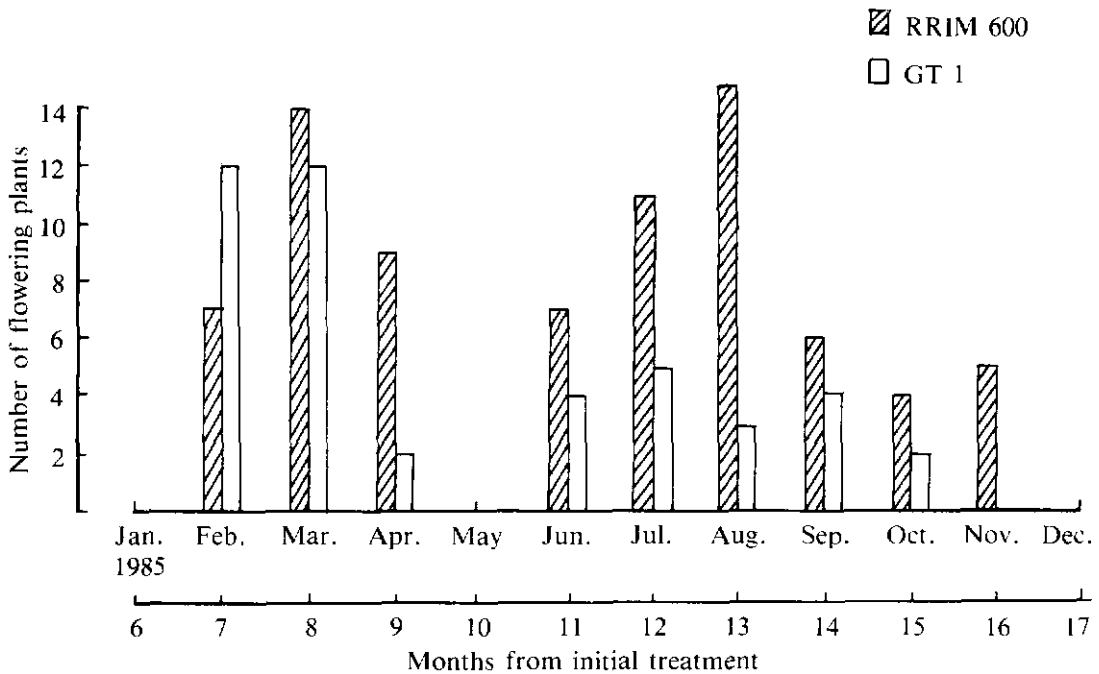

Figure 7. Seasonal variation in precocious flowering induced by PP333. Results combined from all dosages of PP333 applied and are based on a single observation per month.

Pollen germination was generally poor in samples from PP333-treated plants, but this could have been a pathological problem linked to the very wet weather experienced. In fact, pollen from control mature trees fared even worse although no significance should be attached to this observation in view of the overall low germination rate. There was hence no evidence from the available data of adverse effect on pollen germination arising from PP333 treatment.

Seed germination was moderately low in harvests from PP333-treated plants. Nevertheless, the figure of 44% is comparable to some of the germination rates in seeds obtained from girdled trees in previous studies[1,2].

The results from this preliminary study show that PP333 may be kept in view as a potential alternative to stem or branch girdling as a means of inducing precocious flowering to aid *Hevea* breeding. The effectiveness of PP333 in flower induction, the simplicity of its application and its ability to retard growth (thereby keeping the plant short and amenable to pathogen control and hand-pollination from the ground) are points in its favour. However, further evaluation of the chemical needs to be carried out before firm conclusions can be drawn.

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