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Growth Pattern of *Stylosanthes hamata* cv. Verano under Controlled Environmental Conditions

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ABSTRACT

The growth patterns and development of *Stylosanthes hamata* cv. Verano were studied in the Controlled enviroment at DSIR, Palmerston North from seedling emergince to full flowering at 131 days. The results showed that the growth and development of intact Verano were slow at the pre-flowering stage and increased rapidly after the onset of flowering. Maximum growth rate of 2.04 grams/plant/day was recorded between 70 and 80 days and maximum dry weight of 105 grams/plant based on the predicted growth model, was acheived approximately 108 days after seedling emergence. During this post-flowering stage, plant growth in terms of plant dry weight, branch development, leaf number and leaf area increased rapidly.

Flowering commenced 35 days after seedling emergence and continued throughout the experimental period. Stem was the major plant component, followed by the inflorescence and leaf fractions.

INTRODUCTION

There are few publications on the growth and development of Verano stylo (*Stylosanthes hamata* cv. Verano). Gardener (1978) measured the relative growth rate and dry weight increments of a number of *Stylosanthes* species including Verano stylo over a period of six weeks after sowing. He found that relative growth rate and dry weight increments in Verano stylo were higher than those of perennial stylo species like *Stylosanthes scabra* but similar to that of *S. humilis*. However, the relative growth rate during this period was still low compared with that of C_4 grasses (e.g. *Panicum maximum*) (Ludlow and Wilson, 1970). This could be a problem when Verano stylo is grown with such vigorous companion grasses. Gardener *et al.*, (1982) studied the changes in total dry weight and its components from a pure stand of Verano stylo over two growing seasons. Total dry weight production reached a peak a about 128 days after sowing. The contribution of the leaf fraction to dry matter yields was high during the early stages of growth, before declining throughout the wet season, with the subsequent major contribution coming from stem tissue.

This experiment reports the uninterrupted growth and development of Verano stylo under conditions of adequate soil moisture and nutrients over a period of 131 days in the controlled environment of a growth room. It includes various aspects of plant morphological development, including branch numbers and plant height, and describes the increase and distribution of dry matter yield and leaf area development.

MATERIALS AND METHODS

Environmental conditions and planting procedures.

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The experiment was carried out in the Controlled Climate Rooms at the Plant Physiology Division, DSIR, Palmerston North, New Zealand.

Plastic pots of 16 cm diameter and 16 cm depth, with drainage holes at the base were filled with 5.5 kg of a mixture of peat, vermiculite and gravel (15 : 15 : 70). *Stylosanthes hamata* cultivar Verano (Verano stylo) seeds collected from North-Eastern Thailand were selected for similar size and scarified with concentrated sulphuric acid for 30 minutes, then washed thoroughly and dried. Seeds were incubated in a germinator for 48 hours at 20-30°C before planting. Ten germinated seeds were sown in each pot, covered with a thin layer of soil, and watered. Pots were placed directly in the controlled environment room. Environmental conditions imposed were :

Photoperiod	- 12 hours
Light intensity	- 160 Wn^{-2} (400-700 nm)
Temperature	- 30°C day and 24°C night
Humidity	- 70 day and 90 night
Carbon dioxide level	= 290-340 ppm

These environmental conditions were chosen, following reference to climatic records for the Central Plain region of Thailand, to simulate as closely as possible the climate prevailing during the growing season.

A complete nutrient solution was applied twice daily for the first three weeks after planting. Watering frequency was then increased to four times per day until the end of the experiment. After three weeks of growth, some 'tip burn' was observed. To minimise this condition, pots were flushed through with deionized water every week to remove nutrients accumulating in the pot. Rhizobial bacteria (in agar form) were applied to the soil surface one week after sowing. Seedling were thinned to three plants per pot, after two weeks and finally to one plant per pot.

Harvesting schedule

Destructive harvests of the entire plant were made on days 7, 14, 21, 28, 39, 60, 67, 81, 88, 117 and 131 days after seedling emergence. Four plants were removed for dissection on each occasion.

Plant measurements

At each harvest, plants were separated into leaf, stem and inflorescence. The samples were dried in a vacuum air oven for 48-72 hours and weighed. The number of leaves, branches and inflorescences were recorded. Leaf area was also measured using an electronic Leaf Area Meter (Model 3100 Area Meter). Soil was washed from the root system and the tap root (including large woody root) and fibrous roots were separated and weighed after drying.

RESULTS AND DISCUSSION

Stage of growth

Baldos and Javier (1976) recognised three sharply defined development phases in the growth of Townsville stylo (*Stylosanthes humilil*) : juvenile, vegetative and reproductive. In contrast, Verano stylo, a quantitative short-day plant, had no sharp destinction between the vegetative and reproductive phase of development, as shown by Cameron and Mannetje (1977). The commencement of first flowering was also early compared with that of other tropical legumes like *S. guianensis* (Loch *et al.*, 1976) and *S. humilis* (Baldos and javier, 1976). Similar results were observed in the present work and also showed that the juvenile and vegetative stages were difficult to distinguish. Thus, on morphological grounds, growth in this study was divided into two stages, viz pre-flowering and flowering. Pre-flowering covered both the so-called juvenile and vegetative stage while the flowering stage covered the period after the onset of flowering. In this study, the pre-flowering stage extended over the first 35 days of growth, which is shorter than that recorded under field conditions in Queensland (Skerman, 1977; Wilaipon and Humphreys, 1976). These authors reported the pre-flowering period to be about 60-67 days. However, under field conditions in Thailand Wilaipon and

Humphreys (1981) found that Verano stylo had a somewhat shorter pre-flowering period of approximately 40 days from seeding emergence. Such variation may in part be explained by the differences in photoperiod. Under field conditions, day-length varied from month to month while in the controlled room a constant 12 hrs daylength was employed. Cameron and Mannetje (1977) found that under controlled conditions, Verano stylo flowered at all photoperiods tested, but it occurred much earlier in the shorter photoperiods of 10 and 11.5 hrs. In their experiment, Verano stylo flowered 32 days after sowing at 11.5 hrs photoperiod and 32°/24°C (day/night) -which is similar to the present experiment.

Plants at the pre-flowering stage had a slower rate of growth and produced fewer branches and leaves, accompanied by only small increments in plant height compared with the flowering stage. However seed germination was rapid and the production of the first trifoliate leaf was observed within 7 days.

The major portion of dry weight accumulation during the life of the plant occurred after the onset of flowering and included vegetative and reproductive growth. This was also observed by Cameron and Mannetje (1977) and Wilaipon *et al.,* (1979). In this study, flowering continued throughout this stage from 35 to 131 days.

Branch number

Branching of the main stem commenced in the axils of the cotyledons approximately 21 days after emergence. Over a subsequent 2-3 weed period, branch numbers increased only slowly at about 3 branches per week. At the onset of flowering, when secondary branch formation commenced, the rate of branch production rose sharply (Table 1). Branching progressed up the main stem and in this study, the terminal growing point developed an inflorescence after the formation of 11 nodes. This was similar to the figure given by Cameron and Mannetje (1977).

Table 1. Number of branches per plant and plant height of Verano stylo

Days after seedling emergence	Number of branches per plant	Plant height (cm)
7	0	2.6 ± 0.39
14	0	3.4 ± 0.85
21	2 ± 0	4.7 ± 0.29
28	6 ± 0.5	16.4 ± 0.54
39[1]	46 ± 12.6	28.9 ± 0.83
60	317 ± 130.1	64.2 ± 0.79
67	596 ± 148.5	72.7 ± 2.32
81	1040 ± 190.6	90.5 ± 6.81
88	1147 ± 206.8	92.6 ± 4.10
117	1823 ± 666.7	112.2 ± 4.78
131	1505 ± 348.0	114.9 ± 3.40

[1]50% of the plant population flowering

Branch numbers rapidly increased as primary, secondary and higher order branches appeared. As a result, the rate of branch appearance was highest between day 60 and 67 at 39 branches per day. Total branches per plant reached a maximum of 1823 at day 117. The decline in the number of branches at a later stage (day 131) was due to shedding of the small and older reproductive branches.

The results demonstrate that a high rate of branch differentiation continues during flowering in Verano stylo. It also shows that this cultivar is capable of producing large numbers of branches and greater than some other tropical legumes, although results reported elsewhere vary considerably. For example, with Townsville stylo, Baldos and Javier (1976) reported 845 branches/plant 168 days after sowing, with a maximum rate of branch appearance of 16 branches per day between day 112 and 140. Loch and Humphreys (1970), also working with Townsville stylo, reported a maximum rate of branch appearance of only 1.1 branches per day between days 89 and 104. For Verano stylo, Wilaipon *et al.* (1979)

reported that the maximum total branch density was 4980 branches/m^2 at 96 days after sowing, under glasshouse conditions. The high number of branches produced in the present experiment indicates that Verano stylo has a high potential for bud production for subsequent growth. A close relationship (R = 0.952**) be between branch numbers and plant dry weight, suggests that grazing or cutting management should aim at encouraging branch development.

Plant height

The slow increase in plant height during the pre-flowering stage is shown in Table 1 However, there was a steep increase in plant height just before the plant flowered and a maximum height of 1.2 m was measured on day 131. This was similar to field values obtained for Verano stylo in Queensland by Clements (1980) under good growing conditions. The initial show increase of plant height in Verano stylo compared with the more rapid increase in height of associated tropical grasses at the same stage of growth confers something of a competitive disadvantage on Verano stylo under mixed pasture conditions. (Gardener, 1978 and Udchachon, 1985).

Growth analysis

The relationship between total plant dry weight and time was tested using both the logistic and the cubic polynomial model (Evans, 1972). The results suggest that neither model describes the entire growth pattern of the legume accurately. The logistic growth model over-estimated total plant dry weight during the early stages of growth while at the late stage (days 117-131) the model predicted an increase in plant dry weight with time. Thus, maximum yield was unable to be obtained from the logistic curve. However, with the polynomial, the growth of the plant was under-estimated during the early stages of growth (0 – 28 days). These led to the conclusion that the entire growth pattern of this species in the present experiment could not be described using only one model. Hunt and Parsons (1977) suggested that a lengthy and complex series of data should be approached by the segmentation of the growth curve.

The relationship between plant dry weight and time was therefore described using two equations. The first section of the data, up to 50% flowering (0-39 days), was best fitted with a linear equation while the remainder gave the best fit with a cubic polynomial (Figure 1).

This latter equation was able to describe the rapid growth phase as well as the later decline in total plant dry weight.

To obtain more information on the growth pattern of Verano stylo, absolute growth rate was calculated at 10 day intervals

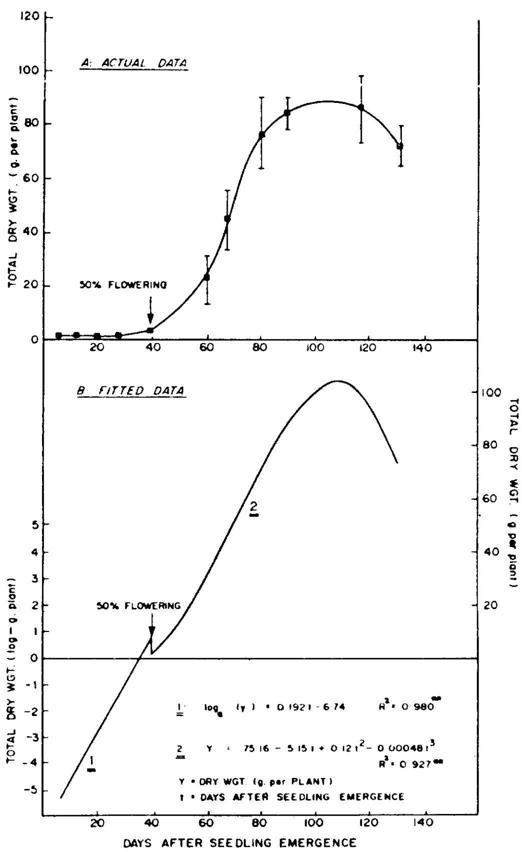

Figure 1 . Changes in total dry weight with time: A. Actual data; B. From fitted data; 1 and 2 represent line of best fit between each phase of growth

using the above equations. The pattern of growth rate at the pre-flowering stage was slow and increased rapidly after the onset of flowering. The highest absolute growth rate occurred between 70 and 80 days after seedling emergence at 2.04 g per day (Figure 2.). Maximum total plant dry weight of 105 g per plant was obtained 108 days after seeding empergence (Figure 1.) using the predicted growth curve. Absolute growth rate decreased rapidly to zero between 110 and 120 days. The relative growth rate calculated using the two equations, is shown in Figure 2. In common with the observation for most plant species, relative growth rate declined as the plant aged, related to the decreasing proportion of actively growing tissues.

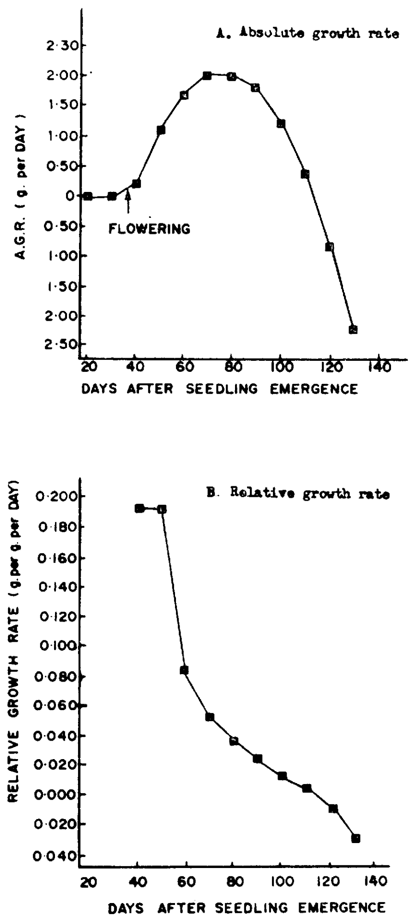

Figure 2. Absolute (A) and Relative growth rate of Verano stylo calculated from fitted growth model (Figure 1)

Dry matter yield

The growth and development of plant components are presented in Figure 3. Leaf and stem fractions increased slowly during the pre-flowering stage (0-39 days). Then all components, including inflorescences, increased significantly with the onset of flowering, despite major differences in the rate of growth for each component. For example, during the first four weeks following the onset of flowering (i.e. to day 63 approximately), all components increased substantially with the maximum rate of increase occurring in the stem fraction, which continued to increase significantly. The leaf fraction increased at a much slower rate during the 4 weeks from flowering, as did the root fraction. After this time, both the leaf and root fractions remained relatively constant until the end of the experiment. All components reached a maximum dry weight by day 117, and then either remained constant or declined to the end of the experiment. Figure 3 clearly shows that under uninterrupted growth stem is the main plant component, followed by the inflorescence fraction. The two contributed more than 50% of total plant dry weight.

Although Verano stylo had a high rate of leaf and branch differentiation after the onset of the flowering, total leaf dry weight (Figure 3) did not show the same trend as leaf number (Table 2) and branch number (Table 1) beyond day 67. This could be explained by reduced dry weight per leaf from day 39, by reduced specific leaf area from days 88 and through an observed increase in leaf senescence (leaf number) near the end of the experimental period (Table 2).

The inflorescence fraction dry weight was strongly related to inflorescence number (R = 0.962**) rather than to the size of the individual

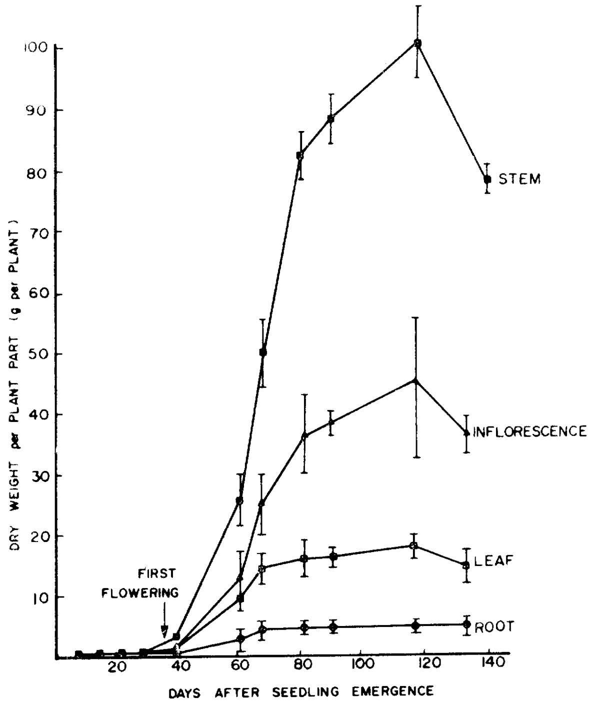

Figure 3. Dry matter yield of leaf, stem, inflorescence and roots of Verano stylo (g/plant).

Table 2. Leaf number, leaf size, dry weight per leaf and specific leaf area.

Days after seedling emergence	Number of leaves per plant[1]	Leaf size (cm^2/leaf)	Leaf dry weight (mg/leaf)	Specific leaf area (cm^2/g)
7	1	0.66	5	132.0
14	3	0.86	5	184.3
21	5	1.28	7	205.8
28	18	1.71	9	197.5
39[2]	107	3.15	15	210.1
60	538	2.78	12	227.6
67	984	2.39	11	227.0
81	1420	2.18	9	241.0
88	1424	2.10	8	254.3
117	1946	1.58	7	224.4
131	1634	1.32	7	202.2

[1] A trifoliate leaf.

[2] 50% of the plant population flowering

inflorescence (R = 0.453*). The decline in inflorescence dry weight over the final 16 days is explained by the reduction in inflorescence number over this period. In addition, shedding of older inflorescences could also be important, as demonstrated by Gardener *et al.*,(1982).

Growth of roots during the seedling stage (0-28 days) was mainly due to elongation of tap root, but with few branch roots. A more rapid growth rate followed immediately after the onset of flowering (day 35) and significantly increased until day 67. Thereafter, root dry weight changed little throughout the experiment (Figure 3.). As a result, the shoot to root ratio increased from 3 on day 7 to 22 on day 117. Leaf area (cm^2/plant), number of leaves and specific leaf area (cm^2/ g)

Leaf characteristics of Verano stylo plants are presented in Table 2 and Figure 4. The number of leaves and total leaf area did not increase during the pre-flowering stage of growth. However, from the onset of flowering, the number of leaves increased rapidly from day 28 up to day 117. Leaf area also increased rapidly from day 28 but reached a maximum on day 81-showing that attainment of maixmum leaf area does not necessarily depend on leaf number, as leaf size and specific leaf area are also important in determining leaf area production. The reduction in leaf number from day 117, due presumably to leaf senescence, was probably the main contributor to the reduction in leaf area at that stage.

Although leaf area index could not be calculated, the maximum leaf area of 3097 cm/plant was noted on day 81. However, the maximum rate of leaf appearance of 63.2 leaves per plant per day was obtained between day 60 and day 67. The results of this study demonstrate that Verano stylo has a high rate of leaf differentiation following the onset of the flowering. Wilaipon *et al.*, (1979) also showed that a leaf density of 35500 per m^{-2} occurred 96 days after sowing under glasshouse conditions. However,

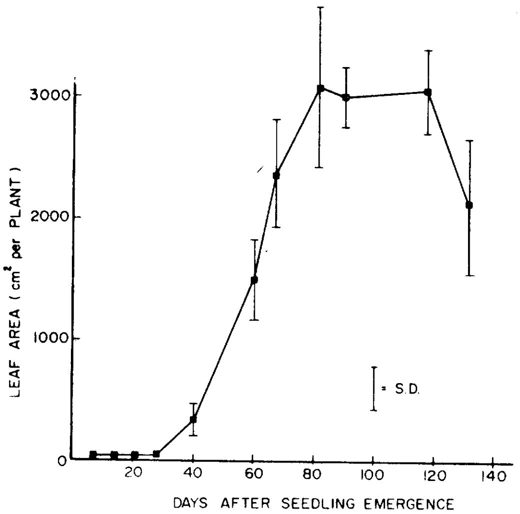

Figure 4 . Changes in total leaf area of Verano stylo with time (cm²/plant).

these results were not shown on a per plant basis.

Growth and development of Verano stylo in the controlled environment clearly showed the highly productive capacity of the species over the experimental period of 131 days. However, early growth in terms of dry matter yield, leaf area, plant height and number of branches, was relatively slow up to the onset of flowering. Nevertheless, growth of the plant increased rapidly after the onset of flowering and reached a maximum absolute growth rate around day 80 which closely approximated the attainment of maximum leaf area. Maximum dry matter production of 105g/plant based on the predicted growth model, occurred on day 108 after seedling emergence. This increase in plant dry weight was mainly through the stem and inflorescence fractions and to a lesser extent the leaves. Beyond the 108 day period production noticeably declined due to ageing processes with the stem fraction remaining the dominant component.

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LITERATURE CITED

Baldos, D.P. and E.Q. 1976. The growth and development of Townsville stylo (*Stylosanthes humilis* H.B.K.). Philippine J. of Crop Sci. 1 : 209-13.

Cameron, D.F. and L.T. Mannetje, 1977. Effects of photoperiod and temperature on flowering of twelve *Stylosanthes* species. Aust. J. of Expt. Agric. and Anim. Husb. 17 : 417-24.

Clements, R.J. 1980. Personal Communication.

Evans, G.C. 1972. The quantitative analysis of plant growth. I. Studies in ecology. Blackwell Scient. Publ., Oxford.

Gardener, C.J. 1978. Seedling growth characteristics of *Stylosanthes*. Aust. J. of Agic. Res. 29 : 803-13.

Gardener, C.J., R.G. Megarrity and M.N. Mcleod. 1982. Seasonal changes in proportion and quality of plant parts of nine *Stylosanthes* lines. Aust. J. of Expt. Agric. and Anim. Husb. 22 : 391-345.

Hunt, R. and I.T. Parsons, 1977. Plant growth analysis : further applications of a recent curve-fitting program. J. App. Ecolo. 14 : 965-68.

Ludow, W.W. and G.L. Wilson, 1970. Growth of some tropical grasses and legumes at two temperatures. J. of Aust. Inst. of Agric. Sci. 36 : 43.

Loch, D.S. and L.R. Humphreys. 1970. Effects of stage of defoliation on seed production and growth of *Stylosanthes humilis*. Aust. J. of Exp. Agric. and Ani. Husbandry 10 : 577-81.

Skerman, P.J. 1977. Tropical Forage Legumes. F.A.O. Rome.

Unchachon, S. 1985. Master of Agricultural Science. University of Massey, Palmerston North, New Zealand.

Wilaipon, B. and L.R. Humphreys. 1981. Influence of grazing on the seed production of *Stylosanthes hamata* cv. Verano. Thai J. of Agric. Sci. 14 : 69-81.

Wilaipon, B., S.A. Gigir. and L.R. Humphreys, 1979. Apex lamina and shoot removal effects on seed production and growth of *Stylosanthes hamata* cv. Verano. Aust. J. of Agricultural Research 30:293-306.