

Project title: *Osteospermum*: Examination of the use of chilling treatments on a range of cultivars and evaluation of the effect of interrupted periods of warmer temperatures on floral initiation and subsequent flowering.

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PRACTICAL SECTION FOR GROWERS

Objectives and background

Cape daisies (*Osteospermum jucundum*) are an increasingly important pot/bedding plant species grown as a half-hardy perennial in the UK. Current estimates suggest that up to two million plants are grown per annum, but there still is little information available regarding the effects of environment on the time to flowering and on overall plant quality. Information on this would be very useful to growers, since the crop is considered to be highly responsive to temperature, in terms of both time to flowering initiation and overall development. Previously HDC-funded work has shown that brief periods of chilling (9°C for 2 weeks) can dramatically hasten the time to flower initiation, whilst high temperatures shorten the duration of post-initiation flower development. This new work was commissioned to further investigate these chilling responses. The work was conducted at three sites; HRI Efford, The University of Reading and Anglia Alpines Ltd. HRI Efford conducted experiments (Part I of this report) to;

- assess chilling responses under greenhouse conditions (previous experiments were only in growth cabinets);
- test whether high temperature interruptions to chilling treatments negates the response.
- examine the response of a more representative range of cultivars.

Research at the University of Reading focussed on details of the temperature response (Part II of this report), examining;

- the optimum temperature for chilling;
- the duration of chilling required;
- the effects of photoperiod during chilling;
- the effects of temperature and photoperiod post-initiation.

The work at Anglia Alpines Ltd (Part III of this report) aimed to confirm the studies conducted at both Efford and Reading under commercial conditions, including the use of plant growth regulators.

Summary of results

HRI Efford

The cultivars assessed in the Efford work were; 'Lubutu', 'Lindi', 'Sunny Lady', 'Pink Fantasy', 'Zulu', 'Sunny Girl', 'Sunny Gustav' and 'Lusaka'. These cultivars represented a range of sensitivity to chilling treatments applied post-pinching, with 'Lubutu', 'Lindi', 'Sunny Lady', 'Pink Fantasy' and 'Zulu' being most sensitive; 'Lusaka' and 'Sunny Gustav' showing almost no response and 'Sunny Girl' with an intermediate response. However in all cultivars the response trends were the same and chilling did reduce time to first anthesis and to marketing. Chilling treatments also reduced the number of leaves and thus the height of plants by cutting down on the number of internodes. Increases in the numbers of flowers initiated were less obvious, although examination of flower initiation in cv. 'Zulu' by dissections and microscope observation showed that chilling did increase this response.

The length of the chilling period was important and if plants were held at 9°C for 7 days the effects of chilling could be reversed by a short period at 22°C. However, after longer periods of chilling (14 or 21 days) the chilling effect apparently became irreversible. When plants were not chilled, but experienced a period of high temperature post-pinching (22°C for 7, 14 or 21 days), their mode of growth became predominantly vegetative and they produced large numbers of leaves, a relatively tall canopy and only came into flower very late (in some cases, plants were not flowering at all by the end of the trial).

The University of Reading

The effects of temperature and photoperiod on time to inflorescence commitment and the rate of progress of inflorescence development of cv. 'Zulu' were investigated. A reciprocal transfer experiment between cool (12°C) and warm (22°C) temperatures showed that plants could be induced to flower by chilling immediately after pinching, and that 11 days at 12°C were sufficient for inflorescence commitment, whilst 68 days were required at 22°C. However, subsequent inflorescence development was then hastened under the warmer temperature regime.

A further experiment on cv. 'Zulu', to study the effects of temperature and photoperiod on the time of inflorescence commitment provided no evidence to suggest that photoperiod affected the time of inflorescence commitment. However, plants grown at 10.6°C were committed to flower 4 days before plants at 13.7°C, indicating a significant effect of temperature and suggesting a relatively low optimum temperature for inflorescence commitment. Plants grown at mean temperatures greater than 18°C had not initiated flowers after 28 days. The data presented here suggest that *Osteospermum* has a low optimum temperature for inflorescence commitment rather than a true vernalisation response.

The effects of temperature and photoperiod on inflorescence development were quite different to those for inflorescence commitment. Once initiated, high temperatures and long days hastened inflorescence development. The rate of progress to flowering from initiation was found to increase linearly with photoperiod and temperature such that plants grown under an 8 h d⁻¹ photoperiod flowered 55 days earlier at 22.5°C compared to 11°C. There was also some indication of an optimum temperature at 23.5°C, a slight slowing in the rate of progress to flowering being seen in plants grown at higher temperatures. The rate of progress to flowering also increased linearly with longer days, such that plants grown at an average temperature of 19.3°C flowered a week earlier when the daylength was extended from 8 to 17 h d⁻¹, indicating a long day response for this developmental stage.

Anglia Alpines Ltd

'Short' (7 days), 'medium' (14 days) and 'long' (21 days) chilling periods at 9°C were compared with 'short', 'medium' and 'long' periods at 22° post-pinch and with a constant 16°C 'ambient' control treatment. All plants also received growth regulator ('Cycocel' = chlormequat) applied as a drench. All treatments produced good marketable plants. However, the best results in terms of both quality and speed of production were obtained with 14 days of post-pinch chilling at 9°C. This treatment reduced the production time over all other treatments by some 4-7 days. The commercial advantage of 100s flowering at an earlier date in this case could give an extra return per pot of some 30-50p, whilst allowing for production costs due to an 8% reduction in time over a 92 day schedule.

Action points for growers

- There were wide differences between cultivars of *Osteospermum* in their degree of response to chilling treatments.
- Chilling treatments applied immediately after pinching are definitely worth considering as part of future schedules as they can effectively reduce plant height and increase flower number, whilst reducing production times. However, further work is needed to provide accurate scheduling information for individual cultivars.
- Chilling time is important and shorter periods (ie when inflorescence commitment is not completed) may be reversible by the effects of short high temperature interruptions.

GENERAL INTRODUCTION

Osteospermum jucundum is a half hardy perennial, originating from South Africa, which is becoming an increasingly popular garden/pot plant. It is estimated that up to two million plants are produced per annum in the UK. However, little is known about the environmental responses of this species. Indeed, it is considered to be highly responsive to temperature and this produces great and hitherto unpredictable variations in flowering and plant quality.

Pearson *et al.* (1995) examined the effect of temperature and photoperiod on the reproductive development of *Osteospermum jucundum* cv. 'Pink Whirls' and showed that it was a long day plant, requiring at least two weeks of chilling at 12°C to induce flowering. Pearson *et al.* (1995) also examined the effects of a range of chilling temperatures in growth cabinet experiments on the cultivars 'Lubutu' and 'Sunny Girl'. 'Lubutu' showed a strong response to chilling, with flowering occurring 3 weeks earlier when plants were maintained at 9°C for two weeks compared to 15°C. Plants of cv. 'Sunny Girl' did not appear to respond to chilling but this was considered to be due to the presence of pre-induced flowers prior to the start of the experiment which obscured treatment differences (the cuttings had been pre-pinned). A similar flowering response to cold temperatures has been observed in the closely related species *Osteospermum ecklonis* (Hendricks and Baumann, 1992). Other work on manipulating the growth of *O. jucundum* has tended to concentrate on the use of growth regulators: optimising their use and assessing the post-production quality of plants (Olsen and Andersen, 1995; Fuller, 1997).

The central aim of this project was to increase our understanding of the response of *Osteospermum jucundum* to the environment, with particular reference to the effects of temperature and to a lesser extent to photoperiod, and with a view to improving plant scheduling and production quality. The study followed three simultaneous lines of enquiry:

- i) an assessment of the impact of chilling treatments on flowering and production times of a range of 8 cultivars under simulated commercial production conditions at HRI Efford;
- ii) a detailed examination of the effects of temperature and photoperiod on the flowering response of representative cv. 'Zulu' using controlled environment facilities at the University of Reading;
- ii) an assessment of the potential interaction between the use of chemical growth regulators and the use of chilling treatments on flowering of cv. 'Zulu' under commercial conditions at Anglia Alpines Ltd.

The objectives of this study were to:

at HRI Efford (Part I);

- assess chilling responses under greenhouse conditions (previous experiments were carried out in growth cabinets only);
- test whether high temperature interruptions to the chilling period negate the chilling response;
- examine the impact of chilling treatments on a widened range of cultivars;

at the University of Reading (Part II);

- determine the optimum temperature for chilling;
- determine the duration of chilling required for a response;
- assess the effects of photoperiod during chilling;
- assess the effects of temperature and photoperiod post-initiation;

at Anglia Alpines Ltd (Part III);

- confirm results of studies carried out at both HRI Efford and the University of Reading under commercial conditions including the use of growth regulator.

PART I: HRI EFFORD

GREENHOUSE ASSESSMENT OF THE IMPACT OF CHILLING TREATMENTS ON FLORAL INITIATION AND FLOWERING IN A RANGE OF CULTIVARS OF *OSTEOSPERMUM*

INTRODUCTION

Commercially-grown cultivars of *Osteospermum* are known to respond to different degrees to environmental stimuli. Experimental work at HRI Efford was designed to assess the response of a range of *Osteospermum jucundum* cultivars to chilling treatments in a commercial-scale greenhouse environment. Under commercial conditions it was also thought possible that short durations of warmer temperatures might occur during chilling treatments which might have a negative 'de-chilling' effect, reversing the benefits of the chilling treatment. In order to determine the importance of this, the impact of short durations of warm temperature (22°C), imposed at the end of the chilling treatment (9°C), on flower initiation, development and plant size was assessed.

MATERIALS AND METHODS

Site

Propagation - H block, southern compartment.
Growing on - Q block, compartments 1, 2 and 3.

Start Material

Unrooted cuttings were supplied by a commercial propagator in Denmark (Karl Axel Sorensen). 1750 plants of each of the following cultivars were grown; 'Lubutu', 'Zulu', 'Pink Fantasy', 'Sunny Lady', 'Sunny Girl', 'Sunny Gustav', 'Lindi' & 'Lusaka'.

Treatments

The following 16 treatments were applied using the schedule illustrated in Figure 1;

1. 16°C throughout = 'ambient' control;
2. 16°C, 9°C for 7 days = 'short duration chilling';
3. 16°C, 9°C for 14 days = 'medium duration chilling';
4. 16°C, 9°C for 21 days = 'long duration chilling';
5. 16°C, 9°C for 7 days, 22°C for 3 days = 'short chilling + short warm interruption';
6. 16°C, 9°C for 7 days, 22°C for 6 days = 'short chilling + medium warm interruption';
7. 16°C, 9°C for 7 days, 22°C for 9 days = 'short chilling + long warm interruption';
8. 16°C, 9°C for 14 days, 22°C for 3 days = 'medium chilling + short warm interruption';
9. 16°C, 9°C for 14 days, 22°C for 6 days = 'medium chilling + medium warm interruption';
10. 16°C, 9°C for 14 days, 22°C for 9 days = 'medium chilling + long warm interruption';
11. 16°C, 9°C for 21 days, 22°C for 3 days = 'long chilling + short warm interruption';
12. 16°C, 9°C for 21 days, 22°C for 6 days = 'long chilling + medium warm interruption';
13. 16°C, 9°C for 21 days, 22°C for 9 days = 'long chilling + long warm interruption';
14. 16°C, 22°C for 7 days = 'short duration warming';
15. 16°C, 22°C for 14 days = 'medium duration warming';
16. 16°C, 22°C for 21 days = 'long duration warming'.

Experimental Design

8	Cultivars	6	Cultivars
x		x	
13	treatments (1-13)	3	treatments (14-16)
x		x	
3	replicates	3	replicates
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312	plots	54	plots

Total = 366 plots, 17 plants per plot of which 6 were recorded.

The layouts of treatments within the glasshouse compartments are given in Appendix I.

Cultural Details

Unrooted cuttings, bought in from C.A. Krage Sorensen, Denmark, were direct stuck into Teku PT104 cell plug trays containing Fisons Levington FI medium on 30 February 1996. Cuttings were dipped in Seradix rooting hormone prior to sticking. Trays were placed onto heated benches, maintaining a base heat temperature of 20°C. Following sticking, cuttings were lightly

watered in with plain water followed by a Bavistin drench. Clear polyethylene sheeting, supported on low metal hoops, was then used to cover the cuttings and maintain a high RH during root establishment.

The initial glasshouse temperature was 18°C, this was reduced to 16°C once cuttings had rooted, with the base heat also being dropped to 16°C. Supplementary lighting was used throughout the propagation period at 2500 - 300 lux for 16 hours per day.

A preventative spray of Rovral was applied 2 weeks after sticking. Four weeks after sticking, cuttings were pinched to 4-5 leaf nodes.

Once pinched, plants were potted on into 12F plastic pots using Fisons Levington M2 medium with 10% grit added. Plants were then grown on at 16°C for 7 days before commencing the main temperature treatments (9°C, 16°C and 22°C). At this point all plants received a Fongarid drench. After this liquid feeding was applied at every watering, providing 175N : 26P : 200K : 28Mg plus additional iron. The pH of the applied water was maintained at pH 6.0 using additions of nitric acid.

A detailed crop diary is presented in Appendix II.

Experimental records

Initiation studies

These records were carried out by staff from both the University of Reading and from HRI Efford. Collected at regular intervals, samples of cv. 'Zulu' from all 16 treatments, were dissected to determine the commencement of floral initiation and potentially its reversal.

During growth of crop

Days to first anthesis

At Marketable stage of growth

Plant height in mm. (a) canopy
(b) flower head

Branch number.

Leaf number.

Bud and Flower number.

Flower diameter.

Days to marketable stage (3-4 open flowers with visible buds).

Photography

Photographs were taken to illustrate treatment comparisons (see Appendix III).

Analysis of compost and leaf samples

Compost samples were collected from all treatments and cultivars at potting, on day 29 prior to final spacing and at marketing. Foliage samples were collected from all treatments and cultivars at pinching, on day 29 and at marketing. Samples of applied liquid feed were collected every 2 weeks. All samples were sent for chemical analysis to the Analytical Chemistry Department at HRI Wellesbourne. The results of leaf tissue and compost analyses are presented in Appendix IV.

Environmental records

Weekly records of day/night glasshouse temperatures and relative humidity were monitored as well as levels of solar radiation received by the crop.

RESULTS

As expected, there was a large variation in cultivar response to chilling treatments. The most responsive cultivars were; 'Lubutu', 'Lindi', 'Sunny Lady', 'Pink Fantasy' and 'Zulu' and the least responsive were 'Lusaka' and 'Sunny Gustav' with 'Sunny Girl' an intermediate (see Appendix V, tables a - h). Although the size of response varied with cultivar, all cultivars showed the same trends in response to both chilling and warm temperature treatments.

The effects of chilling treatments of 9°C generally increased with the length of chilling period from 7 to 21 days. With increasing chilling period, the time to both first anthesis and to a marketable stage of development (2 - 3 open flowers and visible buds) was reduced. The most sensitive cultivar was cv. 'Lubutu' with reductions of 5.58 and 6.78 days in the time to anthesis and to marketing after 21 days at 9°C in comparison with controls grown at a constant 16°C. Chilling treatments also caused similar reductions in canopy and flower heights and in leaf numbers. Changes in flower numbers were a little less obvious, although there was a definite increase in the proportion of flowers to leaves with increasing chilling period and significantly more flowers were present after the longest chilling period than in 16°C controls in cvs. 'Sunny Lady' and 'Zulu'. Chilling treatments had no obvious effects on flower diameter and did not affect branch number.

A short period at a warm temperature (22°C) around the time for flower and leaf initiation had the opposite effect to chilling on plant size and the number of leaves produced. Plants given these treatments became largely vegetative, producing very few flowers and large amounts of foliage. The time for floral development was increased, as reflected in the increased times to first anthesis and to marketing. In some cultivars, too few flowers emerged even before the foliage started to senesce and so values for times to marketing for cultivars 'Lubutu' and 'Zulu' (the most temperature-sensitive cultivars tested) were only estimated for the statistical analysis (Appendix V, tables a & e).

Interruptions to chilling treatments with a period of warm temperature (22°C) did reverse the effects of chilling. Over the range of times tested (3 - 9 days), the length of the warm period did not appear to affect the outcome. However, the length of the preceding chilling period was very important. After 7 days chilling at 9°C, a period at 22°C as short as 3 days generally reversed the effects of chilling on all the plant growth and development parameters mentioned above. However, after between 14 and 21 days of chilling, the period of warming had no effect on the chill-induced responses and may, in some cases, even have very slightly speeded up the rate of flower development.

In the initiation studies, sample shoot primordia were dissected out of three replicate plants per treatment on the following days after sticking:

Treatment	Sample times (Days after sticking)
1	78, 81, 85, 88, 92, 95, 100, 103, 106
2	78, 81, 85, 88, 92, 95, 100, 103
3	85, 88, 92, 95, 100, 103, 106
4	92, 95, 100, 103
5	85, 88, 92, 95, 100, 103, 106
6	85, 88, 92, 95, 100, 103, 106
7	85, 92, 95, 100, 103, 106, 109
8	92, 95, 100, 103
9	92, 95, 100, 103, 106
10	92, 100, 103, 106
11	95, 100, 103
12	95, 100, 103
13	95, 100, 103
14	78, 81, 85, 88, 92, 95, 100, 103, 106, 109, 113, 116
15	85, 88, 92, 95, 100, 103, 106, 109, 113, 116
16	92, 95, 100, 103, 106, 109, 113, 116

Assessments consisted of a count of the number of leaves, including primordia, from the point of pinch and recording the presence/absence of flower initials (see Figure 3, Appendix III). These counts were prone to a small amount of error due to misidentification of some flower initials as leaf initials. Also using the presence/absence measure of flower initiation was somewhat coarse and treatments 1-13 all appeared to start flower initials in week 15 (treatments 14-16 started in weeks 16 and 17). Since for each treatment, the rate of leaf initiation was linear, regressions for this were calculated. It was then possible to estimate the time to flower initiation by extrapolation using the counts of finally fully emerged leaves. These estimated values for the time to flower initiation are presented in Table 1 (The mean leaf counts and regression analyses are in Tables i and j , Appendix V).

Table 1: Estimated times to flower initiation (from sticking), in cv. 'Zulu', following various chilling treatments.

Treatment No.	Estimated time (days) from sticking to first flower initiation
1	99.9
2	70.9
3	72.7
4	96.2
5	100.2
6	102.6
7	103.9
8	97.3
9	99.3
10	94.0
11	96.0
12	94.3
13	93.6
14	109.5
15	116.7
16	119.3

These results confirm that chilling treatments at 9°C encourage flower initiation and speed the process up. The effect of short duration chilling treatments (7 days) was reversed by the application of a short period of warm temperature (22°C). However, after longer periods of chilling (14 and 21 days), the effect of the chilling treatment on the rate of flower initiation was irreversible by the application of heat. Plants treated with a warm period without any chilling, initiated flowers at a greatly reduced rate and remained largely vegetative, producing more leaves than in the other treatments.

It was observed that the 'Sunny' varieties, particularly 'Sunny Lady', rooted quickly, whilst cv. 'Zulu' was much slower. This meant that at the start of the treatment regimes, cv. 'Zulu' received a relatively light pinch compared to cv. 'Sunny Lady', which was pinched back hard. Dissection studies at this time showed that cv. 'Sunny Lady' plants had initiated flowers in their main shoots, but that the sideshoots, which developed after pinching were all vegetative. All other cultivars were vegetative at this stage.

DISCUSSION

These results indicate that it may be possible to use a chilling treatment on a commercial scale to provide some degree of control of plant height by limiting the number of leaves. However the large amount of variation seen between cultivars in response to chilling treatments would mean that much further work would be necessary to produce reliable schedules. The length of the chilling period was essential in maintaining the stability of the response. After short periods of chilling a high temperature interruption could reverse the vernalisation effect and 'de-chill' the plants. However this reversal of the chilling response only appeared important when periods of chilling were less than 14 days. In previous work at the University of Reading a chilling period of 11 - 12 days at 12°C was required to cause the vernalisation effect. In the HRI Efford-based experiments reported here, the vernalisation process may not have been completed after 7 days at 9°C. When this 7 day period was followed by a temperature of 16°C vernalisation process set in motion could possibly have been completed. However, when the temperature was increased to 22°C, the process was possibly reversed and the plants were transferred to vegetative growth. After longer periods (ie 14 or 21 days) at 9°C, full vernalisation would have occurred before the end of the chilling period and a subsequent increase in temperature to 22°C would therefore have had little impact on subsequent development.

PART II: THE UNIVERSITY OF READING

THE EFFECTS OF TEMPERATURE AND PHOTOPERIOD ON INFLORESCENCE COMMITMENT AND SUBSEQUENT INFLORESCENCE DEVELOPMENT IN *OSTEOSPERMUM JUCUNDUM* CV. 'ZULU'.

INTRODUCTION

Research at Reading focussed on the details of the temperature/flowering response, examining;

- the optimum temperature for chilling;
- the duration of chilling required for flower commitment;
- effects of photoperiod during chilling on flower commitment;
- effects of temperature and photoperiod post-initiation on flower development.

In total, three experiments were conducted. The first was a reciprocal transfer growth cabinet experiment where plants were moved to and from cool or warm environments (12 or 22°C) to assess when flowers were induced, and the duration of cool temperature induction required. A second analogous but more extensive glasshouse experiment examined the time to flower commitment for cuttings grown in a factorial combination of six temperatures (set at 6 to 26°C) and four photoperiods (8 to 17 h d⁻¹). The third experiment examined the effects of the same six temperatures and photoperiods on the time of flower development following initiation.

MATERIALS AND METHODS

General plant culture

Un-rooted cuttings of Cape daisy (*Osteospermum jucundum* cv. 'Zulu') were obtained from a commercial propagator in Denmark (C.A. Kragh Sørensen). These were struck into Hassey 104 plug trays containing a peat-based seed and modular compost (SHL; William Sinclair Horticulture Ltd, Lincoln, UK). Plants were rooted in a glasshouse set to provide a minimum temperature of 18°C, they were initially covered with polyethylene with supplementary lighting (Mercury fluorescent) for the first week for 14 h d⁻¹ at an irradiance of 80 μmol m⁻² s⁻¹. After the plants had rooted they were subsequently potted up into 9 cm pots (volume 370 ml) containing a mixture of a peat-based potting compost (SHL; William Sinclair Horticulture Ltd) and perlite (3:1 by volume).

Plants were then pinched at the start of an experiment to reduce the chance of using pre-initiated plant material and to minimise variation between plants, as suggested for chrysanthemum by Cockshull & Hughes (1976). Once rooted, all plants were irrigated as necessary with Sangral 111 liquid feed at a conductivity of 1500 μS (182 ppm N; 78 ppm P; 150 ppm K), acidified to a pH of 5.8.

Experiment 1: Reciprocal transfer

A reciprocal transfer experiment between cool (12°C) and warm (22°C) environments was used to assess the chilling requirement of *Osteospermum jucundum* cv. 'Zulu' for flowering and indicate the stage of development when plants become sensitive to chilling after pinching.

On the 29 April 1996, plants were pinched 11 days after potting and moved to growth cabinets (Fisons Gallenkamp 770 and Fisons Fi-totron 600), at an initial density of 100 pots m^{-2} , these were then respaced as the canopy closed until a final density of 25 pots m^{-2} . Plants were transferred between the two growth cabinets set to provide constant temperatures of either 12°C or 22°C ($\pm 0.5^\circ\text{C}$). Both growth cabinets were set to provide an irradiance of 200 $\mu\text{mol m}^{-1} \text{s}^{-1}$ at plant height for 16 h d^{-1} from cool white fluorescent tubes supplemented with 20% tungsten, determined on the basis of nominal wattage.

At twice weekly intervals, five randomly selected plants were transferred to and from each growth cabinet for up to 45 days whilst a further 13 plants remained in each growth cabinet as untransferred controls. Plants were then grown either until flowering or 153 days when the experiment was terminated. The number of days to flower and the number of leaves from the pinch to the first inflorescence were recorded for each plant, or the number of leaves on the upper most lateral branch for a few plants that had not flowered by 153 days from the start of the experiment.

Experiment 2. Effect of temperature and photoperiod on inflorescence commitment.

A second experiment was conducted to assess the effect of temperature and photoperiod on the time to inflorescence commitment. The experiment was conducted using photoperiod chambers built within in a linear array of five temperature-controlled glasshouse compartments (3.7m x 7m) set to provide minimum temperatures of 6°, 10°, 14°, 18° and 22°C, with ventilation at temperatures 4°C above the set-points. The two coldest compartments were equipped with air conditioning units to maintain temperatures under high ambient temperature conditions. Mean diurnal temperatures within each compartment were recorded on a data-logger (Datataker, DT500), from measurements using aspirated PT100 temperature sensors (15 second scans, logged hourly). Each compartment was equipped with four photoperiod controlled chambers, which were sealed from exterior light sources. Plants remained in the glasshouse for 8 hours. At 16:00h each day, they were wheeled into the photoperiod chambers, where they remained until 08:00h the following morning. Day-lengths were extended inside each of the chambers by illuminating the plants with low intensity lighting at an irradiance of 11 $\mu\text{mol m}^{-2} \text{s}^{-1}$, from a 40W tungsten and a 15W compact florescent light bulb. In all treatments the lamps were switched on automatically at 16:00h for a duration dependent on the daylength extension required. To minimise any temperature increase the compartments were continuously ventilated (average air speed of 0.2m s^{-1} over the plants). The daylengths provided were 8, 11, 14 and 17 h d^{-1} . Thus, the experimental design provided a combination of five temperatures and four photoperiods. Rooted cuttings were pinched on 17 May 1996 and placed on the photoperiod trolleys while in plug trays (Hassey 104). Five randomly selected plants were then transferred on three occasions every week for four weeks from pinching, from each trolley to a

glasshouse set to provide a minimum temperature of 22°C and natural daylengths, where plants were grown on until flowering. A further 100 plants were placed in the same warm glasshouse at pinching. When all plants had been transferred they were potted up into 9cm pots. Plants remained in the second glasshouse until flowering up to a maximum of 153 days when the experiment was terminated. Data was recorded as in experiment 1.

Experiment 3. The effect of temperature and photoperiod on inflorescence development.

A further experiment was designed to assess the effect of temperature and photoperiod on subsequent inflorescence development after inflorescence initiation. This experiment was conducted in the same temperature/photoperiod facility as described in experiment 2, except that a sixth glasshouse compartment was used enabling the provision of 6 temperature regimes (minimum temperatures of 6°, 10°, 14°, 18°, 22°, 26°C) and four photoperiods (8, 11, 14 and 17h d⁻¹).

To initiate inflorescences prior to the start of the experiment plants were pinched on 18 April 1996 and grown under natural daylengths in a glasshouse compartment set to provide a minimum temperature of 6°C, with refrigerative cooling being used to reduce the temperature if above 10°C. Five randomly selected plants were subsequently dissected on two occasions every week, to assess the reproductive state of the uppermost lateral apex and the number of leaf primordia above the pinch. Apices were examined against a flowering scale devised from scanning electron micrographs. Thus, at various stages of growth, apices were fixed for examination under a scanning electron microscope (SEM). Dissected apices were fixed in glutaraldehyde 4% (v/v) in 0.1M phosphate buffer (pH 7) for 2 hours in sealed bottles at room temperature. The fixative was decanted off and the material washed with three changes of glass distilled water. The material was then fixed in 1% (w/v) aqueous osmium tetroxide (OsO₄) for 2 hours and again washed with three changes with glass distilled water. The material was then dehydrated through a graded acetone series (30%, 50%, 70%, 90%, 95%, 100% followed by absolute 100%), the samples were in each acetone dilution for 30 minutes. The material for SEM was dried using the critical point drying technique, mounted on stubs using silver paint and coated with gold using a sputterer coater. Observations were conducted with a Jeol T20 SEM at approximately 20KV.

When 100% of the dissected population were considered to have initiated an inflorescence, as indicated by a whorl of bract primordia (stage "2" of flowering; Appendix III, Figure 3c) replicate plants were placed in each of the 24 temperature/photoperiod combinations. Plants then remained under these conditions until flowering when the number of days to flower and the number of leaves from the pinch to the first inflorescence were recorded for each plant.

RESULTS

Experiment 1. Reciprocal Transfer

Plants grown continuously at 22°C flowered 7 days before those grown at 12°C, however this difference was not significant (Figure 2). However, flowering was most rapid when plants were exposed initially to between 10 and 21 days at 12°C and then grown on until flowering at 22°C. Flowering was however delayed when plants were exposed to longer periods of chilling, presumably because the process of inflorescence development was slowed at the cool temperature regime. Exposing plants to 22°C prior to transfer to 12°C, delayed flowering as inflorescence initiation was delayed at the higher temperature (Figure 2, up to transfer day 38). However, after 38 days, the hastening of flowering due to earlier inflorescence initiation (at 12°C) is offset by the delay caused in subsequent inflorescence development by this cool temperature, resulting in an overall delay in flowering when plants are transferred to 12°C after this time.

The effect of the cool and warm temperatures on inflorescence initiation and development can be seen more clearly in terms of the number of leaves initiated below the inflorescence (Figure 3). Plants grown continuously at 12°C initiated an inflorescence after 24 leaves had been initiated compared to 73 leaves recorded on plants grown at 22°C. This difference in leaf number is due to a delay in the time of inflorescence commitment and possibly an increased leaf initiation rate in the warm temperature regime. Non-linear regression, using SlideWrite Plus 2.0, was used to fit a sigmoidal curve to the leaf number data from plants transferred from 12°C to 22°C. This sigmoidal curve was chosen as it gave good fit to the experimental data and has well defined upper and lower asymptotes, thus;

Figure 2: The effect of transferring plants from 12 °C to 22°C (●) and from 22°C to 12°C (○) on the time to flowering. (Standard errors of the means are shown where larger than the points and arrows indicate the presence of a plant within a treatment which had not flowered after 153 days).

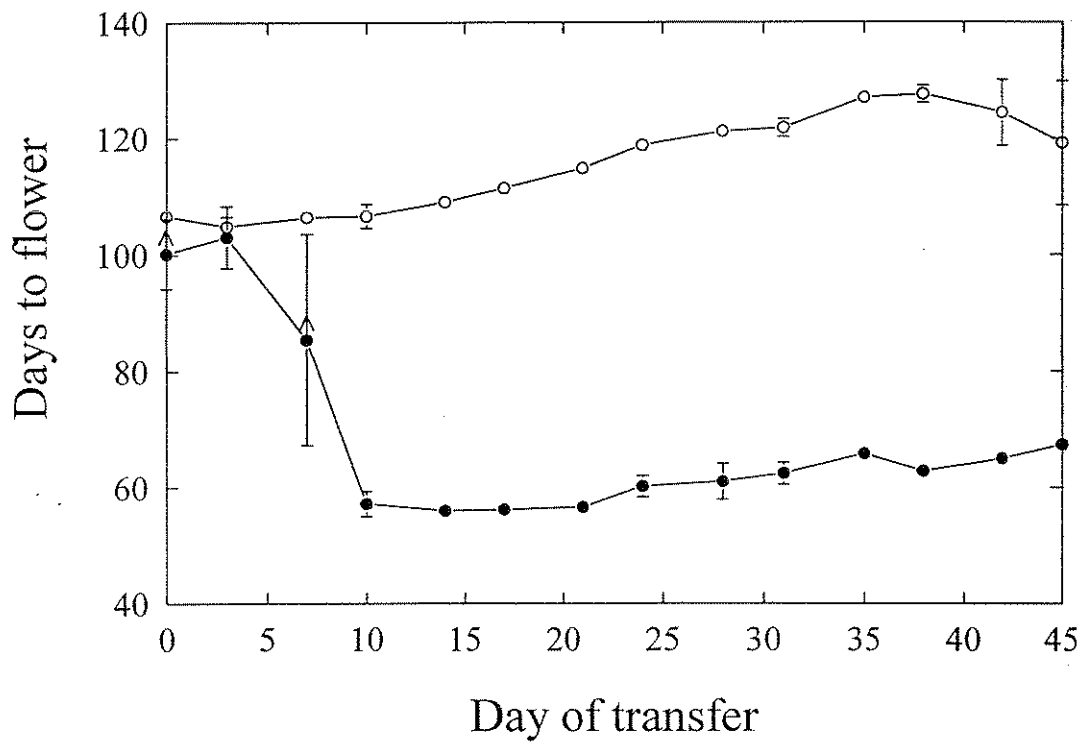
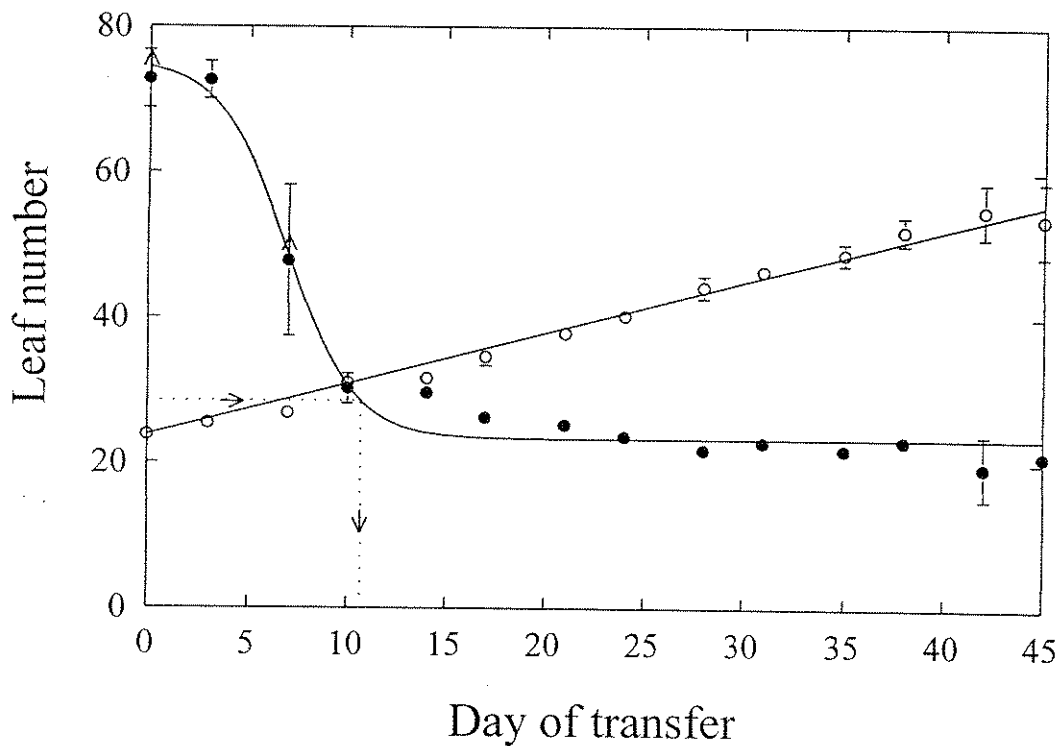


Figure 3: The effect of transferring plants from 12°C to 22°C (●) and from 22°C to 12°C (○) on the leaf number from the pinch to inflorescence (Standard errors of the means are shown where larger than the points. Arrows indicate the presence of a plant within the treatment which had not flowered after 153 days. The relationship for plants transferred from 22°C to 12°C was fitted by regression analysis; leaf number = 23.8 (± 1.21) + 0.70 (± 0.012)D, $r^2 = 0.99$, 13 d.f., where D is the transfer date. The curve was fitted to plants transferred from 12°C to 22°C using non-linear regression, where leaf number = 23.27 (± 0.89) + 52.03 (± 3.35)/(1 + e^{-(D - 6.91 (± 0.45)) - 1.72 (± 0.42)}}, $r^2 = 0.98$, 10 d.f.).



$$\text{Leaf number} = a + b/(1 + e^{-(t-c)/d})$$

Where t is the transfer date, a is the lower asymptote, b is the difference between upper and lower asymptotes and c and d are constants.

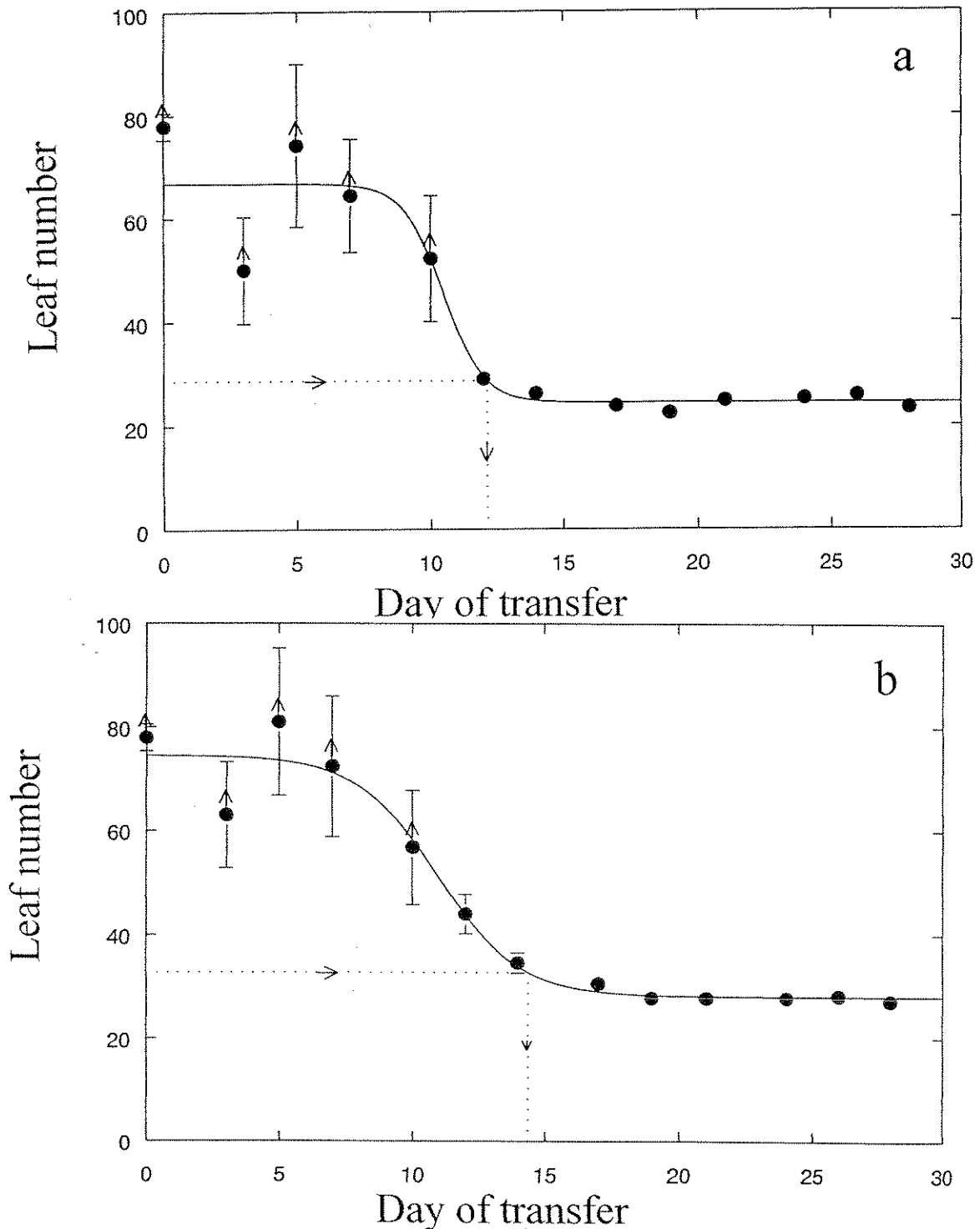
The time of inflorescence commitment was determined as the time at which the leaf number had decreased by 90% of the difference between the upper and lower asymptotes. Eleven days at 12°C were found to be sufficient to cause inflorescence commitment (Figure 3). Increased leaf number with later successive transfers in plants transferred from warm to cool conditions illustrates the delay seen in inflorescence initiation by maintaining plants initially at 22°C. An indication to the time of inflorescence commitment for plants grown at 22°C can be obtained by extrapolating this relationship to the point at which leaf number is predicted to be equivalent to that seen in plants grown continuously at 22°C (73 leaves). This suggests that inflorescence commitment occurs after 68 days at 22°C, although as the error bars (standard errors of the mean) indicate, the leaf number and therefore most probably the time of inflorescence commitment is highly variable at this temperature.

The relationship between leaf number below the inflorescence and transfer date for plants transferred from 22°C to 12°C (Figure 3) can be used to indicate the time when plants became sensitive to chilling, or whether a period of insensitivity to chilling exists immediately after pinching. The regression line from plants transferred from 22°C to 12°C intercepts the Y-axis at 24 leaves a value not significantly different from the leaf number recorded in plants grown continuously at 12°C. Warm temperatures are therefore shown to have increased the leaf number below the inflorescence even if given immediately after pinching. This suggests a delay in inflorescence initiation and therefore that no period of insensitivity to chilling after pinching exists.

Experiment 2. Effect of temperature and photoperiod on inflorescence commitment.

It was found that 11 days were required to cause inflorescence commitment in plants grown at 12°C lit for 16h d⁻¹, and so to examine more fully the effects of both temperature and photoperiod on the time to inflorescence commitment a second experiment was conducted where plants were transferred from a range of photo-thermal conditions to a warm glasshouse. Figure 4 shows data for plants grown at 10.6° and 13.7°C and exposed to a photoperiod of 14h d⁻¹, similar relationships were obtained for other plants grown under the other photoperiods (data not shown).

Figure 4: The relationship between the mean number of leaves between the pinch and inflorescence with transfer date for plants grown initially at **a)** 10.6°C and a 14h d⁻¹ photoperiod (where leaf number = $24.41 (\pm 2.81) + 42.37 (\pm 4.73)/(1 + e^{-(D-10.47(\pm 0.57)/-0.76(\pm 0.54)})$, $r^2 = 0.91$, 9d.f.) **b)** 13.7°C and a 14h d⁻¹ photoperiod (where leaf number = $28.09 (\pm 2.03) + 46.43 (\pm 3.75)/(1 + e^{-(D-10.95(\pm 0.57)/-1.56(\pm 0.54)})$, $r^2 = 0.91$, 9d.f.) before transfer to a glasshouse set to provide a minimum temperature of 22°C. The curves were fitted by non-linear regression analysis using SlideWrite plus 2.0, where D is the transfer date. Arrows indicate the presence of plants within a treatment that had not flowered after 153 days from pinching.



Sigmoidal curves were fitted by non-linear regression analysis using SlideWrite Plus 2.0 as described in experiment 1. The time of inflorescence commitment was again determined as the time at which the leaf number had decreased by 90% of the difference between the upper and lower asymptotes. A proportion of the plants that had not committed to flower before transfer to the warm glasshouse, had not initiated inflorescences by 153 days from pinching. In this instance, the mean leaf number was determined as the mean of the whole population which included plants that had flowered as leaf counts from plants not flowering after 153 days.

The time of inflorescence commitment was calculated for plants grown under each photoperiod in the two coolest temperatures (Table 2). Plants grown in the warmer temperature regimes, irrespective of photoperiod, had not committed to flower after 28 days. Analysis of variance showed a significant ($p > 0.05$) effect of temperature on the time of inflorescence commitment, but no significant effect of photoperiod. The effect of temperature on the time to inflorescence commitment was pronounced the most rapid time for this to occur was in plants grown at 10.6°C and a 2.1°C rise in temperature caused a delay of 4 days. When the temperature was raised to 18.7°C, no inflorescence commitment occurred within the 28 day period over which plants were transferred, irrespective of photoperiod.

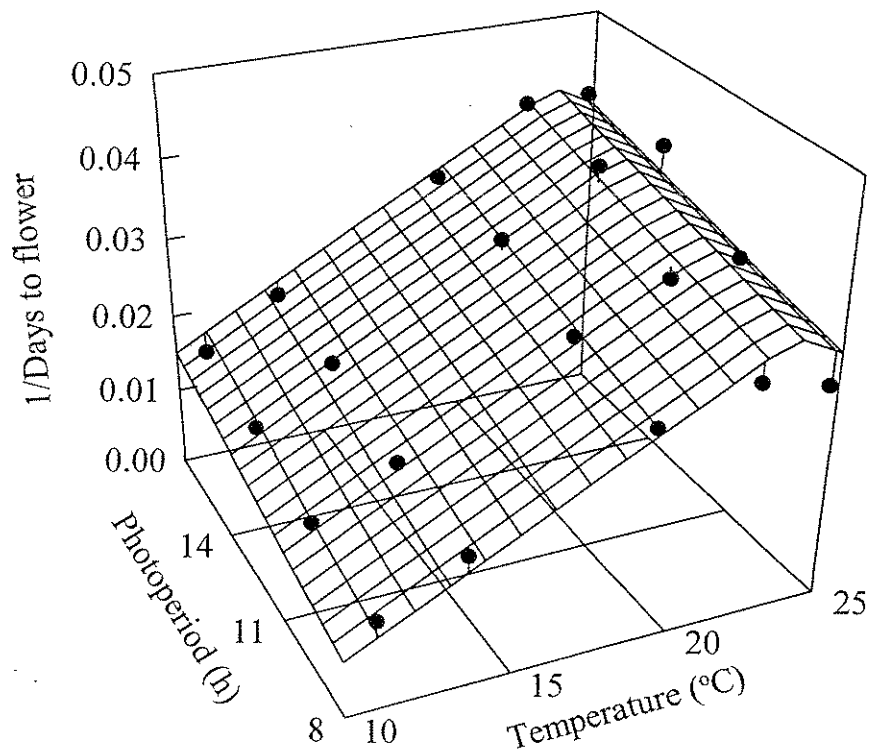
Table 2: The effect of mean temperature from pinching and photoperiod on the number of days for commitment of a terminal inflorescence, as determined by leaf number data.

Temperature (°C)	Photoperiod (hr d ⁻¹)	Days for inflorescence Commitment
10.7	8	14.1
10.6	11	10.5
10.6	14	12.1
10.7	17	14.9
13.7	8	17.9
10.7	8	14.1
13.7	11	17.4
13.7	14	14.4
13.7	17	18.3
≥18.7	8	>28
≥18.7	11	>28
≥18.7	14	>28
≥18.7	17	>28

Experiment 3. The effect of temperature and photoperiod on inflorescence development.

All plants dissected had initiated inflorescences after 35 days growth at an average temperature of 12.2°C and at natural daylengths. Plants were then transferred to the combination of 6 temperatures and 4 photoperiod treatments. Plants flowered in all treatments except in the warmest temperature regime (minimum temperature of 26°C) which senesced before flowering. The rate of progress to flowering (1/f) increased linearly with increasing temperature (Figure 5), such that plants grown under an 8h d⁻¹ photoperiod flowered 55 days earlier at 22.5°C compared to 11°C.

Figure 5: The effect of temperature and photoperiod on inflorescence development, shown as the reciprocal of the days from inflorescence initiation to anthesis. (Each point is the mean of 5 replicate plants. The mesh was fitted by regression analysis where $1/f = 0.019(\pm 0.002) + 0.0019(\pm 0.00009) Te + 0.00089(\pm 0.00013)P$ $r^2 = 0.96$, 17 d.f., where P - photoperiod and Te = effective temperature calculated with an optimum temperature (To) of 23.5°C, where $Te = To - |To - Ta|$ and Ta is the actual temperature.)



There was also some indication of an optimum temperature at 23.5°C, with a slight slowing in the rate of progress to flowering being seen in plants grown at temperatures higher than this. The rate of progress to flowering also increased linearly with longer days, such that plants grown at an average temperature of 19.3°C flowered a week earlier when the daylength was extended from 8 to 17h d⁻¹ (Figure 5) indicating that *Osteospermum* is a long day plant for this developmental stage. There were no data to indicate that plants had reached either a critical or ceiling photoperiod. Multiple linear regression showed that both temperature and photoperiod had significant independent effects of the rate of the rate of progress to flowering with no significant interaction, the analysis giving a good fit to the data ($r^2 = 0.96$, 17d.f.).

Plants grown with day extension lighting to increase the photoperiod to 17h d⁻¹ were also elongated compared to those at 8h d⁻¹. This elongation is clearly undesirable in terms of commercial plant production, but it is not clear whether this response was due to the length of the day *per se* or the tungsten light used in the photoperiod cabinet to extend the day.

DISCUSSION

Vince-Prue (1975) described the early phase of plant growth from germination during which flowering cannot be induced as juvenility. For a plant to respond to conditions inductive to flowering it must be sensitive to these stimuli, and the meristem must be capable of responding. It is usually the incapability of the meristem to respond that prevents flowering in woody plants while juvenile (Bernier, 1988). It is unlikely that cutting-raised plants are incapable of receiving inductive stimuli for flowering, however, Post (1949) identified a period of insensitivity to short days which are normally inductive to flowering in chrysanthemum, when axillary meristems could be released from apical dominance by decapitation. This would therefore, tend to suggest that meristems were incapable of responding to inductive stimuli until they reached a particular age or size. Data presented here, however, show no evidence for a period of insensitivity after pinching, to stimuli inductive to flowering (i.e. chilling) in *Osteospermum jucundum* cv. 'Zulu'. Exposing plants to 22°C after pinching was predicted to increase leaf number presumably due to delayed inflorescence initiation, indicating plants were sensitive to inductive stimuli at this time.

Inflorescence commitment and, as a result inflorescence initiation in *Osteospermum*, has been shown to be hastened by cool temperatures. However, inflorescence initiation resulted directly from the chilling treatment. Therefore, as suggested by Pearson *et. al.* (1995), the chilling treatment was not a true vernalisation response as defined by Vince-Prue (1975), where cool temperatures would act to make the plant sensitive to a second stimulus (such as long days) to induce flowering. The response of *Osteospermum* is therefore comparable to the low optimum temperature for inflorescence initiation seen in cineraria (Larsen, 1985). The optimum temperature for inflorescence commitment in *Osteospermum* has not been determined here, but has been shown to be in the order of 11°C or below. Furthermore, there is no evidence that the requirement for chilling can be modified by photoperiod which is the case in a number of long day plants requiring cold, such as *Campanula medium* (Wellensiek, 1960). The requirement for cold in *Campanula medium* can be overcome by short day; these plants will normally flower if exposed to either low temperatures followed by long days, or short days followed by long days. *Osteospermum* appears, however, to be day-neutral for this stage of development, although once plants are committed to flowering further inflorescence development can be hastened by long days and warm temperatures (up to an optimum of 23.5°C). Inflorescence commitment and development therefore respond very differently to the photo-thermal environment.

The analysis conducted here has greatly expanded our understanding of the flowering physiology in *Osteospermum jucundum*, the understanding gained has important implications for commercial growers. Growers of *Osteospermum* want plants with a short compact habit, to achieve this growth regulators are often applied (Olsen and Andersen, 1995, Fuller, 1996). These not only add to the cost of production, but are becoming increasingly unpopular and alternatives to growth regulators are continuously being sought. By carefully controlling temperature and therefore the time which plants initiate inflorescences, the number of leaves below the inflorescence can be controlled within certain limits. Control of leaf number can therefore be used to limit height, rather than trying to control internode lengths after initiation has occurred. This work has concentrated on the time to inflorescence commitment of the terminal meristem. However, in commercial production, longer periods of chilling will be required to increase bud number, as inflorescence initiation occurs in a basipetal progression (see HDC Report PC 114). The terminal inflorescences therefore initiate first, and as chilling continues, the number of flowering laterals on a branch will increase. Furthermore, there is potential to use long days to hasten flowering when plants are grown on at warmer temperatures after inflorescence commitment. Day extension or night break lighting could hasten flowering by about one week. However, detrimental internodal extension may result, as was seen under long day treatments using tungsten lights.

PART III: ANGLIA ALPINES LTD.

COMMERCIAL EVALUATION OF CHILLING TREATMENTS IN COMBINATION WITH GROWTH REGULATOR APPLICATION

INTRODUCTION

A commercial trial was set up at Anglia Alpines Ltd in order to provide information on the potential interaction between the effects of using a chemical plant growth regulator and the use of chilling treatments on flowering in *Osteospermum*. This trial was also initiated in order to confirm some of the results of the studies carried out at both HRI Efford and at the University of Reading under fully commercial growing conditions.

MATERIALS AND METHODS

The trial was run under a commercial growing environment at Anglia Alpines Ltd and a chemical plant growth regulator for controlling plant height ('Cycocel' = chlormequat) was applied to all treatments. Plants were potted into 13 cm pots and grown on for 7 days at a minimum temperature of 16°C and were then transferred to their various treatments. After completion of each of the chilling treatments, plants were returned to a temperature of 16°C which was maintained until the end of the trial. The cultivar used was the chilling sensitive cv. 'Zulu' and the treatments applied were as follows:

- | | |
|---|--------|
| 1. Control - grown at ambient temperature (16°C) throughout | White |
| 2. 9°C for 7 days - 'short' chilling treatment | Yellow |
| 3. 9°C for 14 days - 'medium' chilling treatment | Pink |
| 4. 9°C for 21 days - 'long' chilling treatment | Green |
| 5. 22°C for 7 days - 'short' warm temperature treatment | Purple |
| 6. 22°C for 14 days - 'medium' warm temperature treatment | Orange |
| 7. 22°C for 21 days - 'long' warm temperature treatment | Blue |

Cycocel (chlormequat) growth regulator was applied as a drench at a rate of 250 ml per 13 cm pot at a concentration of 2 ml l⁻¹. Applications were made to the various treatments on the dates stated below.

Crop notes

All plants potted at 16°C in 13 cmm pots.	1 March
All plants moved to different temperature zones.	6 March
Plants in treatments 2 and 5 returned to 16°C zone.	14 March
Plants in treatments 3 and 6 returned to 16°C zone.	21 March
Plants in treatments 4 and 7 returned to 16°C zone.	28 March
All plants stopped	23 March

Cycocel applications

To treatments 1 and 7.	16 April
To treatments 2, 3, 4, 5 and 6.	22 April
To treatments 2, 3 and 4.	2 May

Crop scores

Treatment 3.	1 June
Treatments 1, 2 and 4.	3 June
Treatments 5, 6 and 7	8 June

Suggested plant specification cv. 'Zulu'

Height above pot	25 to 30 cm
Width	15 to 20 cm
No. branches	5 plus
No. flowers open per plant	2 or more

RESULTS

The earliest treatment to flower was treatment 3 (14 days at 9°C) on 1 June. The 7 and 21 day treatments at 9°C prior to ambient (16°C) and the controls all had flowers open by 3 June whilst those plants which were placed at 22°C for 7, 14 and 21 days, started flowering by 8 June.

The strongest effect of the 9°C chilling treatment was in those plants which were kept chilled for 21 days (treatment 4), these were some 5 cm smaller in height than those kept at 9°C for 7 days (Table 3). The width of plants, number of branches and number of flowers open appeared to be little affected by the treatments, although there was some reduction in bud number in the 14 day chilling treatment (treatment 3). The 7 day chilling treatment (treatment 2) did increase both bud and flower number.

There was a slight reduction in plant height after the longest period of treatment at 22°C (treatment 7 = 21 days). However the warm temperature treatments seemed to have little effect on plant width, branch number and the numbers of open flowers, although there did appear to be some decrease in both bud and open flower numbers after the longer periods at 22°C (treatments 6 and 7; see Table 3). All plants were considered to be of a marketable standard of quality.

Table 3: Effect of chilling treatments on plant height, width, numbers of branches and flowers open in cv. 'Zulu'

Treatment	Date	Height (cm)	Width (cm)	No. Branches	No. Flowers Open	Bud No.	Total No. Flowers & Buds
1 Control; 16°C throughout	3 June	25.7	16.0	5.1	3.2	12.2	15.4
2 Short chilling 7 days 9°C then 16°C	3 June	28.6	18.4	5.7	3.1	13.9	17.0
3 Medium chilling 14 days 9°C then 16°C	1 June	25.1	16.6	4.4	3.2	9.9	13.1
4 Long chilling 21 days 9°C then 16°C	3 June	23.7	16.8	5.0	2.4	13.5	15.9
5 Short warming 7 days 22°C then 16°C	8 June	28.3	16.7	5.0	3.1	11.0	14.1
6 Medium warming 14 days 22°C then 16°C	8 June	28.3	16.4	4.9	2.2	12.0	14.2
7 Long warming 21 days 22°C then 16°C	8 June	26.2	15.1	4.5	2.3	8.2	10.5
Mean		26.6	16.6	4.9	2.8	11.5	14.3

On the first recording date (1 June), treatment 3 (14 days at 9°C) gave 100% flowering (Table 4), whereas no other treatment gave over 33% flowering and in treatments 6 and 7 (14 and 21 days at 22°C respectively) there were no open flowers and only a small amount of flowering in the controls and in treatment 5 (7 days at 22°C). On June 3, treatments 2 and 4 (7 and 21 days at 9°C respectively) were also giving greater than 50% flowering. On the final recording date (8 June) only treatment 6 (14 days at 22°C) still had not reached 100% flowering (Table 4).

DISCUSSION

All treatments produced good plants. However, treatment 3 (14 days 'chilling' at 9°C) gave the best results in terms of quality. This treatment also reduced the production over all other treatments by 4 - 7 days. The commercial advantage of 100% flowering at an earlier date could give an extra return per pot of some 30 - 50p, whilst allowing for production costs due to an 8% reduction time over a 92 day schedule.

Table 4: Percentage of plants of cv. 'Zulu' with one or more flowers open

Treatment	% Flowers open		
	1 June 1996	3 June 1996	8 June 1996
1 Control; 16°C throughout	4	30	100
2 Short chilling 7 days 9°C then 16°C	33	83	100
3 Medium chilling 14 days 9°C then 16°C	100	100	100
4 Long chilling 21 days 9°C then 16°C	25	80	100
5 Short warming 7 days 22°C then 16°C	2	27	100
6 Medium warming 14 days 22°C then 16°C	0	4	76
7 Long warming 21 days 22°C then 16°C	0	2	100

GENERAL CONCLUSIONS

When considered together the results of the three sections of this project on *Osteospermum* were mutually supportive and the following general points can be drawn from the study.

- There was a large variation between cultivars in the degree of response to temperature treatments, although the overall trends in response were the same as in cv. 'Zulu' which was adopted as the project 'standard' cultivar.
- 'Chilling' treatments applied to young plants immediately after pinching could increase flower initiation, reduce the time to flower opening and therefore times to marketing as well as reducing leaf numbers and therefore the canopy height by reducing the number of stem internodes.
- The chilling temperature strongly influenced the length of time required for inflorescence commitment. In plants at 10.6°C, commitment occurred 4 days before those in 13.7°C. Although 11 days at 12°C was sufficient for commitment, after 7 days at 9°C, the process was still apparently reversible by transferring plants to a short duration at 22 °C.
- After long periods of chilling, inflorescence commitment did not appear to be reversible by the implementation of short periods of warm temperature.
- The effect of chilling on inflorescence commitment was considered to be a low optimum temperature mediated process as opposed to a true vernalisation effect.
- Photoperiod did not appear to affect the time to inflorescence commitment.
- At 22°C inflorescence commitment only happened after 68 days and plants grown under these conditions remained largely vegetative with few flowers reaching maturity.
- Once inflorescence commitment was completed, subsequent development could be hastened by warmer temperatures, such as 22°C, although the impacts of this on commercial quality of plants was not assessed. In general, flower development, post-inflorescence commitment, appears to be favoured by warm temperatures and long days.
- In commercial trials, chilling treatments appeared quite compatible with other practices such as growth regulator applications, and a treatment of 14 days at 9°C post-pinching gave promising results.

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APPENDIX I

Trial layout for HRI Efford experiment Part A

Compartment Q1 - treatment position
Temperature 9°C until * then at 16°C

Day No.											Total No. Plots
8		2	5	6	7						96
9		2	5	6	7						96
10		2	5	6	7						96
11		2	5	6	7						96
12		2	5	6	7						96
13		2	5	6	7						96
14		2	5	6	7						96
*15	1	2							14		66
16	1	2							14		66
17	1	2							14		66
18	1	2			5				14		90
19	1	2			5				14		90
20	1	2			5				14		90
21	1	2			5	6			14		114
22	1	2	3		5	6			14	15	156
23	1	2	3		5	6			14	15	156
24	1	2	3		5	6	7		14	15	180
25	1	2	3		5	6	7	8	14	15	204
26	1	2	3		5	6	7	8	14	15	204
27	1	2	3		5	6	7	8	14	15	204
28	1	2	3		5	6	7	8	14	15	228
29	1	2	3	4	5	6	7	8			180
30	1	2	3	4	5	6	7	8			180
31	1	2	3	4	5	6	7	8			180
32	1	2	3	4	5	6	7	8			180
33	1	2	3	4	5	6	7	8			180
34	1	2	3	4	5	6	7	8			180
35	1	2	3	4	5	6	7	8			180
36	1	2	3	4	5	6	7	8			180
37	1	2	3	4	5	6	7	8			180
38	1	2	3	4	5	6	7	8			180
39	1	2	3	4	5	6	7	8			180
40	1	2	3	4	5	6	7	8			180
41	1	2	3	4	5	6	7	8			180
42	1	2	3	4	5	6	7	8			180
43	1	2	3	4	5	6	7	8			180
44	1	2	3	4	5	6	7	8			180
45	1	2	3	4	5	6	7	8			180
46	1	2	3	4	5	6	7	8			180

Space plants to 30/m²

Treatment 4 - cultivars A, C, E and H only

APPENDIX I

**Trial layout for HRI Efford experiment
Part B**

Compartment Q2 - treatment position
Temperature 9°C until * when it reverts to 16°C

Plot No.													Total No. Plots	
8		3	4				8	9	10	11	12	13		168
9		3	4				8	9	10	11	12	13		168
10		3	4				8	9	10	11	12	13		168
11		3	4				8	9	10	11	12	13		168
12		3	4				8	9	10	11	12	13		168
13		3	4				8	9	10	11	12	13		168
14		3	4				8	9	10	11	12	13		168
15		3	4				8	9	10	11	12	13		168
16		3	4				8	9	10	11	12	13		168
17		3	4				8	9	10	11	12	13		168
18		3	4				8	9	10	11	12	13		168
19		3	4				8	9	10	11	12	13		168
20		3	4				8	9	10	11	12	13		168
21		3	4				8	9	10	11	12	13		168
22			4							11	12	13		96
23			4							11	12	13		96
24			4							11	12	13		96
25			4							11	12	13		96
26			4							11	12	13		96
27			4							11	12	13		96
28			4							11	12	13		96
*29	14	15	4					9					16	90
30	14	15	4					9					16	90 Space plants to 30/n
31	14	15	4					9	10				16	114
32	14	15	4					9	10	11			16	138
33	14	15	4					9	10	11			16	138
34	14	15	4					9	10	11			16	138
35	14	15	4					9	10	11			16	162
36	14	15	4					9	10	11			16	162
37	14	15	4					9	10	11			16	162
38	14	15	4					9	10	11			16	186
39	14	15	4					9	10	11			16	186
40	14	15	4					9	10	11			16	186
41	14	15	4					9	10	11			16	186
42	14	15	4					9	10	11			16	186
43	14	15	4					9	10	11			16	186
44	14	15	4					9	10	11			16	186
45	14	15	4					9	10	11			16	186
46	14	15	4					9	10	11			16	186

Treatment 4 (italics) - cultivars B, D, G and J only

APPENDIX I

**Trial layout for HRI Efford experiment
Part C**

Compartment Q3 - treatment position
Temperature 22°C throughout

Day No.														Total No. of plots			
8	14	15	16														54
9	14	15	16														54
10	14	15	16														54
11	14	15	16														54
12	14	15	16														54
13	14	15	16														54
14	14	15	16														54
15		15	16	5	6	7											108
16		15	16	5	6	7											108
17		15	16	5	6	7											108
18		15	16		6	7											84
19		15	16		6	7											84
20		15	16		6	7											84
21		15	16			7											60
22			16			7	8	9	10								114
23			16			7	8	9	10								114
24			16				8	9	10								90
25			16					9	10								66
26			16					9	10								66
27			16					9	10								66
28			16						10								42
29									10	11	12	13					96
30									10	11	12	13					86
31										11	12	13					72
32											12	13					48
33												12	13				48
34													12	13			48
35														13			24
36															13		24
37																13	24
38																	
39																	
40																	
41																	
42																	
43																	
44																	
45																	
46																	

Space plants to 30/m²

APPENDIX II

Crop Diary

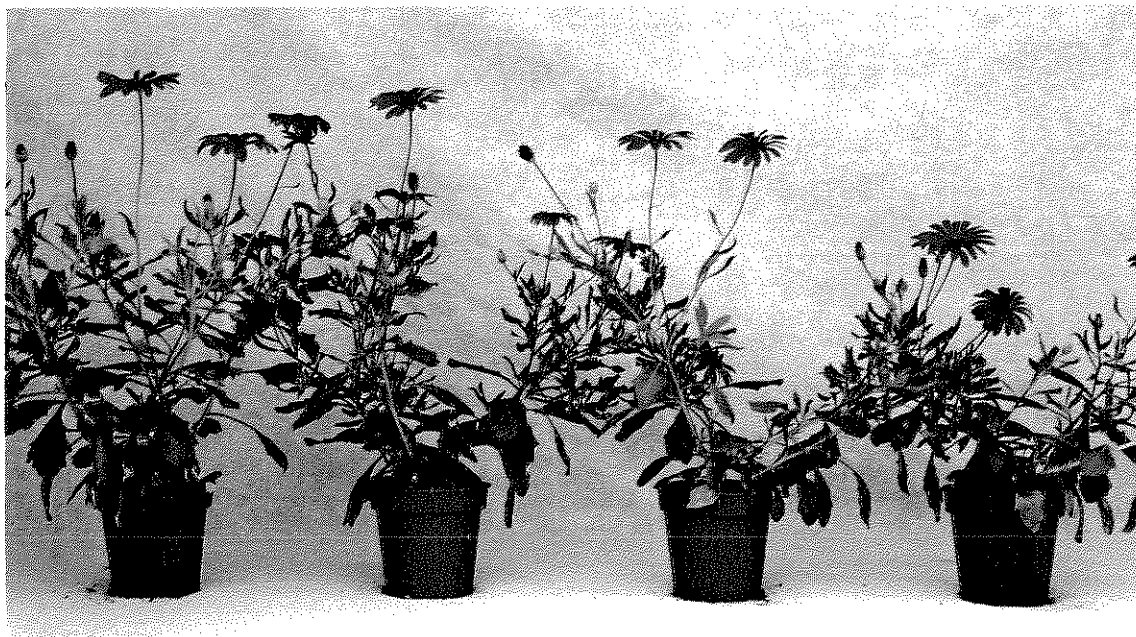
Date Detail

30.01.96	Cultivars 'Sunny Gustav', 'Sunny Lady', and 'Lubutu' struck
31.01.96	Cultivars 'Zulu', 'Lindi', 'Lusaka' and 'Pink Fantasy' struck. All cuttings treated with Seradix 1 rooting hormone and subsequently drenched with Benlate (0.5g per litre). Ambient temperature 16°C, base temp 21°C, assimilation lighting 16 hours, 04.00 to 20.00 hours, 2000 lux.
06.02.96	Rovral spray applied.
13.02.96	Rovral spray applied.
22.02.96	Assimilation lighting stopped
27.02.96	Pinched to 4th leaf node cvs. 'Sunny Landy', 'Sunny Gustav'.
28.02.96	Pinched to 4th leaf node cvs. 'Sunny Girl', 'Lubutu' and 'Pink Fantasy'.
29.02.96	Pinched to 4th leaf node cvs. 'Lusaka', 'Zulu' and 'Lindi'.
05.03.96	Potted trts. 5, 6, 7, 1 & 2.
06.03.96	Potted trts. 3, 4, 8, 9, 10, 11, 12 and 13.
07.03.96	Potted trts. 14, 15 and 16. Start date for temperature treatments.
13.03.96	Fongarid drench applied (5g per m ²).
14.03.96	Dissection sampling of Control treatment 1, each cultivar. Treatment 1 transferred to H south, 16°Cn for 1 week. Q1 and Q2 temperature dropped to achieve average 9°C. Q3 temperature raised to 22°C.
21.03.96	Biological control agent introduced, <i>Amblyseius eucumeris</i> , <i>Phytoseilus persimilis</i> and <i>Aphidius colemunis</i> . Transfers.
22.03.96	Transfers.
24.03.96	Transfers
26.03.96	Nemasys applied Started feed regime
31.03.96	Transfers
03.04.96	Transfers
05.04.96	Transfers
06.04.96	Transfers
07.04.96	Transfers
10.04.96	Transfers
12.04.96	Nemolt spray (0.5ml per litre)
13.04.96	Transfers
15.04.96	Q3 temperature reduced to 16°C.
15.04.96	Q2 spaced 30 plants per m ² .
16.04.96	Q1 spaced 30 plants per m ² .
19.04.96	Nemolt spray (0.5ml per litre). Repeat biological agent introduction.

APPENDIX III Figure 1

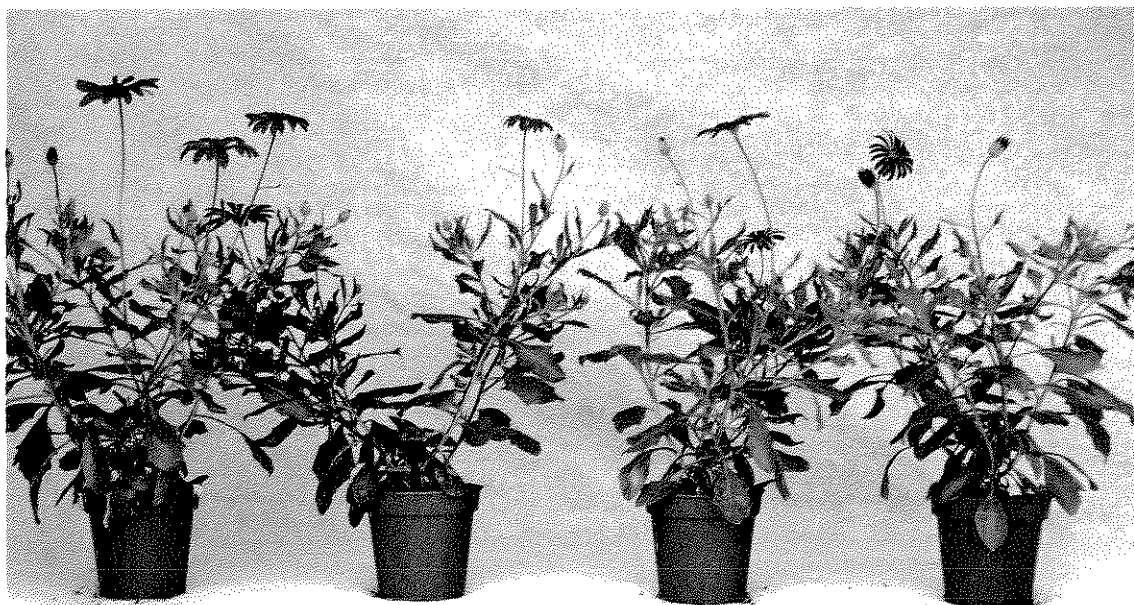
Colour plates showing treatment comparison at marketing stage in cv. 'Sunny Lady'

1a



Treatment	1	2	3	4
	(Control)	(7 days 9°C)	(14 days 9°C)	(21 days 9°C)

1b

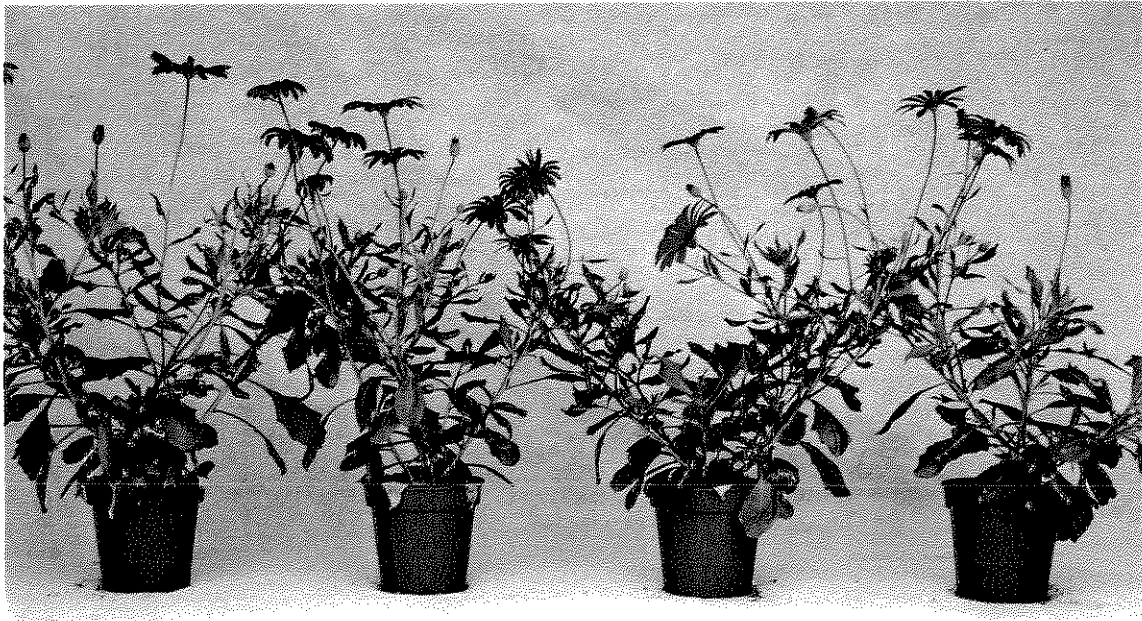


Treatment	1	5	6	7
	(Control)	(7 days 9°C + 3 days 22°C)	(7 days 9°C + 6 days 22°C)	(7 days 9°C + 9 days 22°C)

APPENDIX III Figure 1

Colour plates showing treatment comparison at marketing stage in cv. 'Sunny Lady'

1c



Treatment	1	8	9	10
	(Control)	(14 days 9°C + 3 days 22°C)	(14 days 9°C + 6 days 22°C)	(14 days 9°C + 9 days 22°C)

1d



Treatment	1	11	12	13
	(Control)	(21 days 9°C + 3 days 22°C)	(21 days 9°C + 6 days 22°C)	(21 days 9°C + 9 days 22°C)

APPENDIX III Figure 1

Colour plates showing treatment comparison at marketing stage in cv. 'Sunny Lady'

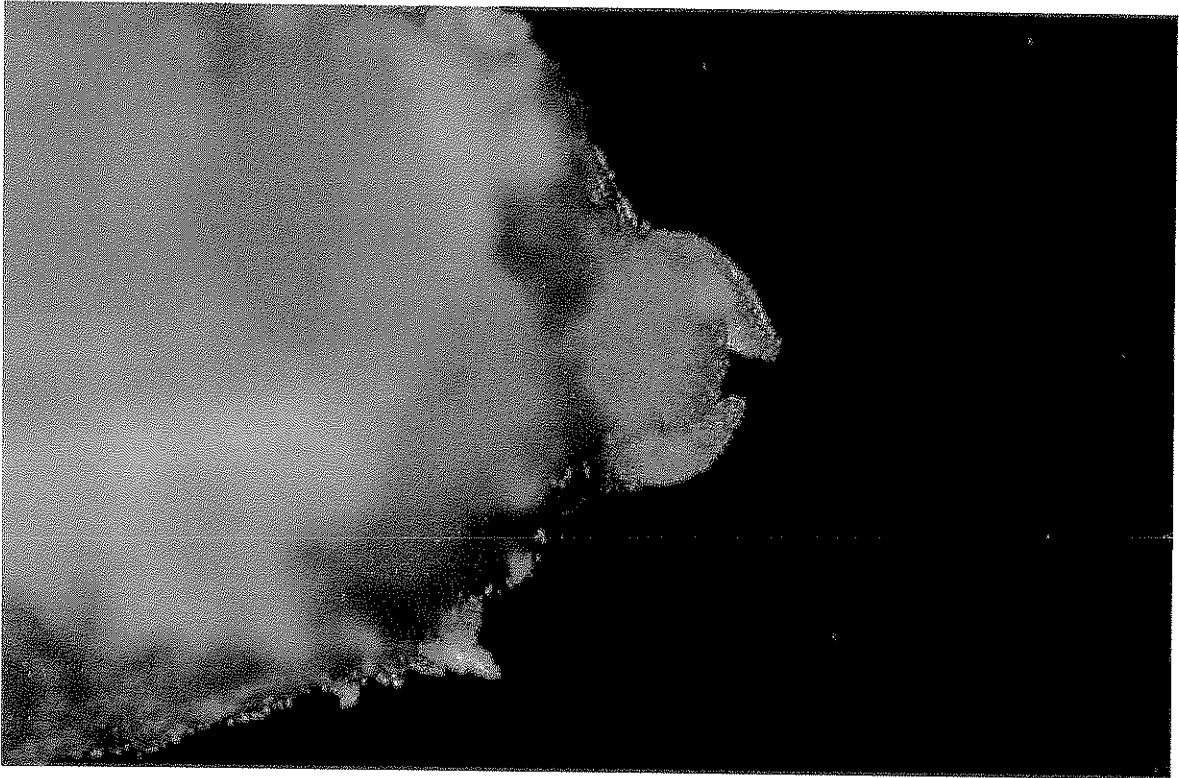
le



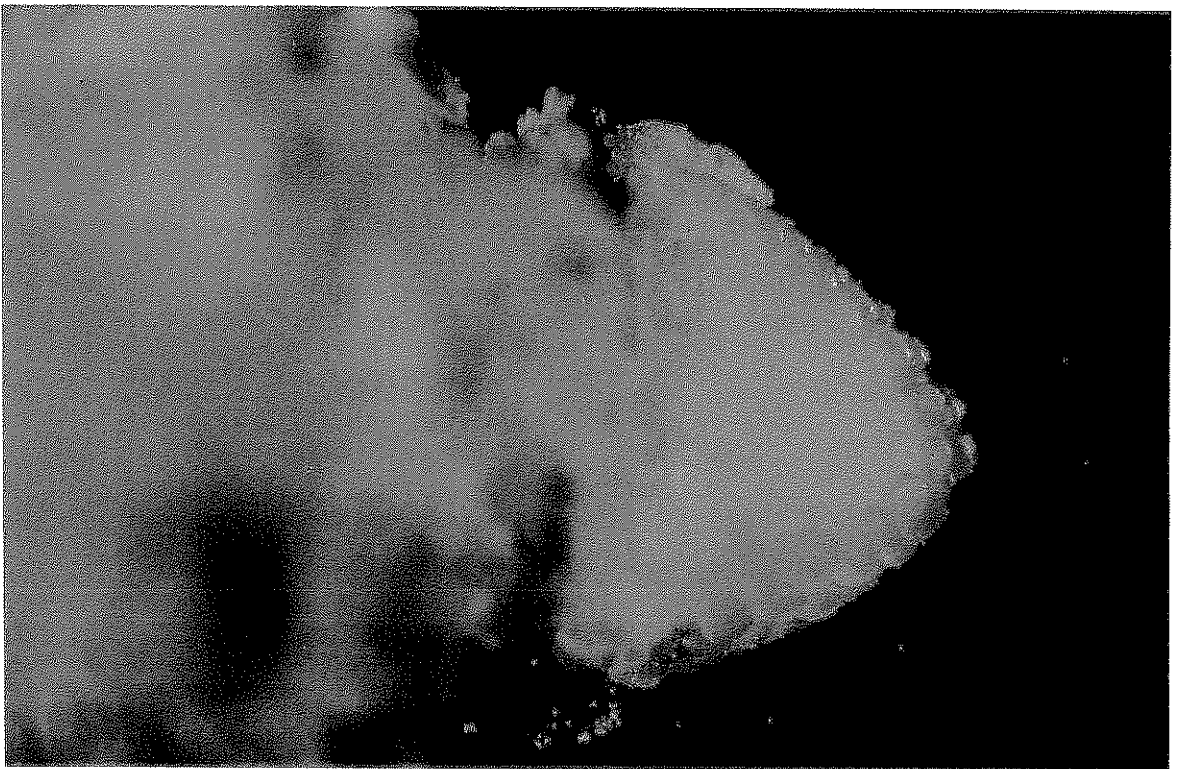
Treatment	1	14	15	16
	(Control)	(3 days 22°C)	(6 days 22°C)	(9 days 22°C)

APPENDIX III Figure 2

Low power light microscope photographs of dissected apices of *Osteospermum* cultivar 'Zulu'.



Uninitiated shoot tip



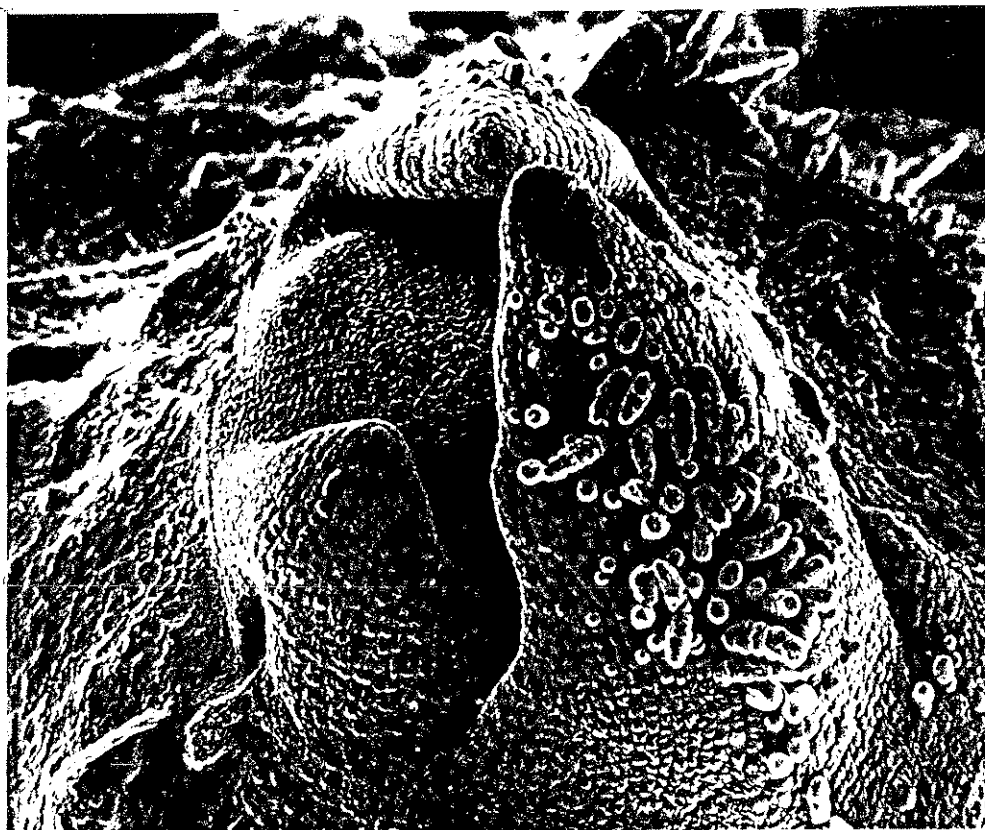
Initiated shoot tip

APPENDIX III Figure 3

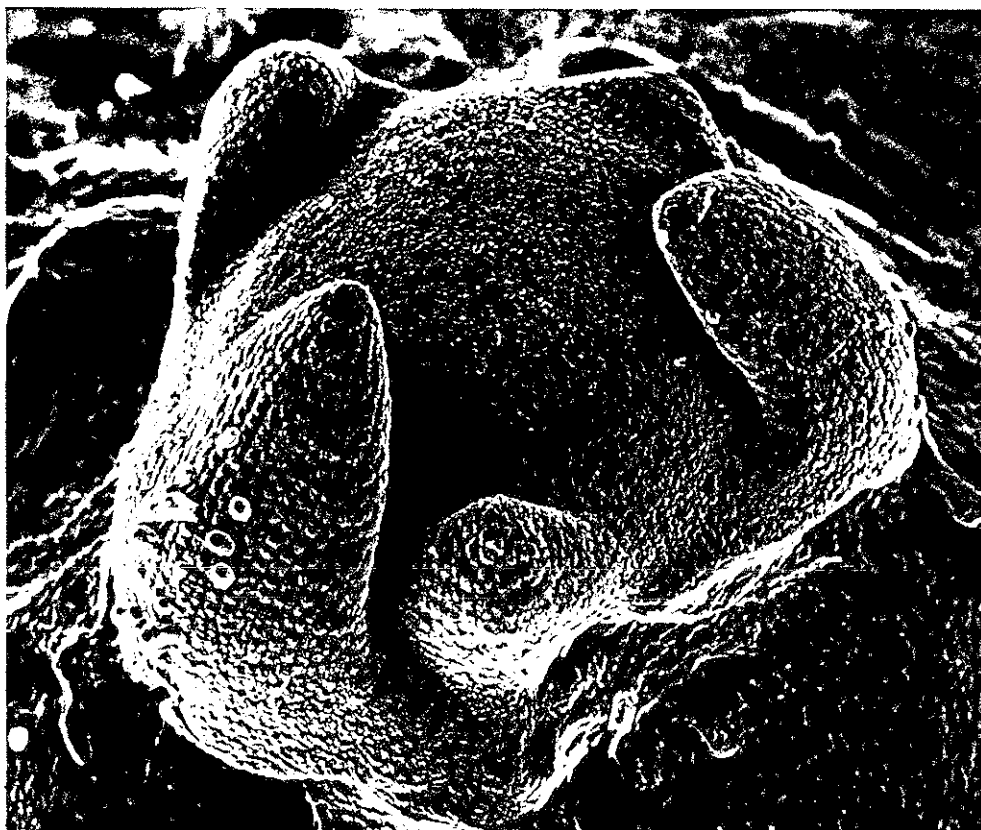
Stages of the development of the *Osteospermum* inflorescence: **a)** Stage 0; vegetative apex, a domed meristem surrounded by leaf primordia (x 250). **b)** Stage 1; enlargement of apex forming the receptacle (x 250). **c)** Stage 2; reproductive apical meristem, a whorl of bract primordia are forming (x 170). **d)** Stage 3; the receptacle is spherical and surrounded by many bract primordia (x 125). **e)** Stage 4; the lower flanks of the receptacle are covered with floret primordia, bracts are enlarged and have an entire margin (x 125). **f)** Stage 5; the entire receptacle is covered with floret primordia, a few primordia still lack the initial stages of the perianth (x 85). **g)** Stage 6; perianth present on all florets, the receptacle is dominated by disc florets surrounded by a single outer whorl of ligulate ray florets (x 60). **h)** Detailed view of disc florets, five stamen primordia formed within the perianth which has developed to form a corolla on which five petal primordia are formed (x 250).

APPENDIX III Figure 3

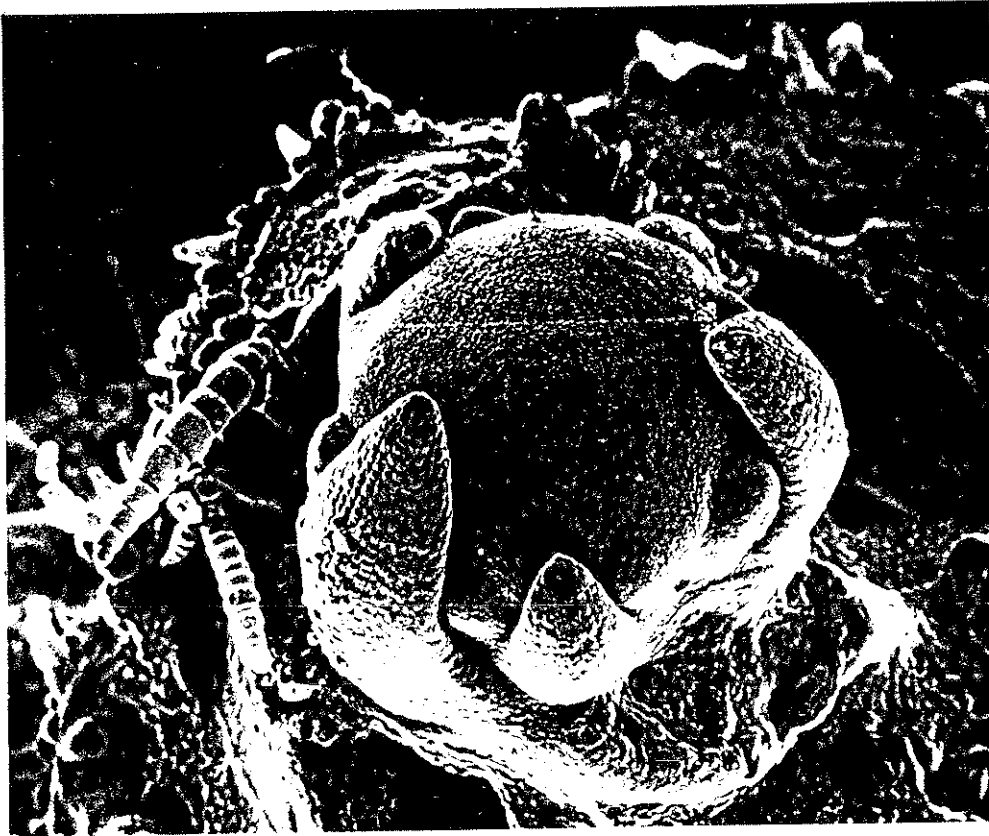
a



b



c



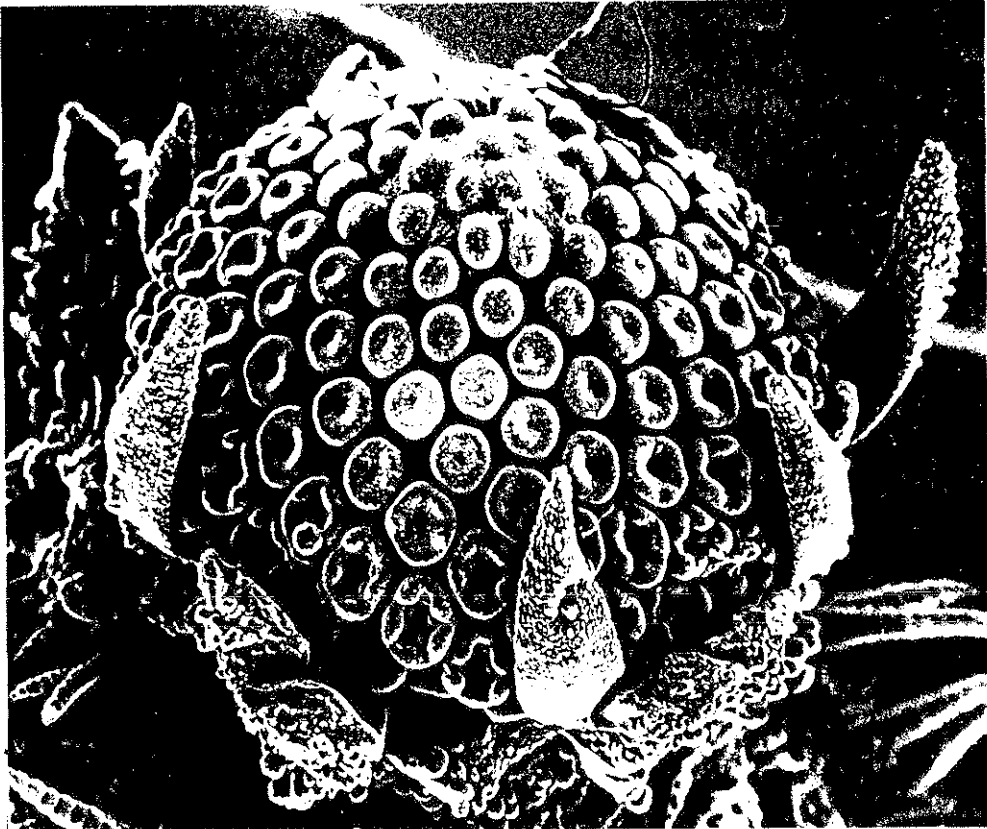
d



e

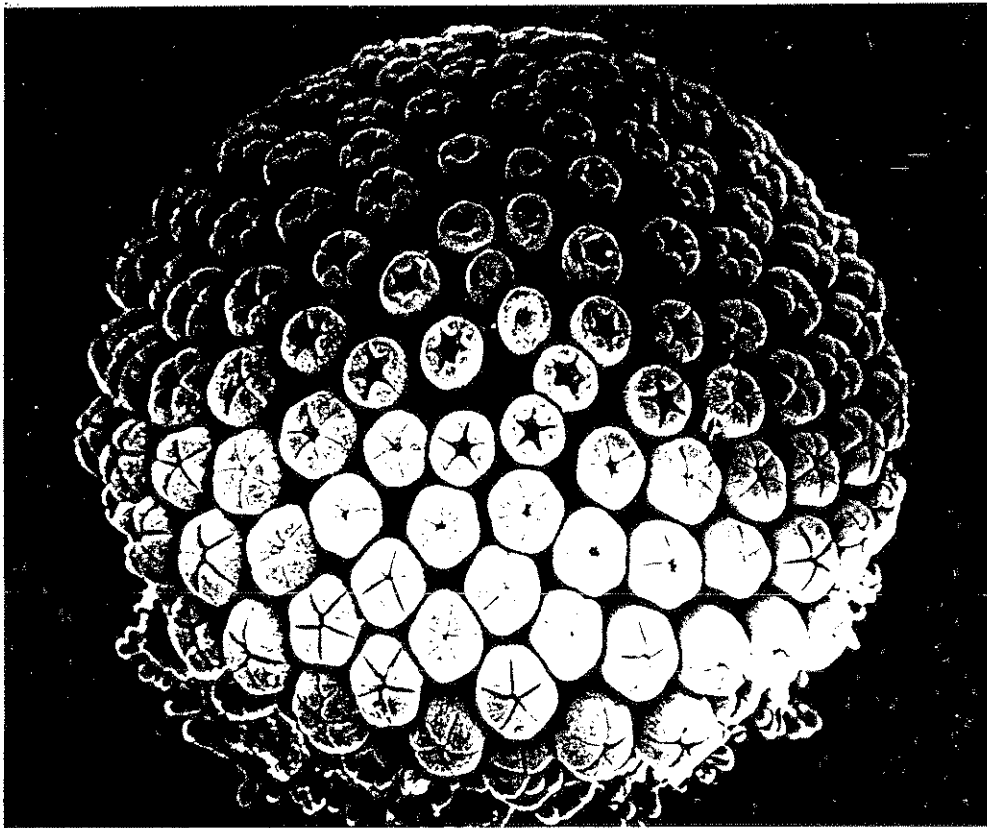


f

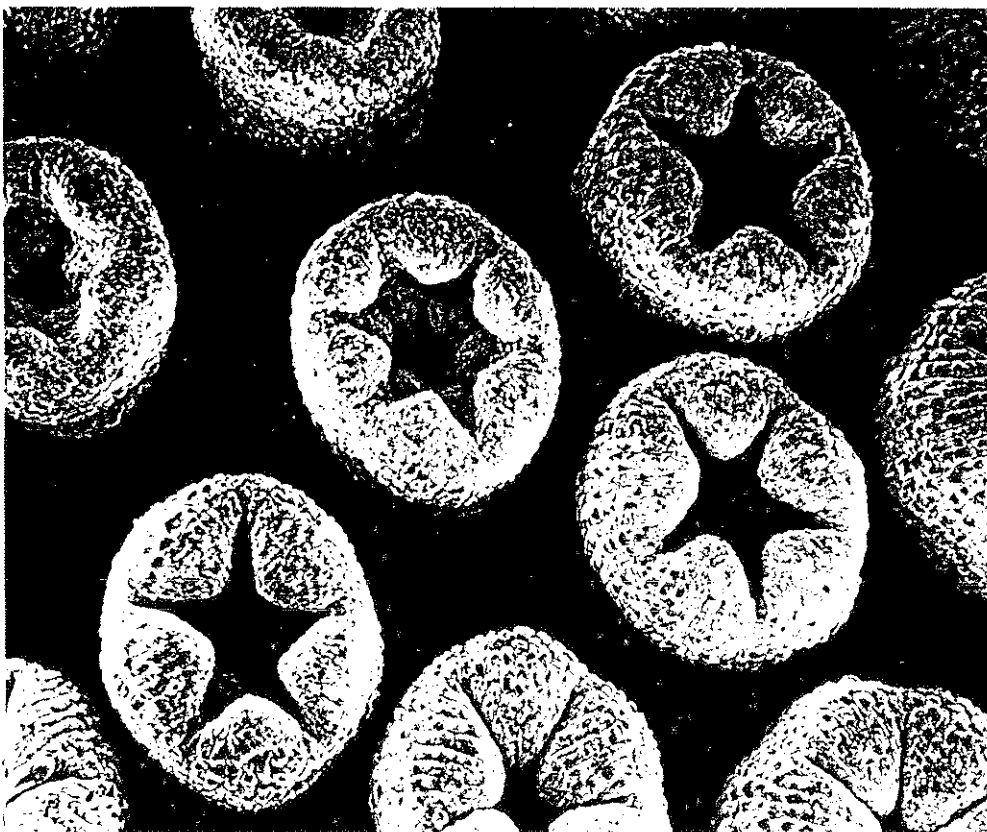


APPENDIX III Figure 3

g



h



APPENDIX IV

Compost and leaf tissue analyses

Compost

Sample type		At potting 01.03.96	Mid crop 10.04.96	At marketing 05.06.96 treatment 1	"Zulu"
Bulk density	g/ml	0.373	0.454	0.457	0511
pH		5.9	5.8	6.7	7.1
Conductivity	uS/20°C	289	299	275	250
Nitrate (as N)	mg/l	90	112	0	0
Ammonium (as N)	mg/l	41.3	28.7	0	0
Potassium	mg/l	114	113	180	186
Calcium	mg/l	81	107	71	60
Magnesium	mg/l	62	88	39	28
Phosphorus	mg/l	72	75	38	33
Iron	mg/l	0.45	0.43	1.59	1.18
Zinc	mg/l	0.28	0.16	0.65	0.87
Manganese	mg/l	0.17	0.17	0.08	0.23
Copper	mg/l	0.07	0.05	0.16	0.17
Boron	mg/l	0.20	0.00	0.09	0.01
Sodium	mg/l	36	42	42	42

APPENDIX IV

Compost and leaf tissue analyses

Liquid feed

Sample type		08.05.96	03.06.96
pH		7.0	6.9
Conductivity	uS/20°C	1522	1696
Nitrate (asN)	mg/l	124	133
Ammonium (asN)	mg/l	10.3	9.4
Potassium	mg/l	198	202
Calcium	mg/l	158	154
Magnesium	mg/l	27	30
Phosphorus	mg/l	24	25
Iron	mg/l	0.73	1.65
Zinc	mg/l	0.07	0.08
Manganese	mg/l	0.03	0.02
Copper	mg/l	0.01	0.01
Boron	mg/l	0.02	0.02
Sodium	mg/l	22	16
Chloride	mg/l	40	29
Sulphate (as S)	mg/l	21	17

APPENDIX IV

Compost and leaf tissue analyses

Leaf 01.03.96

Sample type		'Lubutu'	'Zulu'	'Pink Fantasy'	'Sunny Lady'	'Sunny Girl'	'Sunny Gustav'	'Lindi'	'Lusaka'
Organic									
Dry Matter	%	10.15	10.45	9.80	9.91	9.52	9.49	8.04	9.67
Nitrogen	%	3.30	4.02	3.48	3.61	3.68	3.71	3.93	3.71
Phosphorus	%	0.581	0.566	1.207	0.898	0.634	0.698	1.137	0.840
Potassium	%	3.89	4.06	3.06	3.26	3.35	3.72	3.43	3.96
Calcium	%	1.057	1.169	1.605	1.772	1.894	1.932	1.928	1.597
Magnesium	%	0.863	0.797	0.997	1.130	1.173	1.238	1.053	1.048
Sodium	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Iron	mg/Kg	115.86	103.33	92.44	66.07	98.53	59.45	87.02	77.01
Manganese	mg/Kg	198.79	137.93	155.59	134.81	181.21	150.45	178.81	121.44
Copper	mg/Kg	16.16	8.53	18.72	12.55	15.72	8.33	17.55	14.88
Boron	mg/Kg	34.17	23.35	33.75	65.18	74.27	66.42	35.80	35.99
Zinc	mg/Kg	83.54	68.88	83.23	65.18	74.27	66.42	65.80	69.26

Leaf 10.04.96

Sample type		'Lubutu'	'Zulu'	'Pink Fantasy'	'Sunny Lady'	'Sunny Girl'	'Sunny Gustav'	'Lindi'	'Lusaka'
Organic									
Dry Matter	%	9.37	12.02	9.05	9.27	8.12	7.88	9.35	8.81
Nitrogen	%	5.59	4.94	5.56	5.28	6.00	5.76	5.21	5.67
Phosphorus	%	0.581	0.566	1.207	0.898	0.634	0.698	1.137	0.840
Potassium	%	3.89	4.06	3.06	3.26	3.35	3.72	3.43	3.96
Calcium	%	1.057	1.169	1.605	1.772	1.894	1.932	1.928	1.597
Magnesium	%	0.863	0.797	0.997	1.130	1.173	1.238	1.053	1.048
Sodium	%	0.0	0.0	0.0	0.0	0.0	0.0	0.0	0.0
Iron	mg/Kg	115.86	103.33	92.44	66.07	98.53	59.45	87.02	77.01
Manganese	mg/Kg	198.79	137.93	155.59	134.81	181.21	150.45	178.81	121.44
Copper	mg/Kg	16.16	8.53	18.72	12.55	15.72	8.33	17.55	14.88
Boron	mg/Kg	34.17	23.35	33.75	65.18	74.27	66.42	38.50	35.99
Zinc	mg/Kg	83.54	68.88	83.23	65.18	74.27	66.42	65.80	69.26

Leaf 05.06.96

Sample type		'Lubutu'	'Zulu'	'Pink Fantasy'	'Sunny Lady'	'Sunny Girl'	'Sunny Gustav'	'Lindi'	'Lusaka'
Organic									
Dry Matter	%	20.09	21.99	19.16	19.39	18.61	17.96	16.33	21.54
Nitrogen	%	2.48	2.56	3.18	2.54	2.90	2.81	3.23	3.42
Phosphorus	%	0.345	0.341	0.617	0.575	0.505	0.535	0.910	0.404
Potassium	%	3.94	4.35	4.78	4.38	4.60	4.35	5.30	4.34
Calcium	%	0.693	0.720	1.233	1.494	1.576	1.941	1.541	1.542
Magnesium	%	0.433	0.432	0.550	0.658	0.690	0.879	0.650	0.667
Sodium	%	0.179	0.182	0.279	0.244	0.249	0.213	0.296	0.295
Iron	mg/Kg	42.23	46.35	51.72	89.52	71.76	69.27	67.70	49.00
Manganese	mg/Kg	75.26	94.77	104.10	114.87	62.44	102.02	107.16	80.30
Copper	mg/Kg	5.03	5.80	8.17	11.78	8.78	8.56	7.31	7.91
Boron	mg/Kg	11.29	12.80	18.78	16.78	12.54	15.80	22.11	14.63
Zinc	mg/Kg	21.44	27.27	32.39	27.26	17.56	24.61	28.58	24.58

APPENDIX V

Table a: Plant growth records at marketing for *Osteospermum* cultivar 'Lubutu' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	137.22	141.08	287.80	519.70	2.667	32.22	8.17	9.22	75.50
2	135.06	139.17	253.30	480.30	3.000	28.78	6.61	11.11	74.00
3	132.12	138.02	229.40	452.20	2.778	24.67	7.56	6.72	75.92
4	130.44	135.50	185.30	389.70	2.633	19.78	7.56	7.06	74.11
5	137.78	140.83	276.70	506.40	2.944	31.06	8.78	9.89	76.08
6	138.07	142.00	315.60	548.30	2.611	32.28	10.00	6.89	79.39
7	140.09	141.83	313.30	553.10	2.833	33.50	8.78	8.00	79.67
8	135.22	138.78	248.90	473.60	2.944	27.06	7.72	8.78	75.61
9	134.33	137.86	246.40	473.90	2.722	26.22	6.44	8.00	78.31
10	132.72	137.83	240.00	463.90	2.478	26.50	9.89	7.17	77.97
11	129.50	136.09	183.30	381.90	2.667	19.28	8.28	6.11	74.64
12	128.78	134.06	179.20	374.40	2.778	18.83	6.78	8.06	70.47
13	129.28	134.80	174.20	378.90	3.056	21.11	9.28	6.39	71.44
14	141.89	138.30†	354.20	600.00	2.556	34.78	16.00	3.89	83.47
15	141.75†	138.30†	400.30	629.20	2.444	40.78	14.22	1.06	78.72
16	141.75†	138.30†	401.10	638.60	2.333	41.28	11.44	1.28	84.88
<i>SED</i>	1.364	1.053	6.330	15.850	0.181	1.652	1.010	1.287	2.917
LSD; P=5%	2.804	2.173	33.345	32.360	0.370	3.373	2.062	2.628	5.956

* days from sticking (31/12/95)

† estimated values

APPENDIX V

Table b: Plant growth records at marketing for *Osteospermum* cultivar 'Lindi' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	131.37	133.89	190.80	357.20	2.889	25.06	8.39	8.83	95.24
2	129.94	132.51	155.00	319.40	3.333	19.94	7.06	10.56	95.19
3	129.44	132.33	138.60	303.60	3.778	18.11	6.78	10.39	93.42
4	129.11	131.94	138.90	301.70	4.056	16.56	3.61	9.83	92.14
5	131.94	135.58	199.20	368.60	3.500	22.50	10.22	10.11	95.61
6	131.40	134.91	202.80	371.40	3.333	23.00	9.00	9.39	95.92
7	137.07	139.67	215.80	379.20	3.278	27.61	10.56	5.72	95.40
8	132.22	132.78	183.90	342.80	3.389	21.44	8.44	6.83	95.25
9	131.42	135.39	168.60	339.20	3.389	21.06	8.39	4.28	89.83
10	127.44	131.92	147.20	308.90	3.222	20.11	9.11	3.94	91.14
11	129.01	131.62	140.30	311.90	3.500	17.22	5.17	7.78	92.87
12	128.11	130.61	110.30	267.80	3.667	16.61	2.11	9.11	88.61
13	128.40	131.34	123.90	267.60	3.467	18.01	5.96	5.53	88.92
<i>SED</i>	1.528	1.116	18.000	18.530	0.245	1.498	1.379	1.171	2.342
LSD; P=5%	3.153	2.303	37.152	38.2454	0.506	3.091	2.846	2.416	4.833

* days from sticking (31/12/95)

APPENDIX V

Table c: Plant growth records at marketing for *Osteospermum* cultivar 'Sunny Lady' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	127.50	129.78	145.30	409.20	4.222	26.67	15.00	9.28	78.33
2	126.56	128.39	117.20	366.70	4.500	16.06	12.50	16.67	78.03
3	127.50	128.89	126.90	401.10	4.222	16.11	10.39	13.06	79.14
4	128.17	130.11	102.20	341.40	4.056	15.67	9.89	12.11	78.31
5	127.89	130.17	131.40	403.30	4.222	17.56	15.50	12.61	80.06
6	128.72	130.67	136.40	400.00	4.222	17.78	14.72	9.83	79.97
7	128.33	131.78	121.10	384.40	4.000	17.72	13.89	8.33	81.72
8	127.83	128.94	172.80	386.90	4.222	15.67	10.44	13.11	77.03
9	124.89	128.17	124.40	374.40	4.111	15.44	9.17	13.61	73.03
10	123.89	128.06	92.50	362.20	4.222	15.50	6.00	13.00	75.81
11	128.06	129.72	106.10	376.90	4.222	15.61	11.28	12.22	76.81
12	128.00	129.00	106.90	365.60	4.278	15.89	6.67	14.22	76.53
13	128.06	128.61	101.10	352.80	4.333	15.72	6.78	13.17	75.06
14	133.59	137.72	158.10	404.40	3.833	21.83	8.83	5.33	80.50
15	136.17	141.56	177.80	423.10	3.667	25.78	10.89	5.17	80.67
16	139.29	139.65	212.80	466.40	3.444	28.50	9.67	3.11	79.06
<i>SED</i>	0.871	0.598	22.090	12.920	0.256	3.160	1.568	1.439	1.740
LSD; P=5%	1.778	1.224	45.107	26.382	0.521	6.452	3.201	2.938	3.553

* days from sticking (31/12/95)

APPENDIX V

Table d: Plant growth records at marketing for *Osteospermum* cultivar 'Pink Fantasy' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	125.67	130.33	143.90	356.70	3.500	20.06	9.61	12.28	73.42
2	124.89	129.44	134.20	336.70	3.889	18.11	6.56	12.78	69.61
3	125.94	129.33	124.20	312.20	3.556	16.94	5.44	12.39	70.28
4	128.00	129.17	129.40	295.00	3.389	16.61	3.94	12.17	68.58
5	125.22	131.17	145.60	351.40	3.722	19.44	8.44	10.39	71.94
6	128.50	132.67	152.20	364.40	3.556	20.67	9.39	7.89	72.72
7	128.50	132.53	150.30	375.60	3.444	22.33	8.11	6.61	73.78
8	124.89	129.83	126.40	313.10	3.222	18.61	6.33	10.22	69.47
9	125.06	130.00	112.20	293.90	3.222	18.28	4.78	7.50	69.06
10	125.94	129.96	104.20	300.30	3.733	17.44	5.67	6.50	67.03
11	127.56	129.78	125.30	278.90	3.222	18.11	3.61	9.94	66.83
12	125.17	128.17	128.60	286.70	3.667	17.61	2.06	12.83	67.33
13	124.17	128.39	116.30	274.40	3.500	16.98	2.30	11.53	65.31
14	130.61	134.56	142.20	351.90	3.167	21.33	7.56	6.50	73.00
15	134.56	138.40	166.70	378.30	3.056	24.56	6.33	3.44	73.53
16	137.22	140.61	182.60	381.40	2.889	24.83	7.06	2.11	70.17
<i>SED</i>	1.036	1.258	11.960	14.490	0.300	1.067	1.005	1.796	1.529
LSD; P=5%	2.115	2.568	24.422	29.588	0.608	2.178	2.052	3.667	3.122

* days from sticking (31/12/95)

APPENDIX V

Table e: Plant growth records at marketing for *Osteospermum* cultivar 'Zulu' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	138.93	141.17	162.50	402.50	3.667	26.61	20.33	1.72	91.32
2	136.22	139.49	125.80	368.90	3.444	23.61	15.72	3.22	93.08
3	136.72	139.22	128.90	368.10	3.278	22.39	12.33	2.89	93.58
4	136.83	138.97	127.80	360.30	3.333	20.61	10.33	3.44	92.17
5	138.89	141.90	149.70	396.10	3.389	25.22	18.11	2.11	87.86
6	140.78	141.86	183.30	423.30	3.778	27.67	19.44	2.44	86.62
7	141.17	141.87	170.30	411.17	3.333	28.72	19.94	2.11	88.02
8	137.33	139.53	128.90	367.80	3.222	23.61	13.33	2.22	92.61
9	138.13	139.57	119.20	355.60	3.222	24.61	12.00	5.00	88.53
10	137.44	141.12	133.60	361.40	3.167	23.67	12.61	2.33	90.00
11	135.33	139.28	125.30	343.60	3.667	20.50	8.39	4.50	92.64
12	135.50	138.33	120.30	336.40	3.389	19.83	8.06	4.11	93.28
13	134.72	138.75	115.60	334.20	3.222	19.33	6.78	4.28	90.61
14	137.54†	140.08†	175.60	445.80	2.722	34.11	19.89	1.56	86.95
15	137.54†	140.08†	244.20	449.70	2.944	37.83	12.28	0.06	71.80
16	137.54†	140.08†	255.30	456.70	2.944	37.11	11.44	0.22	77.75
<i>SED</i>	1.068	0.908	11.640	14.040	0.335	1.325	1.324	0.850	2.190
LSD; P=5%	2.204	1.894	23.768	28.669	0.683	2.705	2.703	1.735	4.493

* days from sticking (31/12/95)

† estimated values

APPENDIX V

Table f: Plant growth records at marketing for *Osteospermum* cultivar 'SunnyGirl' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	125.78	132.47	201.10	455.30	3.278	20.39	1.33	5.83	80.03
2	121.32	128.08	113.30	377.40	3.144	15.44	4.50	5.67	84.79
3	122.94	128.70	118.30	367.80	3.056	14.67	5.22	5.67	84.47
4	127.11	129.28	133.90	380.00	3.167	14.89	7.28	6.06	84.61
5	124.17	129.89	140.60	404.40	3.200	16.78	3.17	5.06	84.39
6	125.94	134.30	141.70	419.40	3.611	19.89	1.94	5.11	84.17
7	124.94	135.28	140.30	409.20	3.556	19.33	2.22	4.33	84.40
8	122.61	128.17	119.40	378.10	3.389	15.00	4.39	4.11	82.17
9	122.44	127.67	100.00	355.00	3.111	15.22	2.39	5.39	86.06
10	125.72	131.33	122.20	366.70	3.000	17.44	2.83	5.28	83.22
11	130.78	128.52	158.30	408.10	3.222	19.39	4.89	3.67	80.19
12	132.83	133.69	201.70	438.30	3.222	23.06	4.17	3.33	83.99
13	125.00	128.22	103.90	351.40	3.389	15.39	4.33	7.89	86.89
14	125.22	131.86	123.90	380.00	3.667	18.00	2.06	4.56	82.86
15	137.78	143.06	244.20	493.50	3.111	26.50	4.00	1.50	80.67
16	129.76	128.05	173.90	425.80	3.500	20.28	3.78	4.11	79.72
<i>SED</i>	3.924	2.513	41.820	39.290	0.293	3.383	1.008	1.304	3.795
LSD; P=5%	8.012	5.176	85.396	80.230	0.598	6.908	2.058	2.662	7.601

* days from sticking (31/12/95)

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Table g: Plant growth records at marketing for *Osteospermum* cultivar 'Sunny Gustav' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	127.78	130.72	254.70	456.40	5.278	21.28	6.89	13.44	66.53
2	124.83	128.33	206.40	414.20	5.278	17.39	11.67	11.39	65.06
3	125.89	128.50	182.20	370.60	4.722	17.83	7.72	12.39	65.03
4	128.17	130.28	185.00	352.50	4.500	17.56	5.83	10.50	66.33
5	126.00	129.44	220.80	430.60	5.278	18.50	13.39	10.00	65.69
6	128.50	132.06	213.60	425.00	5.556	19.56	7.72	11.33	63.53
7	129.89	134.00	218.30	436.40	5.389	20.83	6.44	12.17	62.97
8	127.06	128.39	167.20	373.30	5.167	16.78	6.50	10.50	65.11
9	124.33	128.44	144.70	345.80	5.167	16.89	3.72	10.39	64.94
10	123.17	127.61	144.40	346.90	4.944	15.89	1.94	8.67	63.81
11	128.00	129.22	175.60	359.20	5.056	16.06	5.94	13.50	65.92
12	127.78	128.67	165.60	357.50	5.000	16.17	4.44	12.72	63.31
13	127.11	128.56	155.00	33.60	4.444	16.72	2.67	11.89	65.31
14	134.83	137.11	284.40	459.40	4.944	27.33	8.78	8.22	63.58
15	137.61	141.00	291.70	485.80	4.556	31.06	8.56	8.39	64.11
16	141.56	139.05	348.83	532.80	4.667	32.17	11.61	4.44	62.33
<i>SED</i>	1.341	0.775	14.980	13.360	0.396	1.029	2.368	2.780	1.801
LSD; P=5%	2.738	1.587	30.589	27.281	0.809	2.101	4.835	5.676	3.677

* days from sticking (31/12/95)

APPENDIX V

Table h: Plant growth records at marketing for *Osteospermum* cultivar 'Lusaka' (average of 3 replicates, 6 plants per plot)

Treatments	Days to first anthesis*	Days to 3-4 flowers open	Height of foliage (mm)	Height of flowers (mm)	Branch No.	Leaf No.	Bud No.	Flower No.	Flower diameter (mm)
1	123.39	127.87	128.10	397.80	3.056	18.33	17.33	5.44	87.08
2	123.22	127.94	132.80	360.30	3.500	18.28	16.94	5.06	83.38
3	123.78	128.00	116.90	342.80	3.167	17.00	14.22	4.67	83.03
4	128.00	129.00	125.60	348.90	3.167	17.00	12.78	5.67	84.08
5	122.94	127.56	118.60	364.70	3.333	19.61	13.83	4.50	84.52
6	123.00	127.83	112.80	373.10	3.000	19.00	16.00	5.78	88.35
7	123.72	128.44	106.90	373.30	3.056	19.06	12.28	4.50	87.53
8	123.78	128.00	106.90	346.10	2.944	17.28	8.22	5.56	85.17
9	122.83	127.96	83.10	320.30	2.778	16.88	4.83	4.61	86.22
10	122.83	127.83	87.20	326.10	3.056	17.61	3.56	4.67	86.72
11	127.56	128.50	111.10	340.60	3.000	16.72	11.44	4.39	80.22
12	127.61	128.72	102.50	327.50	3.222	16.67	6.17	4.72	83.22
13	125.61	128.11	106.40	318.60	3.389	16.44	7.22	6.17	84.81
<i>SED</i>	0.789	0.547	10.370	15.750	0.174	0.495	2.087	1.043	2.638
LSD; P=5%	1.628	1.129	21.403	32.508	0.359	1.022	4.307	2.152	5.444

* days from sticking (31/12/95)

APPENDIX V

Table i: Change in mean number of leaves, including primordia, in shoots from point of pinch in cv. 'Zulu', with time following various chilling treatments (means of three replicate shoot dissections).

Treatment	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
Time from sticking (Days)																
78	10	10	-	-	-	-	-	-	-	-	-	-	-	10	-	-
81	14	10	-	-	-	-	-	-	-	-	-	-	-	14	-	-
85	16	14	16	-	14	15	15	-	-	-	-	-	-	14	14	-
88	15	16	15	-	16	15	-	-	-	-	-	-	-	16	14	-
92	22	16	16	17	15	19	20	19	16	22	-	-	-	20	19	20
95	20	19	15	18	22	20	20	20	19	-	19	19	20	20	20	19
100	26	26	34	25	24	27	22	31	32	26	27	29	26	26	23	25
103	29	27	26	25	26	27	30	26	29	28	25	27	26	27	27	27
106	34	-	26	-	33	33	31	-	28	31	-	-	-	32	31	32
109	-	-	-	-	-	-	32	-	-	-	-	-	-	36	32	32
113	-	-	-	-	-	-	-	-	-	-	-	-	-	41	35	30
116	-	-	-	-	-	-	-	-	-	-	-	-	-	37	40	35

APPENDIX V

Table j: Regression coefficients for analyses of leaf initiation against time from sticking (days) for cv. 'Zulu' after a range of chilling treatments.

Treatment	Constant	<i>SE</i> Y	r^2	D. f.	X coefficient	<i>SE</i>
1	-51.30	2.13	0.94	7	0.78	0.077
2	-48.02	1.74	0.94	6	0.72	0.074
3	-51.10	5.52	0.58	5	0.76	0.289
4	-63.12	1.87	0.89	2	0.87	0.219
5	-58.95	2.24	0.91	5	0.84	0.117
6	-56.43	1.77	0.94	5	0.82	0.093
7	-58.55	1.98	0.90	4	0.84	0.136
8	-63.98	4.27	0.62	2	0.90	0.500
9	-74.70	4.61	0.67	3	1.00	0.403
10	-31.81	0.60	0.98	3	0.59	0.052
11	-63.05	3.27	0.70	1	0.87	0.571
12	-83.91	3.77	0.73	1	1.10	0.660
13	-50.89	1.72	0.86	1	0.75	0.301
14	-53.48	2.09	0.96	10	0.80	0.050
15	-57.87	1.40	0.98	8	0.82	0.044
16	-38.05	2.16	0.87	6	0.63	0.097