Number 316
December 1992

Acta Horticulturae

ISHS
International Society for Horticultural Science

International Workshop on Intensive Cultivation of Protea

Editors
G.J. Brits
J.T. Meynhardt

Pretoria
22 September 1991

Number 316
December 1992

Acta Horticulturae

ISHS
International Society for Horticultural Science

International Workshop on Intensive Cultivation of Protea

Editors
G.J. Brits
J.T. Meynhardt

Nevelian
2-7 September 1990

International Society for Horticultural Science
Société Internationale de la Science Horticole

ACTA HORTICULTURAE is a publication of ISHS.
Information about Acta Horticulturae and ISHS is given at the end of this book.

Secretary-General of ISHS
Ir. H.H. van der Borg

Secretariat of ISHS
Englaan 1
6703 ET Wageningen
Netherlands

Phone: (31) 8370 - 21747
Fax: (31) 8370 - 21586

International Workshop
on
Intensive Cultivation of Protea

Neve-Ilan

2-7 September 1990

Convener

Jaacov Ben-Jaacov

Editors

G.J. Brits

J.T. Meynhardt

Section for Ornamental Plants

Protea Working Group

International Workshop
on
Intensive Cultivation of Protea

Neve-Ilan

2-7 September 1990

Convener

Jaacov Ben-Jaacov

Editors

G.J. Brits

J.T. Meynhardt

Section for Ornamental Plants

Protea Working Group

Editors

G.J. Brits and J.T. Meynhardt
Agricultural Research Council
Vegetable and Ornamental Plant Institute
Private Bag
ELSENBURG 7607
Republic of South Africa

Organizers:

Section for Ornamental Plants
Protea Working Group

ISBN 90 6605 315 1
Price for non-members of ISHS: Dfl. 47,—

Printed by drukkerij Avanti, Schimminck 11, 5301 KR ZALTBOMMEL

CONTENTS

Session C: Control of Growth and Flowering - Pot plant production

This Acta *is an edited, non-refereed proceedings of the Israel Workshop - Editors.*

LIST OF PARTICIPANTS

Australia
Blakers, Ken
Daws, Joyce, International Protea Association, 26 Edwin Street, Heidelberg West, Victoria 3081, Australia.
Dupee, Steven, University of Sydney, Sydney, Australia.
McEwan, Lena, 29 Royal Pole, Parkville, Victoria 3052, Australia.
Sedgley, Ralph, School of Agriculture, University of Western Australia, Nedlands W.A. 6009, Australia.
Webb, Mark, Western Australian Department of Agriculture, Baron-Hay Court, South Perth 6151, Australia.

France
Allemand, Pierre, I.N.R.A., Route des Colles, Sophia Antipolis, 06410 Biot, France.
Visser, Schalk, South African Agricultural Attache to Israel, 59 Quai D'Orsay, 75009 Paris, France.

Israel
Ackerman, A., A.R.O. Misgav Experimental Station, Kiryat Shmona, Israel.
Ben-Jaccov, Jaccov, A.R.O - Volcani Center, P O Box 6, Bet Dagan 50250, Israel.
Calo, L., Moshav Sde Nitzan, Israel.
Folman, I., A.R.O - Volcani Center, P O Box 6, Bet Dagan 50250, Israel.
Kaddar, G., A.R.O - Volcani Center, P O Box 6, Bet Dagan 50250, Israel.
Karbalov, Dorit, Nit Nursery, Kfar Hes 406920, Israel.
Meltzer, Colin, Moshav Neve Ilan 90850, Israel.
Polag, Shuky, Moshav Neve Ilan 90850, Israel.
Shchori, Y., A.R.O - Volcani Center, P O Box 6, Bet Dagan 50250, Israel.
Silber, A., A.R.O - Volcani Center, P O Box 6, Bet Dagan 50250, Israel.
Tal, Eran, A.R.O. Misgav Experimental, Station, Kiryat Shmona, Israel.
Wallerstein, I., A.R.O - Volcani Center, P O Box 6, Bet Dagan 50250, Israel.
Watad, Abed, A.R.O - Volcani Center, P O Box 6, Bet Dagan 50250, Israel.
Wolfson, Dubbi, Ministry of Agriculture, Tel Aviv, Israel.

South Africa
Brits, Gert, V.O.P.R.I, Private Bag, Elsenburg 7607, South Africa.
Burger, Willie, V.O.P.R.I, Private Bag X293, Pretoria 0001, South Africa.
Coetzee, Cobus, V.O.P.R.I, Private Bag, Elsenburg 7607, South Africa.
Jacobs, Gerard, University of Stellenbosch, Stellenbosch 7600, South Africa.
Malan, Gerhard, V.O.P.R.I, Private Bag, Elsenburg 7607, South Africa.

Spain
Rodriguez, Juan, Escuela University de Ingenieria T. Agricola, Carretera de Geneto 2, 38200 La Laguna, Tenerife, Spain.

U.S.A.
Perry, Dennis, Perry's Panorama, 4800 North Street, P O Box 540, Somis California 93066-0540, U.S.A.

Alphabetical list of Authors

FLORICULTURE AND PROTEAS IN ISRAEL

J. Ben-Jaacov
Department of Floriculture
A.R.O. The Volcani Center
Bet Dagan 50-250
Israel

The floricultural industry for export started in Israel in the late 1960's. It is now the second largest horticultural industry for export in Israel totalling about 150 million US$.

Israel is the third on the list of flower exporting countries after the Netherlands and Colombia.

The following factors determine the importance and expansion of floriculture in Israel:

1) Local markets are saturated with all economically feasible goods, therefore expansion is possible only in export.
2) Main limiting factor in agricultural production is water and floriculture has a high return per unit of water.
3) Israel has a suitable climate for winter flower production.
4) Close distance to the large European markets.

In order to survive in the highly competitive world floricultural industry it is necessary to diversify our production. Proteas are part of this diversification effort.

Proteas

The main advantage of producing Proteas in Israel is our closeness to the markets and our reasonably suitable climate for them. The main problems, however, are related to our high pH soils and water.

Although Proteas were introduced to Israel in 1974, commercial production is still limited to Banksias, Grevilleas, *Leucospermum patersonii*, *Protea obtusifolia*, *Leucadendron* "Safari Sunset", *L. discolor* and some other Leucadendrons. Expansion of planting is limited mainly because of the uncertainty of success.

To overcome soil problems and expand planting area, four main approaches are under trial and investigation:

a) Growing species and their hybrids suited to our soils (mainly lime-tolerant).
b) Using artificial growth media.
c) The orchard approach - the development of suitable rootstocks.
d) Combination of the above.

The main research activities are:

- Development of different methods for cultivation in inert media.
- Large scale introduction and testing of cultivars and wild species suited to our conditions.
- Grafting and testing rootstocks-scions-soils combinations.
- Rapid propagation by tissue culture.
- Development of proteaceous plants as flowering pot plants.

BREEDING PROGRAMMES FOR PROTEACEAE CULTIVAR DEVELOPMENT

G.J. Brits
Fynbos Research Unit, V.O.P.R.I.
(Vegetable and Ornamental Plant Research Institute)
Private Bag
7607 Elsenburg
South Africa.

Abstract

Competition in floriculture is rapidly increasing in the form of improved cultivars and whole new cut flower crops. The slow growing protea is therefore urgently in need of imaginative genetic improvement.

Severe bottlenecks in current breeding technology include inability to hybridize between species, low hybrid seed set and lack of pollen storage techniques. Inefficient evaluation of new breeding products (regional selection) slows down local and international commercial introduction of improved cultivars. To break the pattern of breeding via accidental discoveries, future research should address these problems through concerted and cooperative effort. Instrumental to this is the basic level study of protea reproductive biology and embryo rescue technology. The breeding advantage of the great natural protea gene pool must be exploited systematically for the development of market oriented cultivars.

1. Introduction - the need for protea breeding improvement

Speakers at the most recent international Protea growers' conferences have pointed out that world cut flower marketing turnover has stabilised. The protea cut flower industry is therefore faced with the realities that its "honeymoon" days as a novelty product are over and that its role as a mature product is to grow at the expense of other crops.

Cut flower marketing is a highly competitive field. Whole new flower crops are launched annually into the marketplace and entire scientific symposia are dedicated to the subject of new flower crops (Christensen, 1989).

Proteas are relatively slow growing plants, their regeneration cycle being in the order of 3-4 yrs. This disadvantage, typical of most woody crops, has two important implications in market development: a) that the genetic improvement tends to be relatively slow (e.g. slow turnover rate of cultivars) and thus b) the progress of the crop as a whole is significantly slower than in the case of herbaceous crops with their relatively short life cycles.

Roughly only 25% of all currently marketed protea material is produced from *selected* species and cultivar material. This underscores the slow progress of the protea in the marketplace, where most other floricultural crops are traded only as improved cultivars. Furthermore it is estimated that approximately 95% of all protea cultivars grown are natural crosses, accidentally discovered in flower orchards/gardens and in nature. An example is the well-known *Leucospermum* 'Red Sunset', an F_1 hybrid which originated in South Africa, and which apparently was a chance discovery in a seed batch. This mode of cultivar improvement has become the established pattern for proteas.

It is clear that systematic, controlled breeding towards well-defined goals is essential in order to advance proteas to their full potential as floricultural crops. To this end a look must be taken at the bottlenecks experienced in protea breeding.

2. Bottlenecks in protea breeding

2.1 Crossing incompatibility between species

Circumstantial evidence suggest that hybridization between species is the most useful approach to protea breeding at this time. Strong hybrid vigour is experienced as a rule in interspecies crosses, commonly increasing yields up to 100-150%, e.g. in *Leucospermum* 'Scarlet Ribbon' and *L.* 'Sunrise' (Brits 1985a; b). In addition the definite horticultural weaknesses inherent in most Proteaceae species can theoretically be complemented by gene transfer from other species. Examples are the undesireable lateral shoot growth from below *P. repens* inflorescences, which do not occur in closely related species e.g. *P. aristata*; unsuitable winter flowering period of *P. compacta* in the southern hemisphere - as opposed to summer flowering *P. grandiceps, P. aristata*; and strong preference for acidic soil in *P. magnifica*, compared with calcicole species such as *P. obtusifolia*.

However artificial hybridization has thus far given disappointing results (Brits 1983). This is especially a feature of the genus *Protea* (table 1).

Species of the Proteaceae family are mostly allopatrically reproductively isolated; prolonged geographic isolation of populations is thus probably correlated with the high degree of endemism of species. This may explain the absence of the chromosomal reproductive isolation mechanisms functional in many other plant taxa and may underlie the constancy of chromosome numbers (De Vos 1943) within Proteaceae genera. The incompatibility mechanisms are therefore likely to be more of a physiological nature. Physiological incompatibility mechanisms are generally less difficult to overcome than chromosomal islolation mechanisms. Therefore the prognosis for finding solutions to incompatibility in proteas is relatively good.

Nevertheless the extremely low seed set routinely found in interspecies crosses presents a vexing problem. Hybrid seed set will generally decline drastically with slight increases in phylogenetic distance - e.g. in *Leucospermum* (table 2). Self-incompatibility varies from strong in *Protea* (table 1) to moderate in *Leucospermum* (table 2).

2.2 Low natural seed set (excepting *Leucadendron*)

Seed set is notoriously low in most Proteaceae species quite apart from the problem of crossing incompatibility (table 3). These plants presumably employ physiologically regulated low seed set as a nutrient economizing strategy. Thus low seed set could be an adaptive response to extremely nutrient poor soils. This problem must be addressed within the breeding programme, possibly by means of growth regulator research. The problem of low seed set is further aggravated by poor seed germination especially in the nut-fruited Proteaceae species. The most advanced germination techniques will in many cases not yield germination percentages of more than 25-50% (Brits, unpublished).

2.3 Lack of pollen storage techniques

A strongly exploitable characteristic of Proteaceae species is their widely differing flowering periods. An example is the mid-summer flowering characteristic of *Protea aristata* during a period of peak demand for southern hemisphere flowers, as opposed to the winter/spring flowering period of most other species. The transfer of this and other characteristics of species with disjunct flowering periods are coveted goals in breeding programmes.

In order to successfully cross species flowering in different seasonal windows the effective storage of viable pollen is required. Techniques for this have not yet been fully developed. Desiccated pollen can presently be stored for up to 6 w at 5 °C (Brits & van den Berg, 1990). Clearly the systematic study of pollen cryo-preservation in different species are indicated (figure 1). To this end an effective test for pollen germinability is also needed. Germinating pollen in 12% sucrose and 100 mg.l^{-1} H_3BO_3 gives reasonable results but more study is needed (figure 2).

2.4. Insufficient evaluation of new breeding products

New cultivars are presently spread worldwide in an opportunistic fashion. There are many current so-called cultivars which are really little more than ordinary examples of species, named and released by especially garden nurseries. Such nurseries are often obliged to denominate plants with popular names in order to attract a clientele. Thus very little improvement of the species standard is associated with some of these releases (Brits, 1988).

Responsible regional research organizations can improve on this situation by a) identifying promising cultivars of foreign origin b) evaluating these cultivars under local conditions in cooperation with the owner/breeder c) followed by controlled release of material to the industry.

One danger of an all too orderly introduction and cooperative evaluation scheme is the trend of official bodies to "sit" on material, disregarding the need for speedy evaluation and release. A useful approach to this problem is to cooperatively involve selected producers, who could keep the pressure on institutions for the speedy release of superior introduced material.

Another potential problem in international distribution and evaluation is the reluctance with which breeders normally surrender their breeding material out of their immediate control. Breeders should consider the substantial advantage of *in situ* evaluation of their material at reduced cost by an agent cooperating in another country. Also the possibility that the material could subsequently be found acceptable and then introduced commercially in that country. Reasonable control over material is possible via legal agreements, following the guidelines proposed by IPWG for different negotiable classes of breeding material (Parvin, 1989).

3. The need for cooperative breeding research

The global manpower currently available for protea breeding improvement - and the international protea industry itself - is too limited for effective advance by means of individualistic breeding programmes. In addition much overlap of breeding goals exists. It can therefore be argued that the coordination of breeding efforts are essential in order to advance common breeding goals maximally.

Table 1 - Summary of typical seed set in *Protea* following artificial cross- and self-pollination, using 60 florets per flower head; self-pollination was active (A): controlled, or passive (P): intact flower head, covered; intra-specific crosses included the hybridization of different ecotypes.

	♀ Parent					
	aristata	compacta	cynaroides	eximia	pudens	repens
♂ Parents	aristata repens	compacta eximia [eximia x susannae "Sylvia"]	cynaroides	eximia [eximia x compacta] repens	repens obtusifolia -hybrid [lacticolor x aurea "Ivy"]	repens
Florets pollinated	540	420	4800	360	540	2520
% seed set	0,0	1,2	20,4	0,0	0,0	24,6
Self-pollin.	60(P)	60(A)	60(P)	-	60(P)	-
% seed set	0,0	0,0	0,0	-	0,0	-

Table 2 - Seed set % in *Leucospermum cordifolium* T75 11 03 cross pollinated reciprocally with four selections of varying affinity; and self-pollinated actively (controlled) (⊗) or with flower heads left uncovered (open-pollinated).

		Reciprocal ♀		
♂ Parent	T75 11 03	♀ Reciprocal	Active ⊗	Open-pollin.
L. cordifolium "Flamespike"	0,5	6,0	1,0	8,0
L. cordifolium "Vlam"	1,2	0,15	0,0	1,0
L. vestitum T75 11 27	0,3	0,8	12,0	15,0
L. reflexum	0,15	0,0	0,3	5,0
Active ⊗	0,15	-	-	-
Open-pollinated	4,0	-	-	-

Table 3 - Seed set percentage in open-pollinated *Protea* and *Leucospermum* flower heads (ex Horn, 1962)

Protea		*Leucospermum*	
species	seed	species	seed
compacta	16,8	*bolusii*	2,4
lacticolor	5,2	*conocarpodendron*	4,7
magnifica	2,8	*cordifolium*	2,2
minor	1,2	*cuneiforme*	1,2
obtusifolia	1,2	*reflexum*	2,3

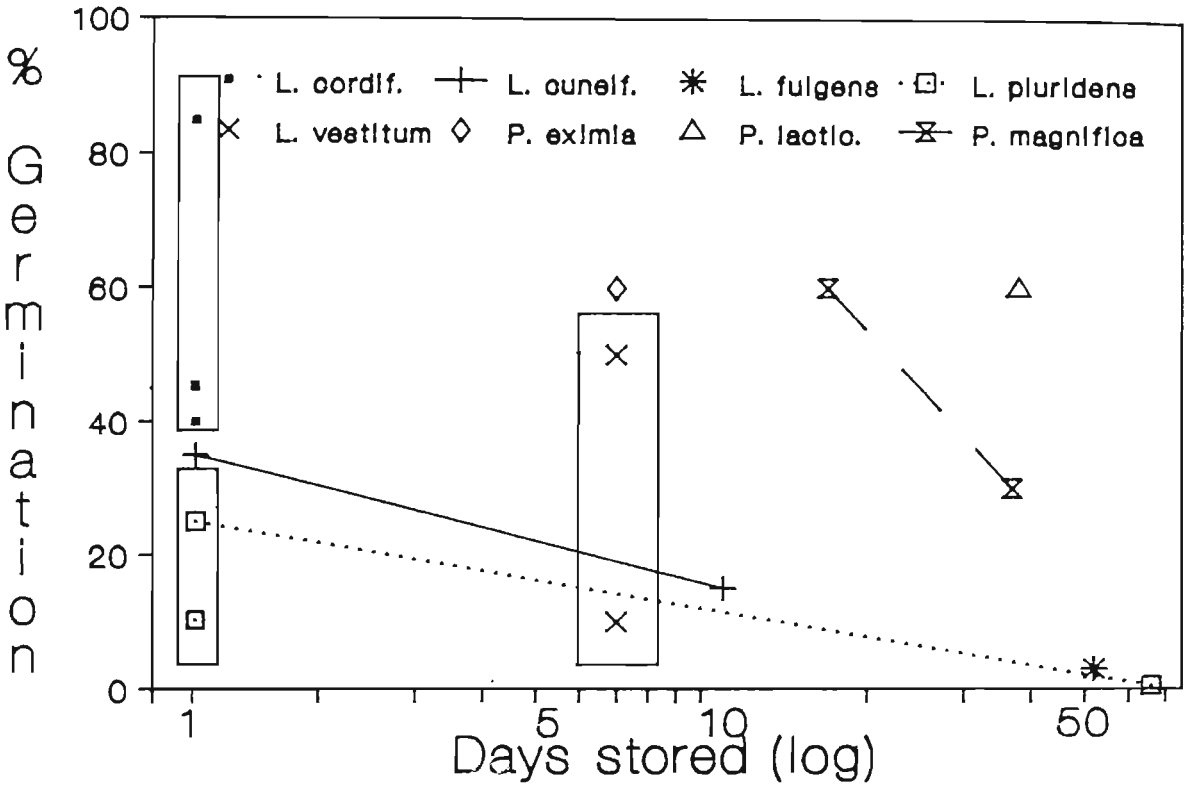
Table 4 - South African protea breeding problems on which co-operative research could ideally be conducted (some projects* already initiated)

Mutual Problem	S. A. Contribution	Cooperation
Protea leaf blackening	Selected material; screen gene source	Develop leaf blackning test (U.S.A / Australia)
Nematode resistance in *Leucospermum*	Breed rootstocks	Field testing (U.S.A.)
P. cinnamomi tolerance in *Leucospermum* rootstocks	Breeding, regional testing	Breeding, basic P.c. study (Australia)
Lime tolerance: *Leucospermum*, *Protea* rootstocks*	Gene source exploitation, regional testing	Field testing, compatibility study (Israel)
Flowering pot plants*	Breeding; technology development	Technology development - microprop.; market test (Israel)
Banksia, Waratah improvement	Bilateral material exchange; testing	Exploit gene sources , breeding (Australia, New Zealand)
Cultivar evaluation	Regional testing	Regional, market evaluation (All countries)
Protea cut flower improvement	Gene sources, breeding	Material exchange (All countries)

Table 5 - Breeding priorities and sub-programmes within the V.O.P.R.I. hibridization programme, for the three main Proteaceae genera; recently initiated programmes are indicated*

Leucadendron	*Leucospermum*	*Protea*
Larger flowers	Late flowering:	Smaller flowers
Late flowering	- natural lateness	Summer flowering
Deeper, varied colour	- ethephon treatability*	Vigour, long stems
Flowering pot plants*	*P.c.* tolerant rootstocks	Improved colour
Phytophthora tolerant rootstocks	Better colour, shape	Improved shapes
	Flowering pot plants *	Less leaf blackening

Figure 1 -Examples of 5 ^{o}C pollen storage in *Leucospermum* and *Protea*. Boxes enclose different pollen sources (plants) of the same species; lines connect germination values of the same pollen source tested at different times. Pollen was germinated in 12% sucrose + 100 mg $H_3BO_3.l^{-1}$



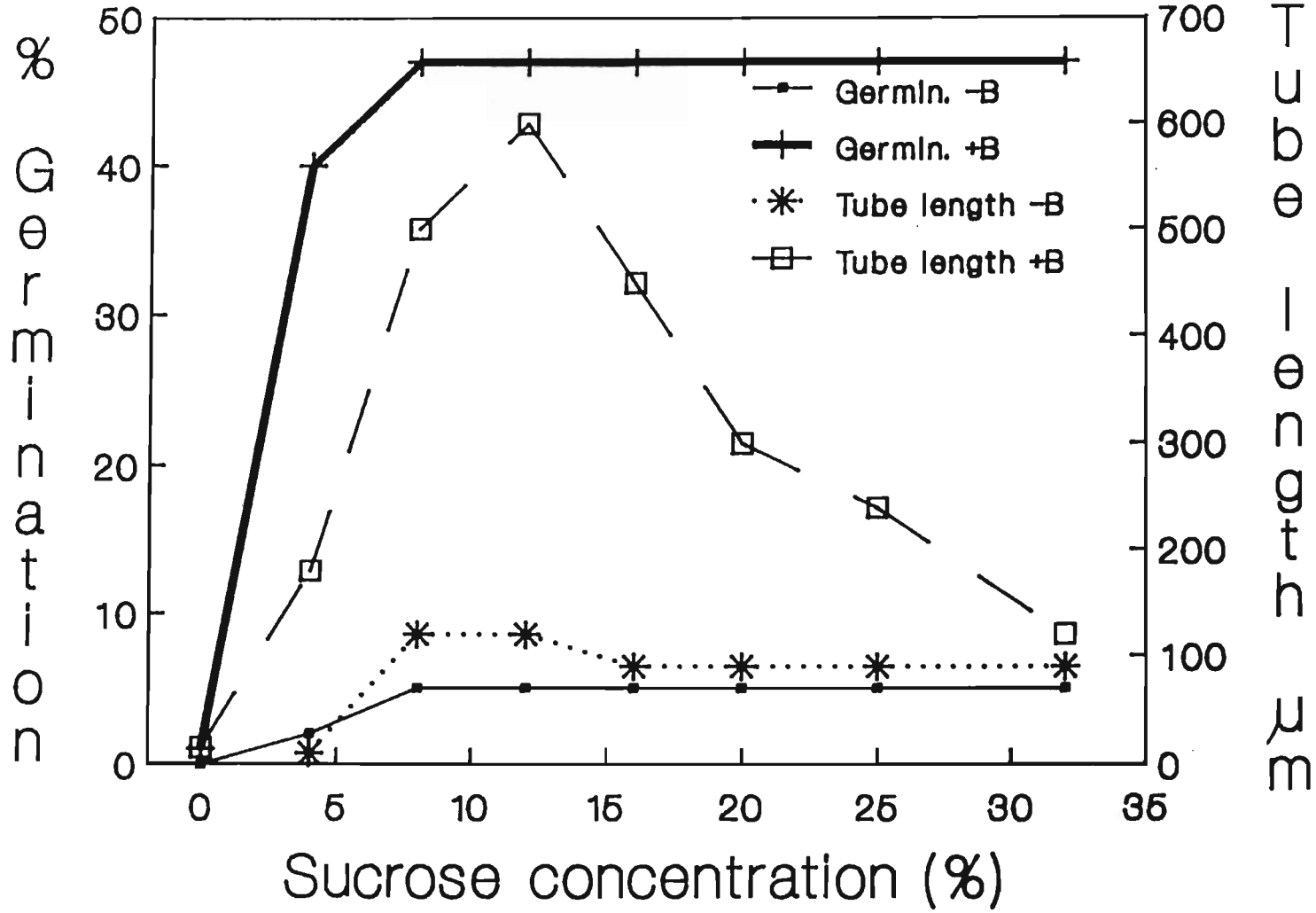


Figure 2 -Effect of sucrose and boron (100 mg $H_3BO_3.l^{-1}$) on pollen germination percentage and pollen tube growth in *L. cordifolium*

POLLEN GERMINATION AND STORAGE IN *BANKSIA* AND SOME OTHER PROTEACEAE PLANTS

Y. Shchori, Tal Goren and J. Ben-Jaacov
Dept. of Ornamental Horticulture
A.R.O. The Volcani Center
Bet Dagan 50-250
Israel

Abstract

Pollen germination of *Banksia*, *Protea* and *Leucospermum* has been tested on several artificial media under different experimental conditions.

Stored *Banksia* pollen viability under different storage conditions has been tested.

1. Pollen germination

In vitro pollen germination of several Proteaceae species: *Banksia ashbyi*, *B. integrifolia*, *B. menziesii*, *B. hookeriana*, *B. prionotes*, *Leucospermum patersonii*, *L. cordifolium*, *L. cuneiforme* and *Protea obtusifolia* were studied.

Pollen germination tests were performed on glass slides in humid chambers at room temperature (22-25°C). The germination media were series of liquid sugar, including Taylor's medium (Taylor, 1972).

Table 1 - Pollen germination percentage of Proteaceae species in two media in upright and in hanging drops.

Plants	Medium:	Taylor's		10% sucrose + 200 ppm B	
	Drop:	hanging	upright	hanging	upright
B. ashbyi		90	60	20.0	6.3
B. prionotes		46	46	7.0	-
B. integrifolia		45	52	-	-
B. hookeriana		35	30	-	-
B. menziesii		20	25	-	-
L. cordifolium		55	36	5.0	5.8
L. patersonii		30	31	6.5	6.2
P. obtusifolia		40	12	6.0	6.0

Highest germination rate was obtained on Taylor's medium in a hanging drop for all the species tested. The maximum germination rate was reached after 22 hours. Some of the results are presented in table 1.

2. Pollen Storage

The effect of storage on pollen viability was examined during four weeks in five *Banksia* species.

Pollen samples were stored in:

1) Room temperature (22-25°C)
2) 5°C refrigerator
3) Desiccator at room temperature
4) Deep-freezer

Pollen germination after storage is shown in figures 1 and 2.

Highest viability level over all storage periods was achieved at 5°C for all the species examined.

5°C cooling showed higher viability than deep freezing during four weeks for all the species tested. In all the tested species a large decrease in viability occurred after the first week of storage. At room temperature low viability was maintained for two weeks and this was prolonged in some cases by dehydration.

In conclusion for viability testing, Proteaceae pollen should be tested immersed in Taylor's medium in a hanging drop. At room temperature germination test counts should be made after 24 hours.

Pollen viability may be maintained to a reasonable degree at 5°C for at least one month.

References

Taylor, R.M., 1972. Crop Sci. 12: 243.

Figure 1 - Effect of storage on pollen germination

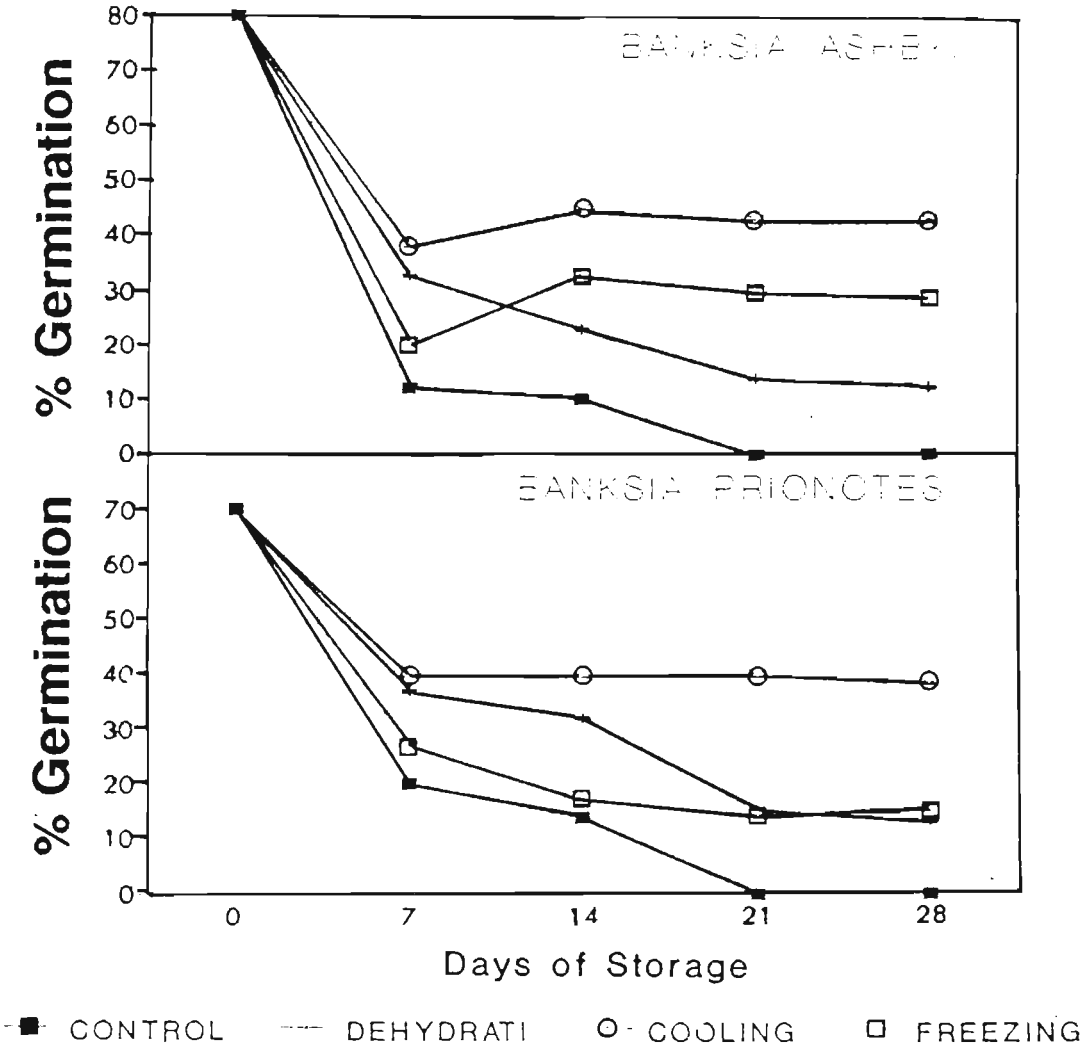
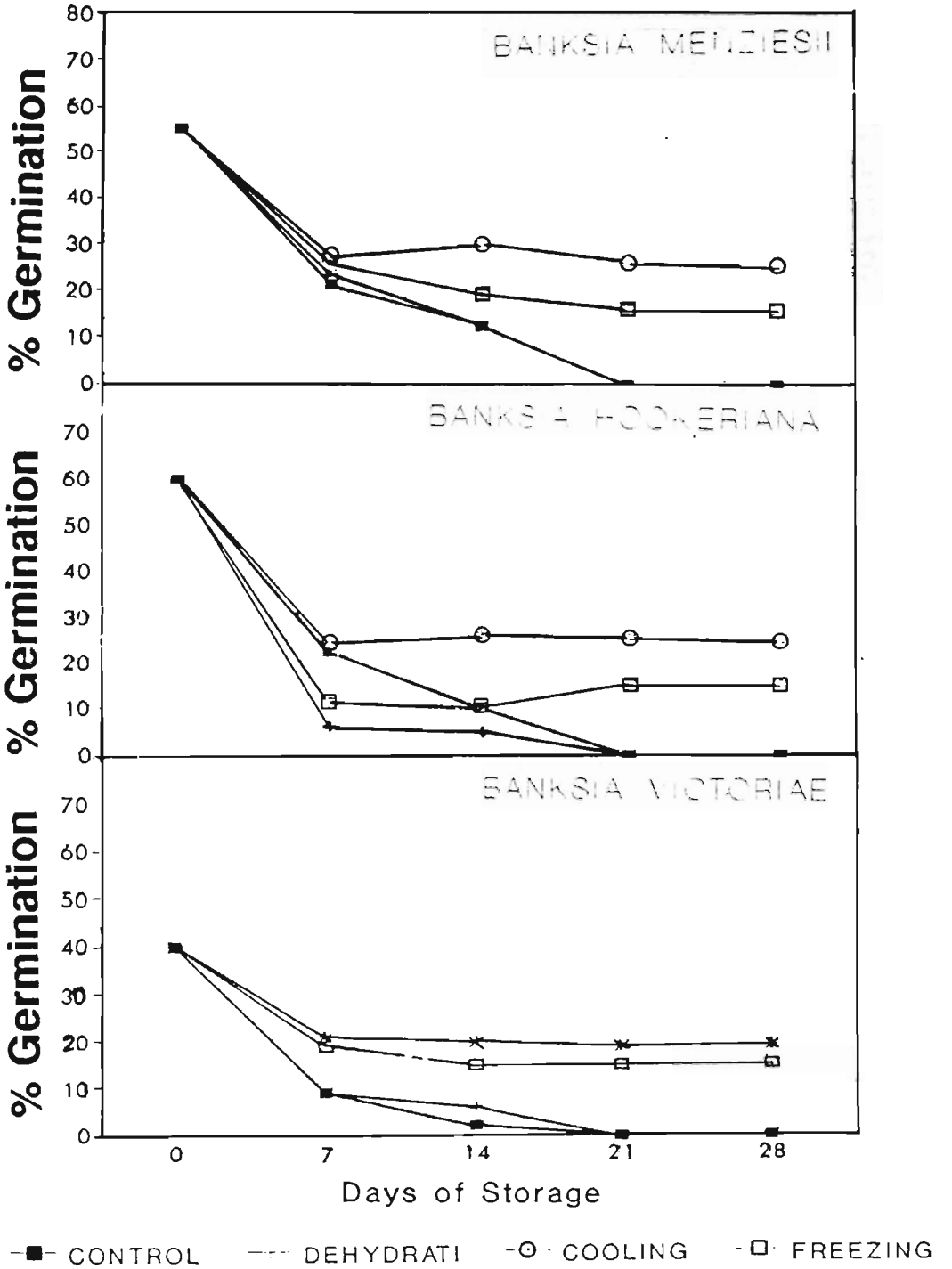


Figure 2 - Effect of storage on pollen germination



AN OVERVIEW OF THE NATIVE FLORA AND PROTEA INDUSTRIES IN WESTERN AUSTRALIA

Mark G. Webb
Western Australian Department of Agriculture
Baron-Hay Court
South Perth 6151
Australia

Abstract

The export of native cut flowers and foliage from Western Australia has increased in importance over the past ten years. From an industry based solely on material picked from the bush, it has developed into a $A7.5 million industry with an increasing amount of product being intensively cultivated on farms. Bush picking is still important for product diversity and for providing material which cannot be propagated, or for which low returns make intensive management unviable.

Proteas have only been grown commercially in Western Australia since 1984, with most plantings occurring in the past three to four years. The current value of exports, mainly to Japan, is estimated to be less than $A1 million.

The export from Western Australia, of cut flowers and foilage from native flora and proteas, could increase by up to 400% over the next two to three years as many plantings reach full production. Industry will undoubtedly be faced with challenges as they promote their product and ensure that market demands are met.

1. Introduction

The intensive cultivation of Australian native flora (native flora) and south African Proteaceae (proteas) for cut flower and foliage production is a relatively new industry in Western Australia (WA). The export of cut flowers and foliage from Western Australia to Europe, Japan and the USA has increased from $1.3 million in 1980/81 to $8.5 million in 1989 (Australian Bureau of Statistics, personal communication).

2. Native Flora

2.1 Background

The export of cut flowers and foliage of native flora from WA began around the 1950s with material harvested from the bush. Most of the plant material was harvested from designated areas in State forest. Operations were generally small scale and provided a supplement to the normal family income. In the 1960s and 1970s, pickers became more professional and bushpicking became the sole source of income for some. In 1980/81, a total of 550 species were used by the industry; 288 for cut flowers and foliage, 308 for seed and 166 for nursery cuttings (Burgman and Hopper, 1982). The most heavily exploited genera for cut flowers and foliage were *Boronia, Verticordia, Stirlingia, Agonis, Banksia* and *Dryandra*. Most of the material was dried, and often dyed with only a small proportion being sold fresh.

The importance of bush picking has declined in recent years although it is still important for providing cut flowers and foliage from species that cannot yet be propagated, that occur widely in the bush or that appear to be uneconomic under intensive cultivation.

2.2 Current Situation

2.2.1 Cultivated Stands

The planting of selected native plant species as row crops under drip irrigation and fertilizer followed in the early 1980s, partly in response to an increasing demand for species which were not widely distributed in the bush. The main genera planted were *Banksia*, *Anigozanthos* (kangaroo paw) and *Chamelaucium* (waxflower). Production from these plantings, together with improved air freight services, provided the impetus to further develop the export of fresh cut flowers and foliage. Marketing difficulties, and some production and management problems caused this expansion into intensive cultivation to briefly falter in 1982. Renewed interest, and corporate investment in the mid 1980s, resulted in a rejuvenation of old plantings, and considerable new plantings. More that 1,000 ha, involving over 50 species of native flora were under intensive cultivation in WA in 1988 (Pegrum, 1989). It is estimated that in Australia in 1990, there were approximately 800,000 waxflower plants and 600,000 kangaroo paw plants under intensive culivation, of which 500,000 and 300,000 respectively were grown in WA (Pegrum, 1990). Based on plant numbers, the potential harvest of *Banksia*, *Anigozanthos* and *Chamelaucium* from cultivated stands in WA could increase by 1992 to a total of 71 million stems (Pegrum and Webb, 1990).

It has become obvious that in WA in the mid 1980s, an insufficient range of some species and cutivars, particularly waxflowers, were planted. Too much reliance was placed on the 'Alba' and 'Purple Pride' forms of *Chamelaucium uncinatum*. Also, major plantings in a single region provided no natural spread of harvest which might have been achieved by planting the same species and cultivars over a wider climatic zone. As a result, the large increase in production over the last two years has led to some oversupply from July to September on export markets, especially of waxflowers. New plantings are concentrating on spreading the harvest period by careful selection of a range of species and cultivars.

The desire for "new" native flowers in the mid 1980s also led to the planting of species for which little or no agronomic or marketing information was available. Many of these species did not grow satisfactorily under cultivation, were not suitable as export cut flowers, could not be grown economically, or were not marketed correctly. Consequently, some of these species have been removed in the past 12 months.

2.2.2 Managed Stands

Since the mid 1980s, there has been considerable interest by farmers and pickers in "semi-managing" native flora, particularly *Agonis* and *Banksia*, that occurs naturally on uncleared, or regenerated private land. This involves chaining and burning (*Agonis*), or pruning or reseeding (*Banksia*), for future production. Some fertilization and insect control may be conducted with access tracks cut to assist harvesting. In the lower south west region of Western Australia, it is estimated that up to 1,000 ha of *Agonis* may be semi-managed (Webb and Dawson, 1989).

2.2.3 Insects and Diseases

Insects and diseases can cause slight to severe problems on native flora. There are three main types of insects causing damage:

(i) those that can kill plants, e.g. a native weevil that attacks Myrtaceous plants. *Chamelaucium* and *Verticordia* spp. are particularly vulnerable;
(ii) those that affect marketability, e.g. various beetles, weevils, caterpillars,

sucking bugs and thrips;

(iii) those that do not necessarily cause damage, but can lead to phytosanitary problems e.g. various thrips, bees, wasps, ants and beetles.

Die-back, caused by fungi of the genus *Phytophthora* is the most important disease of native flora. Many growers have instituted strict hygiene measures to limit the spread of this devastating disease.

2.3 Marketing

Like the rest of the industry, the marketing of native flora is in its infancy. In the past, many agents merely acted as "freight forwarders", with limited experience in flower marketing. This is changing as exporters realize that when supplied in quantity, native flora, no matter how special, loses some of its novelty value and long term marketing plans must be developed.

The development of any marketing plan relies on the ability of growers and pickers to harvest a high quality product, and on all sectors of the industry to correctly handle the product to ensure a vase life of at least seven days. Considerable success has been achieved in post-harvest research over the past five years resulting in improved product quality on export markets.

3. Proteas

3.1 Background

Interest in protea production in WA has increased since 1984 with over 200 ha planted (Webb, 1990). Proteas grow well on the infertile, acid sands common throughout WA. Early plantings were of a wide range of species, mainly seed grown. In some cases, plantings of seed grown material has provided useful selections for subsequent cloning.

3.2 Current Situation

In the past three years, new plantings have been mainly of selected, cutting grown material, with particular emphasis on *Leucadendron* spp. Based on plant numbers, the potential number of stems of *Protea*, *Leucadendron* and *Leucospermum* from WA could increase to over 20 million by 1992 (Pegrum and Webb, 1990). Increasingly, some growers are planting a combination of native flora and proteas, using species from each group to optimize farm operations and maximize market opportunities.

Nearly all the proteas in WA are planted in rows under drip irrigation and regularly fertilized. To minimize potential problems with *Phytophthora* spp., many growers have instituted strict hygiene measures. Other diseases tend to be minor although there are several leaf diseases which can be identified symptomatically, but which have proved difficult to isolate and positively identify. Insect problems are generally minor but can cause some phytosanitary problems on export shipments.

3.3 Marketing

It has only been in the last two to three years that significant quantities of proteas have been exported from WA, mainly to Japan. The value of protea exports is difficult to determine but is probably less than $1 million. The marketing of proteas faces similiar problems to native flora, and some of the solutions will probably be similar for both groups.

4. Conclusion

The area of native plants and proteas under intensive cultivation in Western Australia has increased substantially since 1985. Production from these stands is expected to increase to over 90 million stems by 1992. This increase in supply will require an unprecedented marketing programme involving all sectors of the industry.

References

Burgman, M.A. and Hopper, S.D., 1982. The Western Australian Wildflower Industry, 1980-1981. Report No. 53. Western Australian Department of Fisheries and Wildlife.

Pegrum, J.A., 1989. Outlook for floriculture. In: Agricultural Outlook for Western Australia - 1989, pp. 71-75. Western Australian Department of Agriculture Miscellaneous Publication No. 7/89.

Pegrum, J.A., 1990. Plantings of Wax and Kangaroo Paws across Australia. The Floriculture Industry Newsletter, 13: 8-13. Western Australian Department of Agriculture.

Pegrum, J.A. and Webb, M.G., 1990. An overview of the western Australian native plant and protea industries. J. Int. Protea. Assoc. 19: 30-33.

Webb, M.G., 1990. The effect of selected herbicides on three *Protea* species. J. Int. Protea Assoc. 19: 35-38.

Webb, M.G. and Dawson, P.G., 1989. Bushpicking of wildflowers. Miscellaneous publication no. 11/90. Western Australian Department of Agriculture.

PROPAGATION OF PROTEACEAE

Daniel G. Malan
Fynbos Research,
Vegetable and Ornamental Plant Research Institute,
Private Bag,
7607 Elsenburg,
South Africa.

Abstract

The protea industry has advanced to the stage of growing registered cultivars and selected plant material. Since a multitude of problems are normally experienced with specific soil requirements and certain root diseases of proteas, cultivation practice will probably soon advance to the extensive use of rootstocks. Propagation by seed will probably be restricted to scientists and growers attempting to breed or select seedlings with improved characteristics such as resistant or tolerant rootstocks, improved or different cut flowers or for flowering pot plants. The vegetative propagation of proteas is, however, still not considered easy, and this inhibits the development of a nursery industry. This discussion introduces the techniques currently employed in the propagation of proteas, identifies some of their shortcomings and suggests research priorities.

1. Introduction

In only a few years the protea industry has developed from picking flowers in the wild (Brits *et al*, 1983) to a level where there are currently more than 150 commercial protea cultivars available (Brits, 1988). This development of proteas as internationally cultivated cut flowers [Israel, Spain (main land, Majorca and Canary Islands), Portugal (mainland and Madeira), France (mainland and Corsica), U.S.A. (California and Hawaii), New Zealand, Australia and Zimbabwe] can to a large extent be contributed to the perseverance of protea growers and the exceptional demand for these flowers. Protea propagators can currently expect success rates varying from 0 - 100% in cutting production and between 0 - 80% in grafting and budding . Only six selections out of 62 genera containing approximately 1400 species have been successfully established in tissue culture. These disappointing figures result in proteas still being considered inherently difficult plants to propagate. The techniques currently employed in protea propagation are presented below and some of their advantages and shortcomings discussed.

2. Vegetative propagation

Most commercial protea varieties are propagated by using approximately 20 cm long terminal semi-hardwood cuttings. A 5 second 5 mm indole butyric acid (4000 $mg.dm^{-3}$) basal dip is used and the cuttings are planted in a well aerated medium with intermittent mist and 22-25 °C bottom heat (Rousseau, 1967; Blommaert and Rousseau, 1971; Jacobs and Steenkamp, 1975; Harre, 1988; Malan, 1988). A minimum rooting percentage of 80%, regarded as ideal for effective nursery propagation, is however seldom achieved. This is probably not due to a single factor. One principle has however emerged: proteas as a group of crops require different propagation environments and treatments to the majority of other cut flower crops (Brits, 1986c). These requirements have to be adapted according to the needs of every cultivar and selection for optimum results (Jacobs and Steenkamp, 1976; Brits, 1986c; Harre, 1988). Few scientific results are available on the rooting parameters of the different proteas. Although the cultivation of proteas

on broad spectrum compatible rootstocks would allow for greater standardization of the rooting environment, it is currently strongly desirable that individual cultivars should be released with guidelines on the most important aspects of their rooting. These guidelines should include the following.

A. Environmental factors:

1. Rooting temperature requirement: Most protea selections and hybrids root satisfactory at 22-25 °C, but some will root better at lower temperature (Mackay, 1985; Brits, 1986c; Perry, 1988). More cultivar specific studies on temperature requirements are needed.
2. Light intensity: Different varieties (hairy vs. others) apparently require different light intensities for optimal results (Harre, 1988). This aspect needs to be investigated.
3. Rooting medium aeration: Perforated bags increased rooting of some cultivars considerably, while it had little influence on others (Harre, 1988; Malan, 1988). This suggests that aeration of the medium may be an important factor.
4. Misting frequency: Although most proteas will root best when supplied with hourly mist during daylight hours (Brits, 1986c), some varieties apparently perform better with the more conventional wet-leaf system (Perry, 1988; Parvin, 1979).

B. Treatment of cuttings:

1. Time of harvesting: In *Leucospermum*, *Leucadendron*, and *Serruria* all species terminate shoot growth during autumn and cuttings are harvested shortly there after as semi-hardwood cuttings. With *Protea*, *Mimetes*, *Banksia*, *Telopea* and *Grevillea* growth cycles are less synchronized (Mackay, 1985; De Swardt, 1989; Malan, pers. observation), and all cuttings are not necessarily at the optimum stage for rooting if harvested at the same time (Malan, pers. observation). Manipulation and preparation of mother plants and more careful selection of cutting material are almost totally neglected fields of study which can greatly affect the performance of cuttings (Harre, 1987; 1989a; Perry, pers. comm.).
2. Growth regulator treatment.
   a. Auxin concentration: Research results on the optimum concentrations for different varieties vary considerably (Rousseau, 1967; Perry, 1988; Jacobs and Steenkamp, 1976; Harre, 1989). The requirements of individual cultivars should therefore be evaluated.
   b. Auxin carrier: Apparently talcum powder as a carrier is best under low rooting temperatures and 50% ethanol at higher temperatures (Brits, 1986; Rousseau, 1967; Gouws *et al*, 1990). More research is however needed.
   c. Additional root promoting treatments: Addition of certain growth regulators ("cocktails") e.g. gibberellic acid, ethrel and daminozide (Criley *et al*, 1979; Brits, 1986; Gouws *et al*, 1990) in addition to indole butyric acid; scarring of the base of the cutting (Perez, 1990) and other pre-setting treatments (Harre, 1989b) have been used with varied success. Specific additions for certain cultivars should however be investigated.
3. Type of cutting.
   a. Terminal vs. subterminal cuttings: Terminal cuttings in general root better and faster (Brits, 1986c), but subterminal cuttings can often be used effectively, especially in the types that root more easily

(Harre, 1988). Terminality interacts strongly with other rooting factors (Brits, 1986c) and should therefore be determined for individual cultivars.

b. Length of cutting: The 20 cm cutting is used routinely, but preliminary results indicate that shorter cuttings, down to the single node type, can be rooted effectively (Perez, this Acta). The economizing potential of single node cuttings, for use when propagating scarce material, is an important aspect to investigate.

A number of indirect factors may also contribute to suboptimal rooting of cuttings:

1. Inability to control diseases: Despite stringent sanitation (Benic, 1986; Forsberg, 1988), diseases often break out in the rooting frame and this results in numerous losses and thus a reduced success rate in propagation.
2. Physiological maturity of mother material: The growth mode of non-lignotuberous proteas, resulting in death of basal axillary buds after 18 to 24 months; and continual vegetative propagation from successive mother plants result in wood of increasing physiological maturity being used as cutting material. In South Africa most non-lignotuberous protea cultivars are in the 5 - 6th vegetative generation when released to the industry, and 2 - 3 more generations are required to effect full production. This may contribute to the poor rooting ability of cuttings which is often encountered (Harre, 1987). Manipulation of mother plants or tissue culture of plants, to invigorate cutting material, may offer a solution to this problem.

3. Grafting and budding

With research currently aimed at the development of clonal rootstocks incorporating lime tolerance and *Phytophthora* and/or nematode tolerance the change to cultivation of proteas on rootstocks is inevitable. Grafting and budding can be applied in numerous ways and situations.

Situations in which techniques have been evaluated include:
a. Cuttings (Malan, 1989; Brits, 1990a).
b. Nursery plants: rooted cuttings and seedlings (Rousseau, 1966; Vogts *et al*, 1976).
c. Micro grafting (Ackerman, pers. comm.).
d. Topworking of established plants (Rousseau, 1968; Harrington, 1988; Brits, 1990a).

Of these cutting grafting or budding seems to be the most promising technique as this is the fastest production method of plants with rootstocks, reasonably successful and the easiest to apply (Brits, 1990a).

Initial investigations indicated that both grafting and budding can be readily applied within and between protea genera (Van der Merwe, 1985; McCredie *et al*, 1985a). Techniques that have been evaluated include:

Grafting:

1. Wedge grafting (Rousseau and Blommaert, 1971; Vogts *et al*, 1976; Malan, 1988; Brits, 1990a).
2. Approach grafting (Brits, 1990a).
3. Tongue grafting (Ackerman, pers. comm.).

Budding:

1. Modified T-Budding (Harrington, 1988; Vogts *et al*, 1976).
2. Modified chip budding (Rousseau, 1968)

Of these wedge grafting and modified chip budding were the most successful (Brits 1990a). Optimum results were obtained with wedge grafting two budded scions on which individual leaf size was reduced to approximately 0.5 cm^2, during late summer. Scions and graft wounds must be well protected against dessication and fungal infection. Results obtained when applying these techniques have however been disappointingly variable (Ben-Jaacov *et al*, 1989; Malan, 1989), even when grafting plants onto themselves, especially in *Protea*. This suggests that neither a suitable technique nor the conditions required when applying such technique have been established.

A number of research priorities have emerged:

a. More successful grafting techniques need to be developed for the genus *Protea*.
b. *Protea* selections with high phenolic content apparently do not graft easily (Brits, pers. comm.) - this correlation should be studied.
c. Scion/rootstock interspecific compatibility is poorly understood in especially *Banksia* and *Protea*.

Although initial investigations have indicated possible rootstocks (Rousseau *et al*, 1971; Barth *et al*, 1986; Brits, 1990b), no specific promising rootstock candidates have yet been identified. The search therefore continues. Points to consider in rootstock selection include:

a. Broad range of compatibility with scion (Brits, 1990b).
b. Lime tolerance for specific areas (Ben-Jaacov *et al*, 1989).
c. Fast rooting rootstocks (Malan, 1989).
d. *Protea* rootstocks with low phenolic content may increase union and rooting success.
e. *Phytophthora* (McCredie *et al*, 1985b, Brits, 1990b; von Broembson and Brits, 1990) and nematode tolerance (Ben-Jaacov, 1986) in *Leucospermum* and *Banksia*.
f. Crop science evaluation of rootstock effects on yield and cut flower quality.

4. Micropropagation

The most important uses for micropropagated material of protea may be as follows:

a. Rapid multiplication of new selections.
b. Overcoming quarantine requirements for international exchange of material.
c. Overcoming problems experienced with conventional propagation techniques.
d. Rejuvenation of physiologically mature material.
e. *In vitro* physiological and other studies.

Tissue culture techniques for the propagation of *Telopea* (Seeley *et al*, 1986; Offord *et al*, 1990); *Leucospermum* (Ben-Jaacov and Jacobs, 1986; Rugge *et al*, 1989a), *Serruria* (Ben-Jaacov and Jacobs, 1986; Rugge *et al*, 1989b) and *Grevillea* (Gorst, 1978) have been developed. These consist mainly of induced axillary bud sprouting of multi-nodal explants in culture, on modified Murashige and Skoog medium with cytokinin. Important new developments are the "feeder leaf" system for *Leucospermum* resulting in substantially improved axillary bud sprouting (Rugge *et al*, 1989b), establishment of single buds in liquid medium resulting in direct proliferation (Kunisaki, 1990), and the forcing of development of axillary buds of *Protea*, by direct application of benzyladenine to buds either before (Rugge, pers. comm.) or during culture (Tal, pers. comm).

Establishment in culture is followed by proliferation in subcultures. This is done by forcing axillary bud development through altered cytokinin and/or gibberellin and/or agar concentrations and rooting of these axillary shoots *in vitro*, in agar or perlite, or following their removal from culture. All of these techniques are however in great need of streamlining for commercial use. They also need to be extended to other more difficult to propagate cultivars of the same genera.

Techniques have been developed to prevent phenolic browning (Ben-Jaacov, 1986a), which originally constituted the main stumbling block in micropropagation of the genus *Protea*. *Protea repens* (Rugge, pers. comm.), *P. obtusifolia* (Tal, pers. comm.) and *P. cynaroides* (Ben-Jaacov, 1986a) have since been induced to sprout in culture. However further phases of micropropagation still need to be developed. This is an area which needs urgent attention as *Protea* material is probably the most difficult and slowest of all the Proteaceae to propagate.

Callus and proteoid roots (Van Staden *et al*, 1981) and embryos (De Lange, pers. comm.) have been raised from mature cotyledons of *Protea*. Although this is of no immediate benefit to the agricultural industry the techniques are available if required.

5. Reproductive propagation

As was mentioned before, propagation of cut flower species by seed will probably soon be restricted to plant breeders attempting to breed material with improved characteristics. It is likely that seed propagated cultivars will be developed from certain foliage species and from cut flower types such as *Serruria florida* and *Protea cynaroides*. The need to obtain good germination of all fertile seed will therefore be even more important than before, especially in the case of hybrid seed artificially fertilized at great cost.

Two main seed types exist namely: a) nut-like achenes which have an oxygen impermeable testa (Van Staden and Brown, 1973a; 1973b; 1977; Brits, 1986a) and an alternating temperature requirement (Brits, 1986b; 1986d; 1987). This can be satisfied by scarification in concentrated sulphuric acid, soaking in 1% H_2O_2 (Brits and Van Niekerk, 1976), and incubation at 8 °C alternated with 24 °C (Brits *et al*, 1986; Brits 1990). Germination can be improved further by a combined treatment with gibberellic acid and cytokinin following the above germination criteria (Brown and Van Staden, 1986; Brits, 1990c). b) Serotinous achenes have only a low temperature requirement which can be satisfied by incubating seed at approximately 8 °C (Brown and Van Staden, 1975; Mitchell *et al*, 1986; Brits, 1986a; Deall and Brown, 1981).

Improved control is necessary in the following commercial operations:

1. Harvesting of seed in the correct mature stage in serotinous (Van Staden, 1978) and in nut-fruited species (Van Staden and Brown, 1977).
2. Seed disinfection by hot water and fungicide treatment prior to sowing (Benic, 1986; Perry, 1988).

Important areas of future research are:

1. The development of a commercial seed viability test to control the widespread sale of poor quality seed of especially nut-fruited species.
2. Dormancy breaking in some difficult to germinate nut-fruited species e.g. *Orothamnus* and *Mimetes*.
3. The development of a commercial pre-treatment in nut-fruited species to break seed dormancy prior to sale.

Considering the number of problems mentioned above, one realizes that we have a long way to go. These problems can however not all be addressed at once, highlighting the importance of determining research priorities before a programme is initiated.

References

Barth, G.E. and Bennell, M., 1986. Selection and grafting studies of *Banksia coccinea* and *Banksia menziesii*. Proc.Inter. Plant Prop. Soc. 36:220-224.

Benic, L.M., 1986. Pathological problems associated with propagation material in protea nurseries in South Africa. Acta Hort. 185:229-236.

Ben-Jaacov, J., 1986a. Plant propagation by tissue culture and its development for rapid vegetative propagation of proteas. IPA Journal 10:18-19.

Ben-Jaacov, J., 1986b. Protea production in Israel. Acta Hort. 185:101-110

Ben-Jaacov, J. and Jacobs, G., 1986. Establishing *Protea*, *Leucospermum* and *Serruria* in vitro. Acta Hort 185:39-52.

Ben-Jaacov, J., Ackerman, A., Gilad, S. and Shchori, Y., 1989. New approaches to the development of proteaceous plants as floricultural commodities. Acta Hort. 252:193-199.

Blommaert, K.J.L. and Rousseau, G.G., 1971. Watermist propagation of cuttings. FFTRI Information Bulletin No. 24. Department of Agriculture Tech. Services.

Brits, G.J., 1986a. The effect of hydrogen peroxide treatment on germination in Proteaceae species with serotinous and nut like achenes. S. Afr. J. Bot. 52:291-293.

Brits, G.J., 1986b. Influence on fluctuating temperature and H_2O_2 treatment on germination of *Leucospermum cordifolium* and *Serruria florida* (Proteaceae) seeds. S. Afr. J. Bot. 52:286-290.

Brits, G.J., 1986c. The influence of genotype, terminality and auxin formulation on the rooting of *Leucospermum* cuttings. Acta Hort. 185:23-30.

Brits, G.J., 1986d. The effect of air temperature, oxygenating treatments and low storage temperature on seasonal germination response of *Leucospermum cordifolium* (Proteaceae) seeds. S. Afr. J. Bot. 52:207-211.

Brits, G.J., 1987. Germination depth vs. temperature requirements in naturally dispersed seeds of *Leucospermum cordifolium* and *L. cuneiforme* (Proteaceae). S. Afr. J. Bot. 53:119-124.

Brits, G.J., 1988. Sample list of validly published protea cultivar names. Protea News 7:6-10.

Brits, G.J., 1990a. Rootstock production research in *Leucospermum* and *Protea*: I. Techniques. Acta Hort. 264:9-26.

Brits, G.J., 1990b. Rootstock production research in *Leucospermum* and *Protea*: II. Gene sources. Acta Hort. 264:27-40.

Brits, G.J., 1990c. Techniques for maximal germination of six commercial *Leucopsermum* R.Br. species. Acta Hort. 264:53-60.

Brits, G.J., Jacobs, G. and Vogts, M.M., 1983. Domestication of fynbos Proteaceae as a floricultural crop. Bothalia 14(3&4):641-646.

Brits, G.J. and van Niekerk, M.N., 1986. Effects of air temperature, oxygenating treatments and low storage temperature on seasonal germination response of *Leucospermum cordifolium* (Proteaceae) seeds. S. Afr. J. Bot. 52:207-211.

Brown, N.A.C. and van Staden, J., 1975. Inhibitors are not responsible for poor germination of *Protea compacta* seed. J.S.Afr. Bot. 41(4):225-230.

Criley, R.A. and Parvin, P.E., 1979. Promotive effect of auxin, ethephon and daminoside on the rooting *Protea neriifolia* cuttings. J. Amer. Soc. Hort. Sci. 104(5):592-596.

Deall, G.B. and Brown, N.A.C., 1981. Seed germination in *Protea magnifica* Link. S.Afr. J. Sci. 77:175-176.

De Swardt, D.C., 1989. Aspekte van die vegetatiewe en reproduktiewe ontwikkeling by geselekteerde Proteaceae kultivars. (Aspects of the vegetaive and reproductive development of selected Proteaceae cultivars). Unpublished thesis, University of Stellenbosch, Stellenbosch, R.S.A.
Forsberg, L.I., 1988. Protea diseases and their control. Queensland Agric. J. Nov.:347-359.
Gorst, J.R., 1978. Tissue culture propagation of two *Grevillea* hybrids. Comb. Proc. Int. Plant Prop. Soc. 28:435-446.
Gouws, L., Jacobs, G. and Strydom, D.K., 1990. Factors affecting rooting and auxin absorption in stem cuttings of protea. J. Hort. Sci. 65 (1):59-63.
Harrington, B., 1988. Budding or topworking of *Protea longifolia*. IPA Journal 16:15.
Harre, J., 1987. Proteaceae propagation - Pre and post harvest treatments of cuttings. Sappex News 56:19-22.
Harre, J., 1988. Proteas: The propagation and production of Proteaceae. Fisher printing and Stationery, Fielding, New Zealand.
Harre, J., 1989a. Procedures for improving vegetative propagation percentages. Protea News 8:2.
Harre, J., 1989b. Delayed propagation technique. Protea News 8:2-3.
Jacobs, G. and Steenkamp, J.C., 1975. Rooting stem cuttings of *Leucospermum cordifolium* and some of its hybrids under mist. Fmg. S. Afr. flowers and ornamental shrubs. B.7.
Jacobs, G. and Steenkamp, J.C., 1976. Proteas: the rooting of stem cuttings. Fmg. S. Afr. Flowers and Ornamental shrubs B.3.
Kunisaki, J.T., 1990. Micropropagation of *Leucospermum*. Acta Hort. 264:45-48.
Malan, D.G., 1988. Propagation of proteas. Sappex News, Special Edition:11-13
Malan, D.G., 1988. An improved rooting bag for protea cuttings. Sappex News 61:9-10.
Malan, D.G., 1989. Rooting and graft compatability of hybrid *Leucospermum* cuttings. IPA Journal 20:65-66.
McKay, B., 1985. Propagating your own proteas. IPA Journal 5:34-35.
McCredie, T., Dixon, K. and Sivasithamparam, K., 1985a. Grafting banksias to avoid root rot. Austr. Hort., April:75-79.
McCredie, T., Dixon, K. Sivasithamparam, K. and Watkins, P., 1985b. Seeking *Phytophthora* resistant banksias. Austr. Hort. April:48-49.
Mitchell, J.J., Van Staden, J. and Brown, N.A.C., 1986. Germination of *Protea compacta* achenes: the relatioship between incubation temperature and endogenous cytokinin levels. Acta Hort. 185:31-39.
Offord, C.A., Goodwin, P.B. and Nixon, P., 1990. Clonal selection and micropropagation of Waratah. Acta Hort. 264:49-51.
Parvin, P.E., 1979. The art of growing proteas - current recommendations. Protea Workshop Proc.:26-30.
Perry, D., 1988. Succesfully growing proteas. Int. Plant Prop. Soc. Comb. Proc. 112-115.
Perez, J.A.R., 1990. A technique to improve the propagation of stem cuttings of *P. obtusifolia*. Acta Hort. 264:41-43.
Perez, J.A.R. Propagation by leaf-bud cuttings of *Leucadendron* "Safari Sunset", *Leucospermum cordifolium, Leucospermum patersonii* and *Protea obtusifolia*. (This *Acta*).
Rousseau, G.G., 1966. Proteas can be grafted. Fmg. S. Afr. 42(6):53-55.
Rousseau, G.G., 1967. Propagation of proteas from cuttings. Tech. Comm. Dept of Agric. Tech. Services No. 70.
Rousseau, G.G., 1968. Okulering van proteas. Fmg. S. Afr. 43(1):17-19. Rousseau, G.G. and Blommaert, K.L.J., 1971. Protea propagation by grafting and budding. Information Bulletin No. 18, FFTRI, Stellenbosch, RSA.

Rugge, B.A., Jacobs, G. and Theron, K.I., 1989a. Factors affecting bud sprouting in multinodal stem segments of *Leucospermum* cv. Red Sunset in vitro. J. Hortic. Sci. 65(1):55-58.

Rugge, B.A., Van Der Merwe, P., Jacobs, G. and Theron, K.I., 1989b. Light microscopy of somatic embroygenesis from shoots of micropropagated *Serruria florida* (Proteaceae). Electronmicroscopy Soc. S.A. 19:93-94.

Seeley, J.F., Butcher, S.M. and Dennis, D.J., 1986. Micropropagation of *Telopea speciosissima*. Acta Hort. 185:281-285.

Van der Merwe, P., 1985. The genetic relatioship between the South African Proteaceae. Protea News 3:3-5.

Van Staden, J., 1978. Seed viability in *Protea neriifolia*. I. The effects of time of harvesting on seed viability. Agroplantae 10:65-67.

Van Staden, J. and Brown, N.A.C., 1973a. The role of the covering structures in the germination of seed of *Leucospermum cordifolium* (Proteaceae). Aust. J. Bot. 21:189-192.

Van Staden, J. and Brown, N.A.C., 1973b. The effect of oxygen on endogenous cytokinin levels and germination of *Leucadendron daphnoides* seed. Physiol. Plant. 29:108-111.

Van Staden, J. and Brown, N.A.C., 1977. Studies of the germination of South African Proteaceae - a review. Seed Sci. Technol. 5:633-643.

Van Staden, J. and Gilliland, M.G., 1977. The effect of harvesting procedure and storage conditions on the viability of *Protea neriifolia* seed. Proc. Elec. Microsc. Soc. S. Afr. 7:89-90.

Van Staden, J., Choveaux, N.A., Gilliland, M.G., McDonald, D.J. and Davey, J.E., 1981. Tissue culture of South African Proteaceae. 1. Callus and proteoid rootlet formation on cotelydonary explants of *Protea neriifolia*. S. Afr. J. Sci. 77:493-495.

Vogts, M. M., Rousseau, G.G. and Blommaert, K.L.J., 1976. Propagation of proteas. Fmg S. Afr. Flowers and ornamental shrubs B.2.

Von Broembson, S.L. and Brits, G.J., 1990. Evaluation of the resistance of pincushion (*Leucospermum* spp.) breeding lines to root rot caused by *Phytophthora cinnamomi*. Acta Hort. 264:115-121.

PROPAGATION BY LEAF BUD CUTTINGS OF *LEUCADENDRON* 'SAFARI SUNSET', *LEUCOSPERMUM CORDIFOLIUM, LEUCOSPERMUM PATERSONII* AND *PROTEA OBTUSIFOLIA*

J.A. Rodríguez Pérez
Departamento de Producción Agraria
Escuela Universitaria de Ingeniería T. Agrícola
38200 La Laguna, Tenerife
Spain

Abstract

Leucadendron 'Safari Sunset', *Leucospermum patersonii* and *Protea obtusifolia* can be propagated on propagating beds provided with bottom heat (25 ± 2°C) and a mist system, using leaf bud cuttings, put under different treatments with IBA (0, 2 000, 4 000 and 8 000 ppm) and obtaining maximum percentages of transplantable cuttings of 20%, 31.25% and 10%, respectively. Although some *Leucospermum cordifolium* cuttings produced roots, they were not able to develop shoots.

1. Introduction

Some species like the black raspberry (*Rubus occidentalis*), the lemon tree (*Citrus limon*), the camellia (*Camellia* spp.) and the azalea (*Rhododendron* spp.) can be easily propagated by leaf bud cuttings. This type of cutting consists of a complete leaf (blade and petiole) and a little portion of stem supporting the corresponding axillary bud (Hartmann and Kester, 1974).

The propagation of proteas is normally carried out by seed or by stem cuttings (Meynhardt, 1974; Jacobs and Steenkamp, 1975 and 1976; Vogts, 1982; Jacobs, 1983; George, 1984).

However, Ellyard and Butler (1985) managed to propagate *Telopea speciosissima* x *T. mongeansis* using leaf bud cuttings, obtaining the best results with IBA treatments at a concentration of 2 000 ppm. At 9 weeks they reached 90% rooting and at 22 weeks 100%. The rooting medium was a mixture of sand, peat and perlite (1:1:1) and the process was carried out under a mist system and bottom heat at 30°C.

With the purpose of observing if this kind of propagation could be applied to *Leucadendron* 'Safari Sunset', *Leucospermum cordifolium, L. patersonii* and *Protea obtusifolia*, proteas of commercial interest for the island of Tenerife, the following experiments were carried out.

2. Material and methods

2.1 General aspects

The cuttings were prepared from terminal shoot sections, from semi-hardwood. These shoots were divided in pieces, each of them with a leaf and its corresponding well developed axillary bud. Then the piece of stem was cut longitudinally down the middle, obtaining cuttings which consisted of a leaf, axillary bud and a section of stem.

Once the cuttings were prepared, the stem sections were dipped in the experimental hormonal solution for 5 seconds, then they were dipped into talc containing benomyl and captan, both at 5% of active material.

The cuttings were planted, inserting the piece of stem shallowly in a mixture of polystyrene grains and peat moss (1:1 in volume). Rooting was done in plastic propagating trays, which were put on a rooting bed with bottom heat (25 ± 2°C) and a mist system with misting nozzles at 50 l/h, for 36 seconds every half hour, between 9:00 and 17:00 in spring-summer and between 8:00 and 16:00 in autumn-winter. They were situated in a well ventilated greenhouse, with a polyethylene roof and plastic nettings walls, at a 60% reduction of natural light.

A randomized block design was used with four treatments and four replications. The number of cuttings was 10 or 20 for each treatment. The total number of cuttings was 160 and 320 for each species or cultivar.

The treatments used were the following:
Treatment A: ethyl alcohol at 50% (control)
Treatment B: 2 000 ppm of IBA in ethyl alcohol at 50%
Treatment C: 4 000 ppm of IBA in ethyl alcohol at 50%
Treatment D: 8 000 ppm of IBA in ethyl alcohol at 50%

The cuttings were sprayed weekly with a mixture of benomyl, mancozeb, iprodione, metomilo or diazinon to control pests and diseases.

Every 4 weeks, at 8 or 12 weeks from the beginning of the experiment, cuttings were scored according to the following scale:
a = dead cuttings
b = cuttings without callus
c = cuttings with callus
d = rooted cuttings
e = cuttings with roots and shoots, but not transplantable
f = cuttings with shoots, but no roots
g = cuttings transplantable

The results as percentage of transplantable cuttings at the end of the experiment, were subjected to analysis of variance, using the arcsin transformation.

2.2 *Leucospermum cordifolium*

The cuttings were taken from two *Leucospermum cordifolium* plants of about 4 years old. They were from seed imported from South Africa, cultivated in experimental plots in the Acclimatization Garden of La Orotava (Puerto de la Cruz). The number of cuttings per treatment was 20. The total number of cuttings was 320.

Data were taken at 12, 16, 20, 24, 28, 32, 36 and 40 weeks from the beginning of the experiment. The trial started on 13 July 1988 and concluded on 21 April 1989.

2.3 *Protea obtusifolia*

The cuttings were taken from a single *Protea obtusifolia* plant, about 5 years old, cultivated in the experimental plots in the Acclimatization Garden of La Orotava. The number of cuttings per treatment was 10. The total number of cuttings was 160.

Data were taken at 12, 16, 20, 24, 28, 32, 36 and 40 weeks from the beginning of the experiment. The trial started on 28 July 1988 and concluded on 4 May 1989.

2.4 *Leucospermum patersonii*

The cuttings were taken from four *Leucospermum patersonii* plants. They were from vegetative material imported from New Zealand, and were cultivated in experimental plots from the Acclimatization Garden of La Orotava. The number of cuttings per treatment was 20. The total number of cuttings was 320.

Data were taken at 8, 12, 16, 20, 24, 28 and 32 weeks from the beginning of the experiment. The trial started on 29 September 1988 and concluded on 4 May 1989.

2.5 *Leucadendron* 'Safari Sunset'

The cuttings were taken from seven plants of *Leucadendron* 'Safari Sunset'. These were from vegetative material imported from New Zealand, cultivated in the Acclimatization Garden nursery in La Orotava. The number of cuttings per treatment was 10. The total number of cuttings was 160.

Data were taken at 8, 12, 16, 20, 24, 28, 32 and 36 weeks from the beginning of the experiment. The trial started on 13 October 1988 and finished on 22 June 1989.

3. Results and discussion

3.1 *Leucospermum cordifolium*

The development of the cuttings throughout the experiment can be seen in figure 1. The use of hormones slowed down the formation of callus on the cuttings, because at 12 weeks treatment A (control) showed 95% of cuttings with callus, whilst treatment D (8 000 ppm of IBA) only showed 55%. The other two treatments had inferior values. However, the formation of roots was favoured by the use of IBA, because at 12 weeks treatment D had already shown 3.75% of cuttings with roots, whilst in treatment A no cuttings rooted and this kind of cuttings only appeared at 28 weeks.

Cuttings that did not receive hormonal treatment were able to form shoots at a low percentage (2.5%) but the ones treated with IBA were not able to form them. This might be due to the opposing interaction of IBA with cytokinins which take part in shoot formation.

Throughout the experiment the percentage of dead cuttings of the treatments with IBA increased continuously, exceeding 80% at the end of the experiment, whilst in the control treatment the final value was 31.25%.

A problem experienced was that a layer of algae formed after 4 months. At first it was a slight layer, but finally it covered both the surface of the rooting medium, reducing its aeration, and part of the base of the cuttings as well.

3.2 *Protea obtusifolia*

The development of cuttings can be seen in figure 2. Contrary to *Leucospermum cordifolium*, the use of IBA accelerated callus formation at a high concentration, as at 12 weeks treatment A (control) showed 50% of cuttings with callus, whilst in treatment D (8 000 ppm of IBA) this value was 70%. Also it favoured root formation, because treatment A showed 10% rooting as opposed to 27.50% of treatment D.

Treatments A and B resulted in 7.50% and a 10% of cuttings with roots and

shoots, respectively, at 28 weeks. At 32 weeks the first cuttings of these treatments were transplanted. At 36 weeks the first cuttings of treatment D were transplanted.

At 40 weeks, treatment B showed a higher percentage of transplantable cuttings (10%), whilst treatments A and D gave the same value (7.50%). If the sum of rooted cuttings and of rooted cuttings with shoots is considered, treatment D showed the highest value (40%), as opposed to treatment A (15%), treatment B (12.50%) and treatment C (22.50%). However, the analysis of variance gave a value of F = 2.220, not significant at the level of 5%. Thus there were no significant differences among treatments.

There were problems with algae which developed on the surface of the rooting medium as in *Leucospermum cordifolium*. These reduced aeration to the substrate and covered the base of the cuttings as well.

Cuttings clearly suffered much during transplantation, therefore, when using this technique of propagation, it is recommended to root cuttings individually in plastic bags.

3.3 *Leucospermum patersonii*

The development of cuttings is presented in figure 3. Analysis of the results showed that the treatment with IBA accelerated root formation, because at 8 weeks the percentage of rooted cuttings was 40% in treatment D (8 000 ppm of IBA), whilst in treatment A (control) it was only 26,25%. However, also at 8 weeks, treatment A showed 21.25% of cuttings with shoots but without roots, whilst in hormonal treatments, only B showed 1.25% of cuttings of this kind. Possibly the IBA inhibited the cytokinins which participate in the formation of shoots. Throughout the experiment it was observed that treatment A always showed a higher percentage of cuttings with shoots but without roots, as opposed to treatments in which IBA was used, which showed a higher percentage of rooted cuttings.

At 16 weeks the first cuttings of treatments A and B were transplanted and those of treatment C at 20 weeks and of treatment D at 24 weeks. However, at 28 weeks the percentages of the cuttings which could be transplanted of treatments A and D were very close (25% and 22.5%, respectively). The same happened at 32 weeks, but treatment D already showed the higher value, 31.25%, compared with 30% of treatment A. The analysis of variance showed a value of F = 0.803, not significant at the 5% level, that is, there were no significant differences among the treatments.

The cuttings were transplanted to pots without any difficulty, containing a medium of sifted peat moss and lapilli (1:3 v/v) which were put on a propagation bed provided with bottom heat and mist until the plants were established (about 1 month). They were then taken out of the bed and left in the greenhouse for 1 or 2 months, and finally exposed to full sun. The percentage of transplanting success was 100%.

3.4 *Leucadendron* 'Safari Sunsent'

In figure 4 the development of the cuttings can be seen. The use of IBA accelerated both the formation of callus and roots, because at 8 weeks the percentage of rooted cuttings was 12.5%, 20% and 22.5% for hormonal treatments B, C and D, respectively, compared with 0% for treatment A (control). However, this last treatment gave 10% of cuttings with shoots but without roots. Throughout

the experiment the percentage of rooted cuttings increased in treatments B, C and D, so that at 20 weeks it was 85% in treatment D as opposed to 2.5% in treatment A.

At the end of the experiment the percentage of the cuttings which could be transplanted was 20% for treatment B (2 000 ppm of IBA) and 2,5 % for treatment A. If the sum of the corresponding values of transplantable cuttings and cuttings with roots and shoots, but not transplantable, is considered, treatment B showed the highest value (25%), following treatment C with 10%. Interestingly treatment D (8 000 ppm IBA) gave the highest value of rooted cuttings (85%) during the experiment, but at the end it resulted in the highest percentage of dead cuttings (22.5%), producing only 2.5% of rooted cuttings with shoots, but not transplantable. This might be due to the opposing interaction of IBA with the cytokinins which take part in shoot formation.

The analysis of variance gave F = 2.877, not significant at the level of 5%. Thus there were no significant differences between the treatments.

The transplantation of the cuttings was carried out without difficulty as in the case of *Leucospermum patersonii*.

4. Conclusions

Leucadendron 'Safari Sunset', *Leucospermum patersonii* and *Protea obtusifolia* could be propagated by leaf bud cuttings, but the process was slow and a low yield was found. In *Leucadendron* 'Safari Sunset' 36 weeks were needed to obtain 20% of transplantable cuttings; in *Leucospermum patersonii*, which rooted easiest, 32 weeks were needed to obtain 32.25% of transplantable cuttings; and in *P. obtusifolia*, 40 weeks to obtain 10% of transplantable cuttings(figures 5, 6). In *Leucospermum cordifolium*, though some cuttings produced roots (5%), cuttings were not able to develop shoots and no transplantable cuttings were obtained.

In conclusion this technique of propagation can only be recommended presently for multiplication of new cultivars or clones of *Leucospermum patersonii* of which there is little vegetative material available.

Clearly, however, it is necessary to continue investigating this promising technique with the purpose of accelerating the propagation process and obtaining a higher cutting yield, by means of modifying some of the factors which play a role: pH, humidity, temperature of the rooting medium, levels of IBA, etc.

5. Acknowledgements

I want to thank Ms. Isabel La Roche for her thorough job as a translator and language adviser when translating this article into English; Dr Juan-Felipe Pérez Francés for his co-operation and assistance in drawing the figures.

References

Ellyard, R.K. and Butler, G., 1985. Breakthrough in waratah propagation. Australian Horticulture, Marzo: 27-28.

George, A.S., 1984. The *Banksia* book. Kangaroo Press, Kenthurst, Australia: 19-22.

Hartmann, H.T. and Kester, D.E., 1974. Propagación de plantas. Compañía Editorial Continental, S.A., México, D.F.: 352-353.

Jacobs, G., 1983. Vegetative propagation of proteas - recent developments. In Growing and Marketing of Proteas Vol. 1. Proteaflora Enterprises, Melbourne, Australia: 7-16.

Jacobs, G. and Steenkamp, J.C., 1975. Proteas: the rooting of stem cuttings. Farming in South Africa, Series: Flowers, Ornamental Shrubs and Trees, B.3. Department of Agricultural Technical Services, Pretoria.

Jacobs, G. and Steenkamp J.C., 1976. Rooting stem cuttings of *Leucospermum cordifolium* and some of its hybrids under mist. Farming in South Africa, Series: Flowers, Ornamental Shrubs and Trees, B.7. Department of Agricultural Technical Services, Pretoria.

Meynhardt, J.T., 1974. Propagation of proteas. Farming in South Africa, Series: Flowers, Ornamental Shrubs and Trees, B.2. Department of Agricultural Technical Services, Pretoria.

Vogts, M.M., 1982. South Africa's Proteaceae, know them and grow them. C. Struik (Pty) Ltd., Cape Town: 220-223.

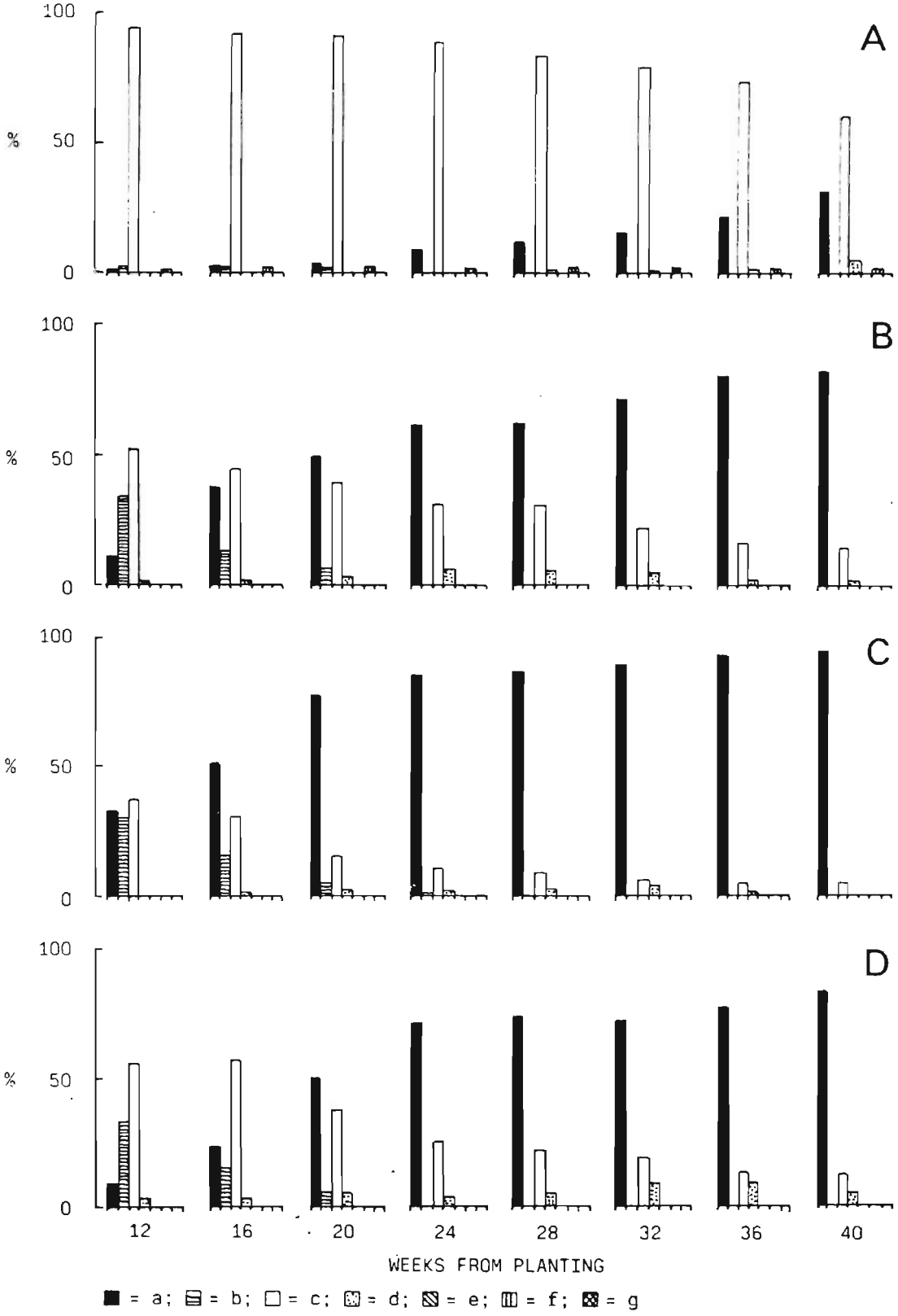


Figure 1 - Effect of IBA on rooting of *Leucospermum cordifolium* leaf bud cuttings. A = control; B, C and D = 2 000, 4 000 and 8 000 ppm ofIBA, respectively. a = dead; b = without callus; c = with callus; d = rooted; e = with roots and shoots, but not transplantable; f = with shoots, but not roots; g = transplantable

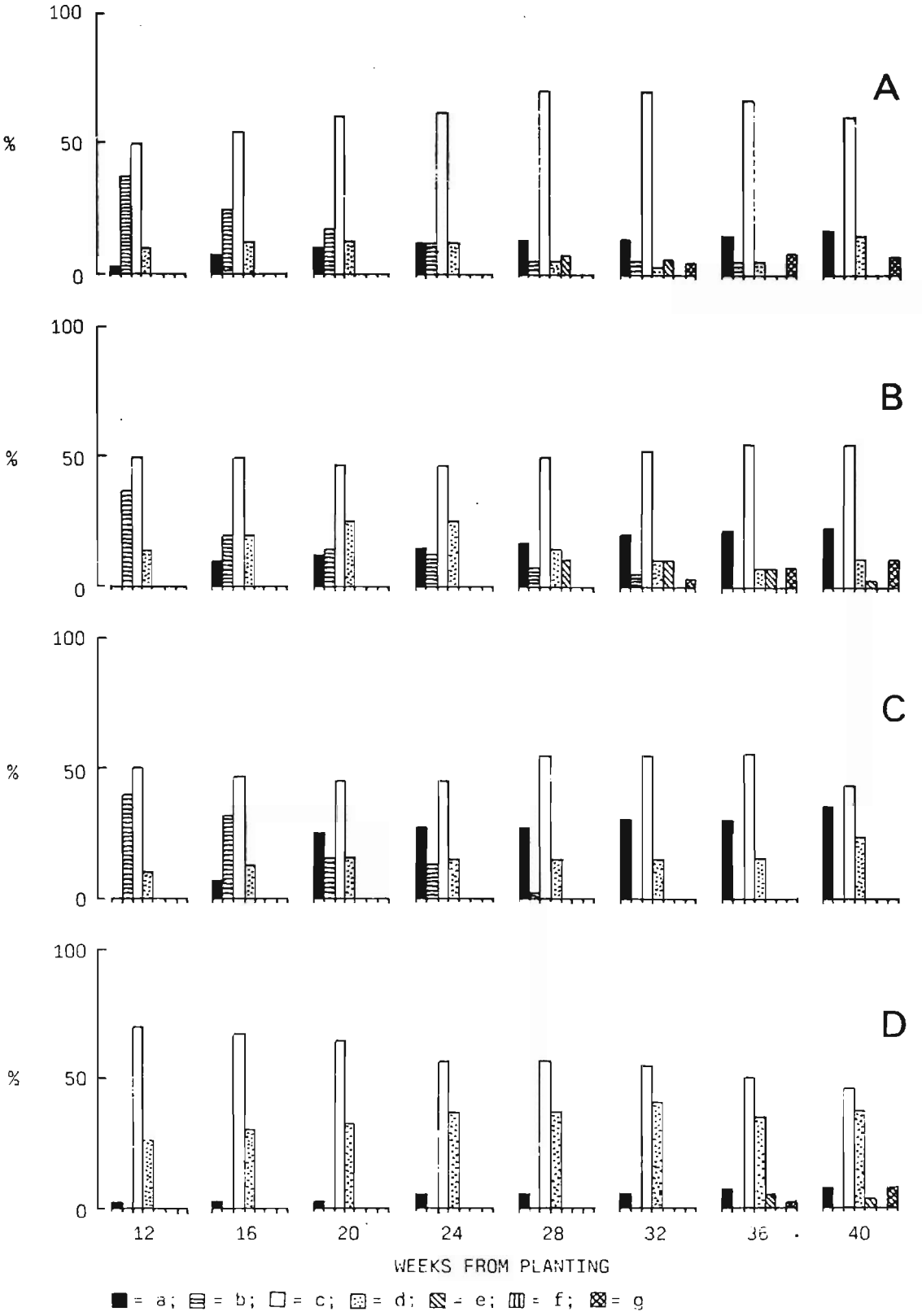


Figure 2 - Effect of IBA on rooting of *Protea obtusifolia* leaf bud cuttings. A = control; B, C and D = 2 000, 4 000 and 8 000 ppm of IBA, respectively. a = dead; b = without callus; c = with callus; d = rooted; e = with roots and shoots, but not transplantable; f = with shoots, but not roots; g = transplantable

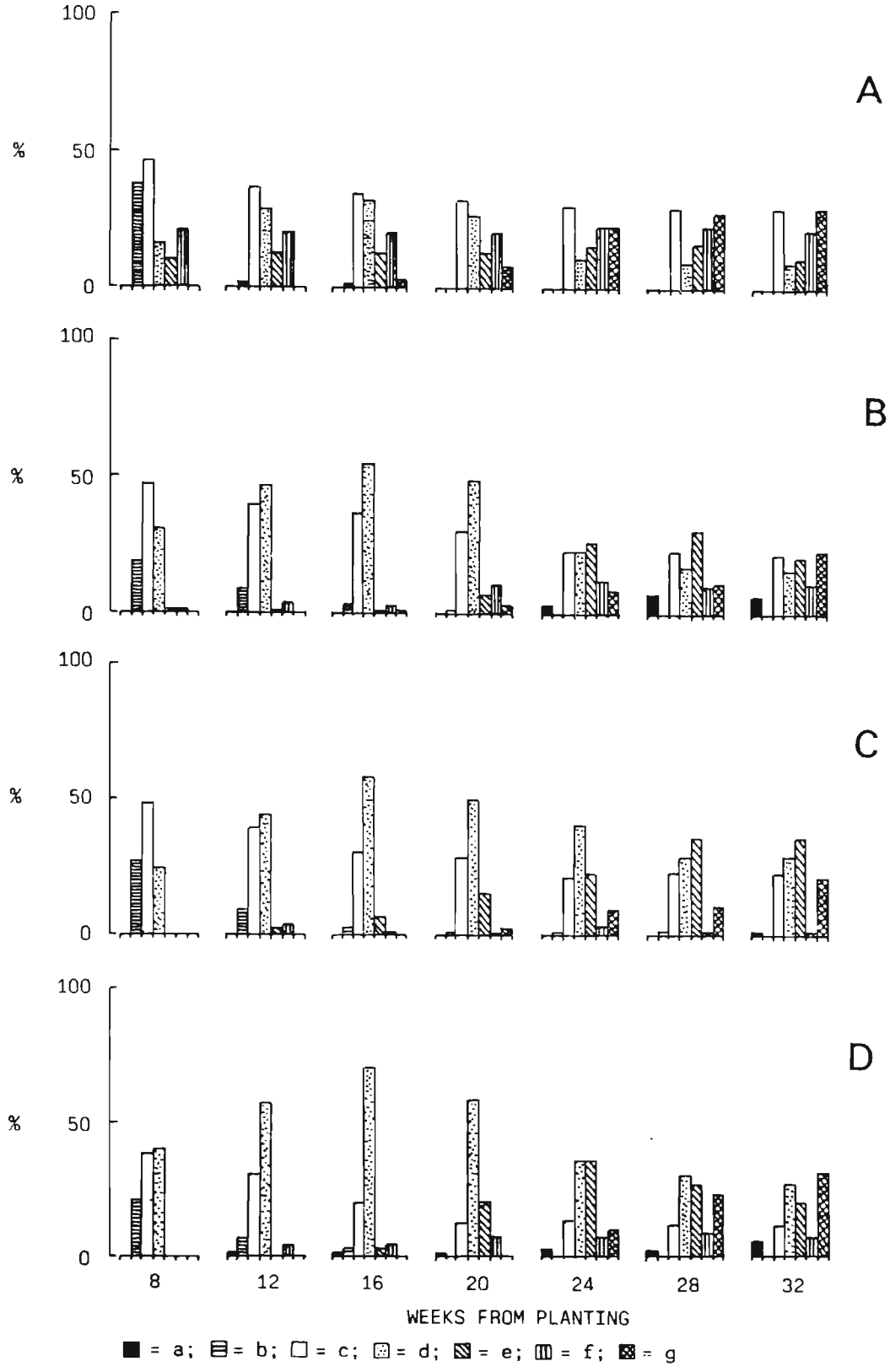


Figure 3 - Effect of IBA on rooting of *Leucospermum patersonii* leaf bud cuttings. A = control; B, C and D = 2 000, 4 000 and 8 000 ppm of IBA, respectively. a = dead; b = without callus; c = with callus; d = rooted; e = with roots and shoots, but not transplantable; f = with shoots, but not roots; g = transplantable

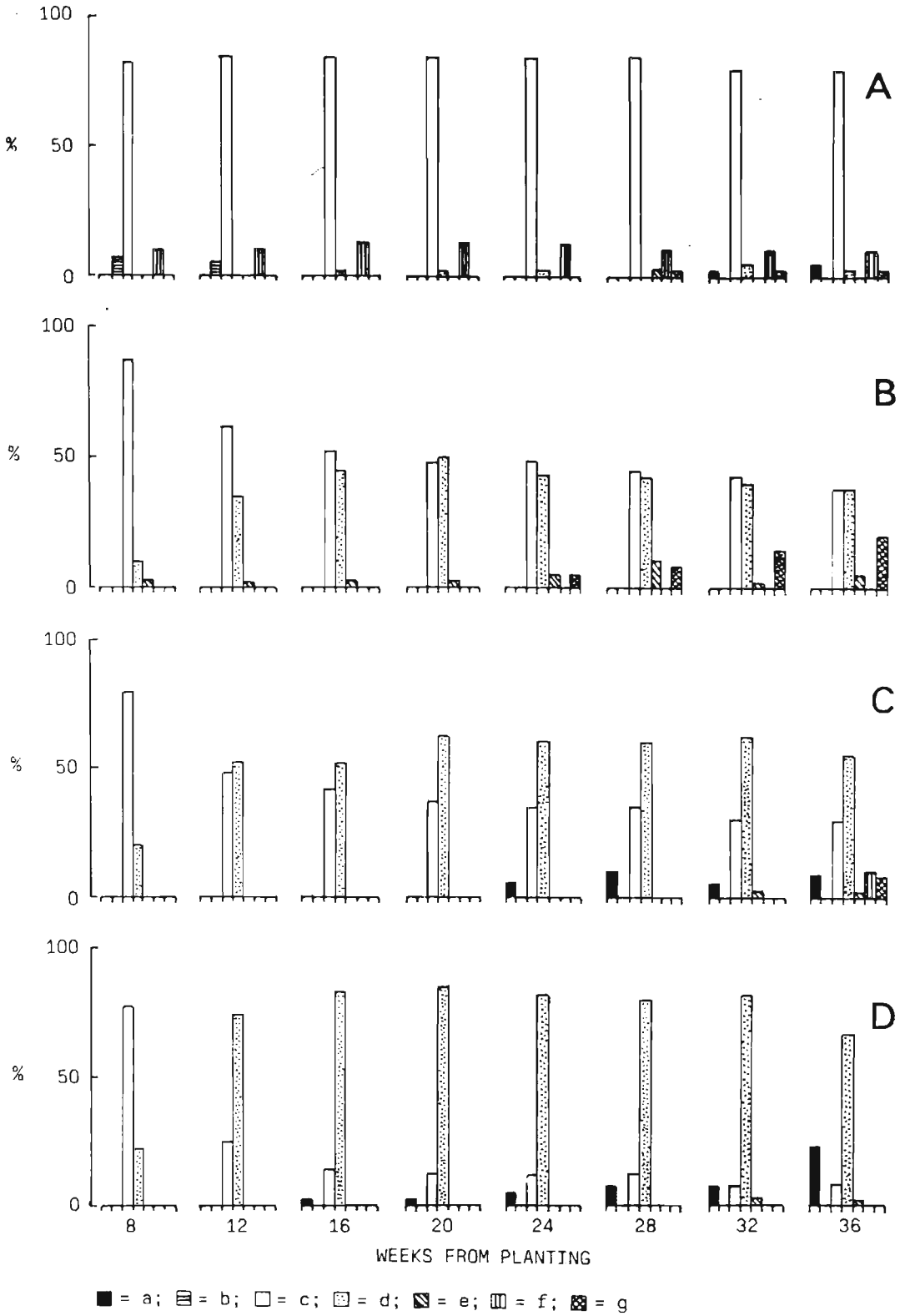


Figure 4 - Effect of IBA on rooting of *Leucospermum* 'Safari Sunset' leaf bud cuttings. A = control; B, C and D = 2 000, 4 000 and 8 000 ppm of IBA, respectively. a = dead; b = without callus; c = with callus; d = rooted; e = with roots and shoots, but not transplantable; f = with shoots, but not roots; g = transplantable

Figure 5 - Transplantable leaf bud cuttings of *Protea obtusifolia*

Figure 6 - Transplantable leaf bud cuttings of *Leucospermum patersonii*

PROPAGATION OF *PHAENOCOMA PROLIFERA* IN *IN VITRO* CULTURE

G. Kaddar, A.A. Watad, T. Ben-Noon, J. Ben-Jaacov
Dept. of Ornamental Horticulture
A.R.O. The Volcani Center
P.O. Box 6, Bet Dagan 50-250
Israel

Abstract

Phaenocoma prolifera of the *Asteraceae* family is an evergreen shrub, widely distributed along the coast of the south western Cape of South Africa. A micropropagation method was developed in order to propagate special plant forms with high potential as flowering pot plants. The addition of growth regulators to Murashige and Skoog's medium at the initial stage in culture has a negative effect on shoot development, and much callus developed. However, after a few subcultures, addition of 1 $mg.l^{-1}$ 6-benzyl-aminopurine (BAP) promoted shoot growth and proliferation. Rooting of the *in vitro* mini-cuttings was achieved by 1 $mg.l^{-1}$ 1-naphthalene acetic acid (NAA).

1. Introduction

Phaenocoma prolifera of the *Asteraceae* is an evergreen shrub which has a widespread distribution along the coast of the southwestern Cape of South Africa (Pienaar, 1984). The flowers remain attractive for a very long time, and serve as good cut flowers and can also be dried for use in arrangements. We are interested in testing its potential as a flowering pot plant. Usually propagation is done by seeds but special selections should be propagated vegetatively. Propagation by cuttings is difficult and slow, and therefore *in vitro* propagation was tried.

2. Materials and Methods

Explants were obtained from mother plants growing in pots kept under a shade net. Shoot segments were cut into 5-10 cm long pieces and rinsed under running tap water for 1 h, and then surface sterilized with 1% sodium hypochlorite, with the addition of 0.1% Tween-20 for 20 min. After three successive rinses in sterile distilled water, the explants were cultured on autoclaved media. The explants were cultured in different media: basal medium with the addition or not of 6-benzyl-aminopurine (BAP) and kinetin in concentrations varying from 0 to 2.0 $mg.l^{-1}$, for shoot proliferation; and 1 $mg.l^{-1}$ 1-naphthalene acetic acid (NAA) for rooting.

The basal medium contained MS salts (Murashige and Skoog, 1962) and vitamins, 3% sucrose and 9 $g.l^{-1}$ agar. The medium was adjusted to pH 5.5 before autoclaving. Cultures were kept at 25 °C and illuminated for 16 h at 4000 lux with cool white fluorescent tubes.

3. Results and Discussion

3.1 Effects of growth regulators on shoot establishment proliferation

Best shoot proliferation was obtained when explants from mother plant stock were planted in culture medium in early spring before summer sprouting. The minimum size of explants for introduction into culture was found to be 10 mm. The addition of growth regulators to the basal medium at the initial stage of propagation (the first three monthly subcultures) had negative effects: the

meristematic edges dried out and much callus developed. At late stages, addition of 1.0 mg.l^{-1} BAP led to the best shoot establishment and proliferation (figure 1). The established explants in culture were maintained for three monthly subcultures in basal medium without hormones (figure 2) before the addition of BAP, or NAA. Browning and blackening of the explants *in vitro* and of the growth medium, due possibly to hydroxy-phenol compounds (George and Sherrington, 1984), was reduced by the frequent subculturing (three) into fresh basal medium (every 2 days).

3.2 Rooting

Subculturing of explants with proliferated shoots in basal medium containing 1.0 mg.l^{-1} NAA, caused roots to develop within 30 days (figure 2).

References

George, E.F. and Sherrington, P.D., 1984. Plant propagation by tissue culture. Handbook and Directory of Commercial Laboratories. Exegetics Ltd., Eversley, Basingstoke, Hants RG27 0QY, England.

Murashige, T. and Skoog, F., 1962. A revised medium for rapid growth and bioassay with tobacco tissue culture. Physiol. Plant. 15: 473-497.

Pienaar, K., 1984. The South African, What flower is that? C. Struik Publishers, Cape Town, South Africa.

Figure 1 - Shoot proliferation of *Phaenocoma prolifera* after one month on basal medium plus 1.0 mg.l^{-1} BAP. (Subsequently it was kept on basal medium plus 1.0 mg.l^{-1} NAA or rooting).

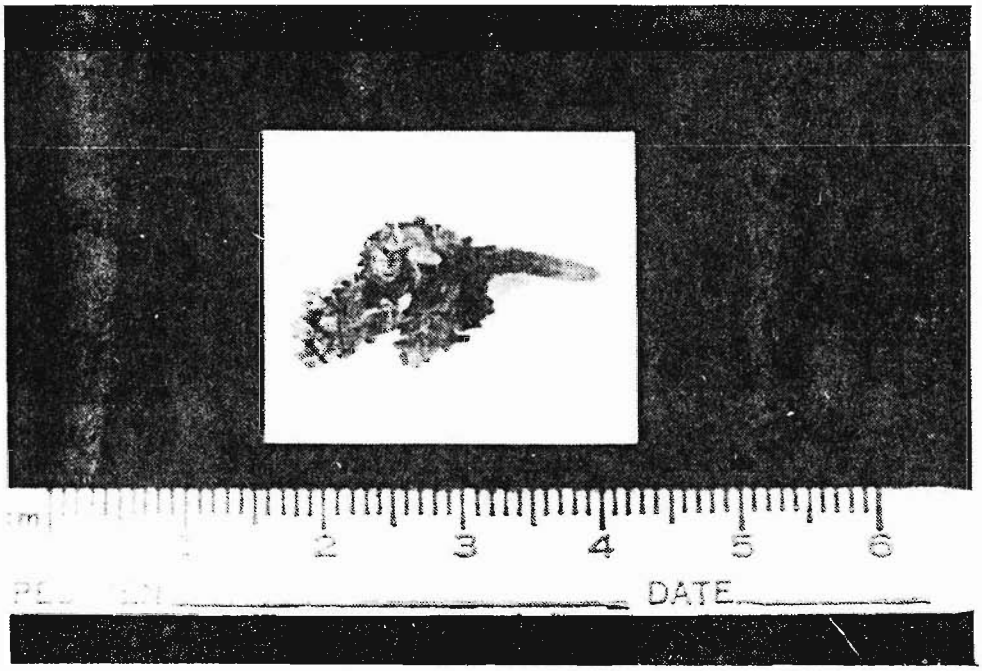
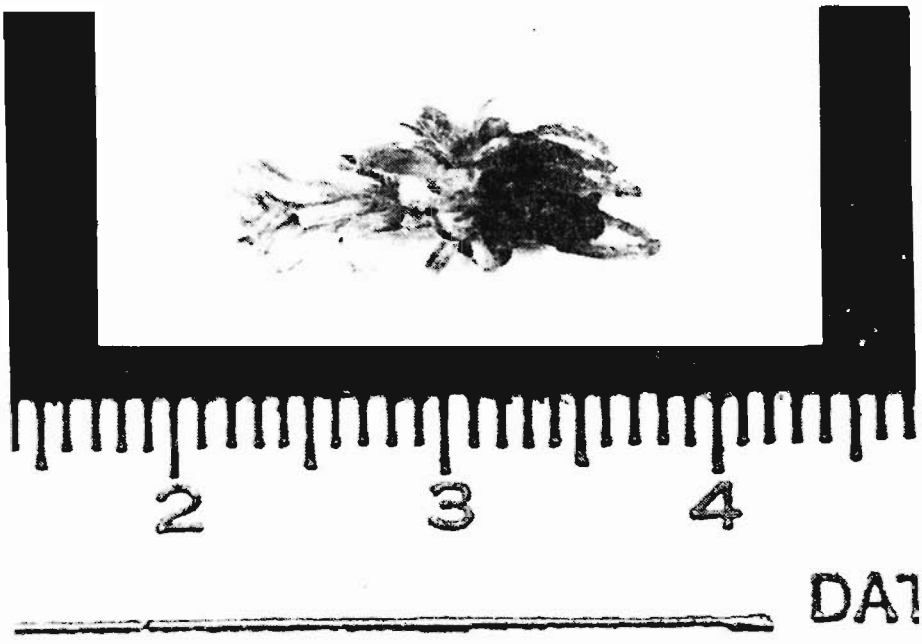


Figure 2 - *In vitro* shoot proliferation of *Phaenocoma prolifera* after three subcultures on basal medium, before subculturing on basal medium containing 1 mg.l^{-1} NAA.



IN VITRO PROPAGATION OF *GREVILLEA* SPECIES

A.A Watad, J. Ben-Jaacov
Dept. of Ornamental Horticulture
A.R.O. The Volcani Center
P.O. Box 6, Bet Dagan 50-250
Israel

E. Tal, H. Solomon
ARO N. R&D Project
Misgav Exp. Station
M.P. Misgav 20-179
Israel

Abstract

Grevillea species are in different stages of development as flowering pot plants. *In vitro* propagation technique was used in order to achieve large scale production of six species and cultivars of *Grevillea*: *G.* 'Roundo', *G.* 'Robyn' Gordon', *G. pinaster*, *G. crithmifolia*, *G. petrophiloides* and *G.* 'Robyn Hood'. The optimal 6-benzyl-aminopurine (BAP) level for shoot proliferation of two-nodal segments was 1 $mg.l^{-1}$. Average propagation factor of all these *Grevillea* was 2.5 - 6.2 new axillary shoots in two months. The highest multiplication rate of 6.2 was in the case of *G. crithmifolia* as compared with only 2.5 in the case of *G. petrophiloides*. The highest percentage of *in vitro* rooting was achieved by 1 $mg.l^{-1}$ NAA.

1. Introduction

A micropropagation project is carried out for introducing and developing different products as new woody flowering pot plants and cut flowers. The work has concentrated on different Proteaceae species originating in Australia and South Africa. The importance of this approach in Israel has been discussed by Ben-Jaacov *et al* (1989). *In vitro* propagation of Proteaceae is important because of several reasons, mainly because it might provide procedures for introduction and for breeding programmes, for new crops. Also because of propagation difficulties by classical methods in the case of some species.

Grevillea is a diverse Australian genus belonging to the Proteaceae and consists of some 270 known species and hundreds of selected hybrids and clones (Burke, 1983). In a previous report (Ben-Jaacov and Dax, 1981), a method of *in vitro* propagation of *Grevillea rosmarinifolia* was described. Tissue culture propagation of two *Grevillea* hybrids was also described by Gorst *et al*, (1978). Results presented here is a summary of a study which focused on developing *in vitro* methods of propagation for six species and cultivars of *Grevillea*.

2. Materials and Methods

Aseptic cultures were started during the summer growth flush from actively growing shoots of six *Grevillea* species, and cultivars: *G.* 'Roundo', *G.* 'Robyn Gordon', *G. pinaster*, *G. crithmifolia*, *G. petrophiloides* and *G.* 'Robyn Hood'. Shoot segments were surface sterilized for 20 min in a solution containing 2% sodium hypochlorite and 0.1% Tween-20 and then rinsed three times in sterile distilled water. Each shoot was sectioned into two-nodal segments and cultured in 25x80 mm glass tubes containing 10 ml of autoclaved medium.

The growth medium contained half strength of the revised Murashige and Skoog salts and vitamins (Murashige and Skoog, 1962) supplemented with 30 $g.l^{-1}$ sucrose, 9 $g.l^{-1}$ Bactoagar (difco) and 1.0 $mg.l^{-1}$ of 6-benzyl-aminopurine (BAP). Rooting was tested with three different auxins: indole-3-acetic acid (IAA), indole-3-butyric acid (IBA) and naphthalene acetic acid (NAA). The pH of the medium was adjusted to 5.7 before autoclaving.

3. Results and Discussion

3.1 Shoot proliferation

The six *Grevillea* species and cultivars were successfully established and rooted in culture. After seven monthly subcultures on MS medium at half strength containing 1 $mg.l^{-1}$ BAP, shoot proliferation was recorded. The shoot multiplication rate was calculated from the average of new axillary shoots during four two-monthly subcultures of two-nodal segments in shoot proliferation medium. The lowest number of new axillary shoots was obtained in the case of *G. petrophiloides*, while the other five species gave higher shoot proliferation (table 1).

The highest number of new axillary shoots was produced by *G. crithmifolia*. When the net two-nodal segments (yield) per tube, which is the real multiplication rate for routine commercial large scale production, was recorded (table 1), larger differences between the six species and cultivars were obtained: the yield in the case of *G. crithmifolia* and *G. pinaster* was 3-4 times that of *G. petrophiloides* while similar results were given by *G.* 'Robyn Hood', *G.* 'Robyn Gordon' and *G.* 'Roundo'.

3.2 *In vitro* rooting

The effect of different auxins on *in vitro* rooting was tested (table 2). After one month in hormone-free medium only 15-38% of shoots from the different species and cultivars developed roots. The highest rooting percentage was obtained in the presence of 1 $mg.l^{-1}$ NAA, also more branched roots per explant were observed. Differences in rooting between species and cultivars were observed. *G.* 'Roundo' was rooted easily, *G. petrophiloides* rooting was low in all three different tested auxins.

In conclusion, results presented here show that six *Grevillea* species and cultivars could be propagated efficiently by tissue culture. Plants from all tested species and cultivars were successfully established in the field after acclimatization.

References

Ben-Jaacov, J. and Dax, E., 1981. *In vitro* propagation of *Grevillea rosmarinifolia*. HortScience 16: 309-310.

Ben-Jaacov, J., Ackerman, A., Gilad, S. and Shchori, Y., 1989. New approaches to the development of proteaceous plants as floricultural commoditites. Acta Horticulturae 252: 193-199.

Burke, D., 1983. Growing Grevilleas in Australia and new Zealand. Kangaroo press, Kenthurust 2154, Australia.

Gorst, J.R., *et al.*, 1978. Tissue culture propagation of two *Grevillea* hybrids. Comp. Proc. Int. Plant Prop. Soc. 28: 435-446.

Murashige, T. and Skoog, F., 1962. A revised medium for rapid growth and biosassays with tobacco tissue cultures. Physiologia plantarum 15: 473-497.

Table 1 - Shoot multiplication rate* of six *Grevillea* cultivars, when two-nodal segments were grown on half MS medium containing 1 $mg.l^{-1}$ BAP (after two months).

Species or Cultivars	No. of new axillary shoots	Total two nodal segments per tube
G. 'Roundo'	4.6 ± 0.8	11.4±1.7
G. 'Robyn Gordon'	4.3 ± 0.6	10.8±1.9
G. pinaster	5.3 ± 1.1	20.0±2.8
G. crithmifolia	6.2 ± 1.5	24.0±3.8
G. petrophiloides	2.5 ± 0.8	6.1±1.4
G. 'Robyn Hood'	3.8 ± 1.2	11.0±2.1

* Multiplication rate is an average value of two-monthly sub-cultures, 20 tubes each.

Table 2 - Effect of different auxins, IAA, IBA, NAA at 1 $mg.l^{-1}$ on shoot rooting percentage of *Grevillea* 'Roundo', *G.* 'Robyn Gordon', *G. pinaster*, *G. crithmifolia*, *G. petrophiloides* and *G.* 'Robyn Hood' after one month. Control: without hormones.

Cultivar	Rooting per cent			
	Control	IAA	IBA	NAA
G. 'Roundo'	38	61	92	90
G. 'Robyn Gordon'	25	NT*	NT	87
G. pinaster	20	NT	NT	85
G. crithmifolia	15	50	75	83
G. petrophiloides	-	5	12	30
G. 'Robyn Hood'	30	20	27	85

* Not tested

MICROPROPAGATION OF SELECTED *LEUCOSPERMUM CORDIFOLIUM*: EFFECT OF ANTIBIOTICS AND GA_3

E. Tal, H. Solomon
A.R.O. Northern R& D Project
Misgav Experimental Station
M.P. Misgav 20-179
Israel

J. Ben-Jaacov, A.A. Watad
Dept. of Ornamental Horticulture
A.R.O. The Volcani Center
P.O. Box 6, Bet Dagan 50-250
Israel

Abstract

The establishment of a dwarf clone of *Leucospermum cordifolium* in culture was difficult because of heavy endogenous bacterial contamination. Two-nodal segments were placed on half strength Murashige and Skoog medium supplemented with 6-benzyl-aminopurine to which the antibiotics streptomycin and rifampicin were added. The addition of 100 $mg.l^{-1}$ rifampicin to the growth medium was most effective in suppressing the bacteria and enabling the growth of the *Leucospermum*. GA_3 at 1-2 $mg.l^{-1}$ was found to be essential for rapid shoot proliferation and elongation. Shoots multiplied at the rate of approximately five new shoots within 8 weeks. The best rooting was achieved when 1 $mg.l^{-1}$ indole butyric acid (IBA) was added to the medium. Relatively high light intensities promoted *in vitro* rooting. Following rooting, plants were successfully transferred to outdoor conditions.

1. Introduction

The flowering pot plant industry requires a high level of success and reliability with propagation material. Species of the genus *Leucospermum* are not considered difficult to root in South Africa (Jacobs and Steenkamp, 1975), but results in Israel vary (mainly seasonally, but some with no clear pattern or explanation), from 50% to 90%.

Micropropagation has many advantages and disadvantages (George and Sherrington, 1984). Its main potential value in the development of *Leucospermum* for the flowering pot plant industry lies in the reliability in supply of propagation material and its adjustability to the growth cycle in pots.

2. Material and Methods

Two-nodal segments of selected *Leucospermum cordifolium*, selected as a potential pot plant, were sterilized with 2% sodium hypochlorite followed by three washes in sterilized distilled water. The segments were placed on half-strength Murashige and Skoog medium to which was added 3% sucrose and 0.9% agar (Difco Bacto). The pH was adjusted to 5.6.

Heavy infestation with bacteria appeared in all test tubes two months later. Two antibiotics, rifampicin and streptomycin, were introduced into the medium at concentrations of 0, 10, 50 and 150 $mg.l^{-1}$. Two methods of sterilization of the antibiotics were tested: filter sterilization and autoclaving. Recording of visual presence of the bacteria as the percent of total clean explants, was done 4 and 8 weeks later.

In order to improve unsatisfactory multiplication rates and elongation the cytokinins 6-benzyl-aminopurine (BAP) and zeatin at 1 $mg.l^{-1}$ were compared with and without the addition of 1-naphthalene acetic acid (NAA) at 0.1 $mg.l^{-1}$ and

gibberellic acid at 1 $mg.l^{-1}$.

A rooting experiment was conducted using three auxins: indole acetic acid (IAA), naphthalene acetic acid (NAA) and indole butyric acid (IBA), at concentrations of 0.5, 1 and 3 $mg.l^{-1}$ and three light intensities, of 110, 150 and 210 $\mu E\ m^{-2}.s^{-1}$. All results were analyzed using Duncan's Multiple Range Test.

3. Results

3.1 Antibiotics

Rifampicin gave better results according to visual evaluation than did streptomycin. The highest concentration (150 $mg.l^{-1}$) and autoclaving gave satisfactory results (table 1).

3.2 Multiplication and shoot length

Both cytokinin and GA_3 had a strong effect on multiplication and growth but the medium containing BAP was better than that with zeatin (table 2). The addition of NAA at 0.1 $mg.l^{-1}$ made no significant difference.

3.3 Rooting

IBA at all concentrations gave much better rooting than IAA or NAA. The highest light level ($210 \mu E\ m^{-2}.s^{-1}$) produced the best rooting with all auxins and all concentrations (tables 3,4).

4. Discussion

The use of antibiotics in plant micropropagation is not well defined and standard formulas do not exist. Falkiner (1990) reviewed the many antibiotics in use and concluded that a specific test is needed for each species.

In the case of *L. cordifolium* rifampicin gave satisfactory results which enabled shoot growth. The use of a high concentration of the antibiotic plus (150 $mg.l^{-1}$) and autoclave sterilization may be of commercial importance.

Good effectiveness of GA_3 in stimulating shoot proliferation and shoot length was reported in several species including some in the Proteaceae family (Ben-Jaacov and Jacobs, 1986), and the significance is even greater due to the need for long shoots for the weaning stage (Tal *et al.* 1991)

The need for higher light intensities, found for rooting of *L. cordifolium*, applies also to micropropagation of some rose cultivars (Bressan *et al*, 1982). These are all woody species, some of which require different *in vitro* rooting environments.

References

Ben-Jaacov, J. and Jacobs, G., 1986. Establishing *Protea, Leucospermum* and *Serruria in vitro*. Acta Hort. 185: 39-51.

Bressan, P.H., Kim, Y.J., Hyndman, S.E., Hassegawa, P.M. and Bressan, R.A., 1982. Factors affecting *in vitro* propagation of rose. J. Am. Soc. Hort. Sci. 107: 979-990.

Falkiner, F.R., 1990. The criteria for choosing an antibiotic for control of bacteria in plant tissue culture. Int. Ass. Plant. Tissue Culture 60: 13-23.

Jacobs, G. and Steenkamp, J.C., 1975. Proteas: the rooting of stem cuttings. Series: Flowers, Ornamental Shrubs and Trees. No. B.3/1975. Department of Agriculture, Republic of South Africa.

George, E.F. and Sherrington, P.D., 1984. Plant propagation by tissue culture. Handbook and Directory of Commercial Laboratories. Exegetics Ltd., Edington, U.K.

Tal, E., Ben-Jaacov, J. and Watad, A.A., 1991. Hardening and *in vivo* establishment of micropropagated *Grevillea* and *Leucospermum*. Submitted to *Acta Hort.*

Table 1 - Percentage of visually bacteria free explants in medium with and without various concentrations ($mg.l^{-1}$) of rifampicin (r) and streptomycin (s) which was added after autoclaving (+) or filter terilization (-).

10r. +	150r. +	1r. -	10r. -	50r. -	150r. -	10s. +	150s. -	10s. -	0
43%	100%	60%	58%	100%	100%	0%	66%	33%	50%

Table 2 - Effect of BAP, Zeatin, NAA and GA_3 on number and length of *Leucospermum cordifolium* shoots.

		BAP		Zeatin	
	0	1 $mg.l^{-1}$	1$mg.l^{-1}$+NAA	1$mg.l^{-1}$	1$mg.l^{-1}$+NAA
		Number of shoots			
0	2.14d	7.43bc	7.86bc	7.0bc	5.71c
GA3 1$mg.l^{-1}$	-	11.14a	9.87ab	8.57ab	6.29c
		Shoot length (cm)			
0	3.07cd	2.07d	2.71cd	4.07bc	4.14bc
GA3 1$mg.l^{-1}$	-	3.71cd	3.14cd	5.14ab	5.57a

Figures followed by a common letter do not differ at P=0.05, according to Duncan's Multiple Range Test.

Table 3 - Effect of auxins (in $mg.l^{-1}$) and light intensity ($\mu E\ m^{-2}.s^{-1}$) on rooting percentage *in vitro* of *Leucospermum cordifolium*.

Light intensity	IAA			NAA			IBA			Control
$\mu E\ m^{-2}.s^{-1}$	0.5	1	3	0.5	1	3	0.5	1	3	0
115	0	50	30	0	0	0	0	55	66	0
150	13	36	44	0	0	0	13	36	66	13
230	33	82	-	42	25	0	62	88	85	22

Table 4 - Effect of auxins (in $mg.l^{-1}$) and light intensity (in $\mu E\ m^{-2}.s^{-1}$) on number of roots per rooted cutting.

Light intensity	IAA			NAA			IBA			Control
$\mu E\ m^{-2}.s^{-1}$	0.5	1	3	0.5	1	3	0.5	1	3	0
115	0	2.8c	2.1cd	0	0	0	0	3c	4b	0
150	1d	3.5bc	1.2d	0	0	0	2cd	3c	4b	2cd
230	2cd	2.3cd	-	1.5d	2cd	0	4.4b	4b	6.1a	2.3cd

Figures followed by a common letter do not differ at P=0.05, according to Duncan's Multiple Range Test.

IN VITRO ESTABLISHMENT OF *PROTEA OBTUSIFOLIA*

A.A. Watad, J. Ben-Jaacov, S. Cohen
Dept. of Ornamental Horticulture
A.R.O., The Volcani Center
P.O. Box 6, Bet Dagan 50-250
Israel

E. Tal, H. Solomon
A.R.O. N. R & D Project
Misgav Experimental Station
M.P. Misgav 20-179
Israel

Abstract

In vitro establishment of *Protea obtusifolia* faces four major problems: browning of tissue and media, no sprouting of axillary buds, endogenous contamination, and tissue vitrification. Multinodal shoots segments of *P. obtusifolia* were established. Etiolation treatment of the new actively growing shoots on the mother plants was helpful to overcome browing and promoted sprouting of axillary buds. Addition of gibberellic acid (GA_3) to the basal medium supplemented with 1 $mg.l^{-1}$ benzylaminopurine (BAP) promoted bud sprouting and elongation.

1. Introduction

The fact that *Protea obtusifolia* is very tolerant to alkaline soil and grows in limestone soil in nature (Eliovson, 1983), makes it very important as a cut flower in Israel, where most of the soils are alkaline. *Protea obtusifolia* can be used also as rootstock for other protea species.

Development of *in vitro* techniques of propagation of *P. obtusifolia* is important for propagation of selected clones, because plants grown from seed have high genetic variability, and vegetative propagation of *P. obtusifolia* by cutting is difficult, slow and unreliable (Perez, 1990).

A major factor that limits tissue culture propagation of proteaceous plants is apparently related to the difficulty of getting axillary buds on explants to sprout (Rugge *et al*, 1989).

2. Materials and Methods

Explants were removed from actively growing terminal shoots from both field- and greenhouse-grown plants and used to initiate cultures. Some of these shoots were kept covered with brown paper bags in order to achieve conditions of lower light intensity and to obtain partly etiolated shoots.

Leaves were cut off, leaving 1-2 cm petioles with some of the leaves. The stems were cut into 4-6-cm-long pieces including two to four nodes. These segments were soaked in 0,1 % Tween 20 for 5 min and rinsed under running tap water for 1 h. Segments were surface-sterilized by stirring them constantly in 1,75 % sodium hypochlorite with 0,1 % Tween 20 for 15 min. After three successive rinses in sterile distilled water, the explants were cultured on the media.

The basal medium contained half strength MS salts (Murashige and Skoog, 1962), vitamins, 3 % sucrose, 9 $g.l^{-1}$ agar. Medium was adjusted to pH 5.5 before autoclaving. The growth regulators used were 6-benzylaminopurine (BAP) and glibberellic acid (GA_3). Cultures were maintained at 25 ± 2 °C, on shelves illuminated for 16 h at 4 000 lux with cool, white fluorescent tubes.

3. Results and Discussion

The main difficulties that were observed with *P. obtusifolia* explants introduced into cultures were (a) browning and blackening of the growth medium and the explants, which usually caused death of the explants. This pheonomenon is known in the case of species that naturally contain high levels of tannins or other hydroxyphenols (George and Sherrington, 1984). (b) No sprouting of axillary buds under *in vitro* culture. (c) Vitrification of the explants tissues. (d) Release of endogenous bacterial contamination from the injured shoot segments into the growth medium. Most of the shoot segments originating from uncovered mother plant shoots turned brown, and died when they were cultured on basal medium or onto basal medium supplemented with 1 $mg.l^{-1}$ BAP, with or without 2 $mg.l^{-1}$ GA_3. Shoots originating from the bag-covered branches remained green but did not show any bud sprouting. This could be due to more lignified tissue, as reported by Rugge *et al* (1989) in the case of *Leucospermum* cv. Red Sunset.

Earlier, Ben-Jaacov and Jacobs (1986) reported that all attempts to induce bud break in *P. neriifolia* x *P. compacta* hybrids in culture failed. Using a semi-etiolated shoot by covering the actively growing shoots on the mother plants, gave softwood explants. Segments from softwood explants did not turn black except for the basal segments and the axillary buds that sprouted. Sprouting and elongation of axillary buds *in vitro* were promoted by the addition of 2 $mg.l^{-1}$ GA_3 and 1 $mg.l^{-1}$ BAP (figures 1, 2). A similar effect of GA_3 was reported in *P. cynaroides* (Ben-Jaacov and Jacobs, 1986). Some of the developed axillary buds became wet or vitrified in culture. A similar observation was reported for *P. cynaroides* as with the soft growth of *Leucospermum* and *Mimetes* (Ben-Jaacov and Jacobs, 1986). Exposing the *in vitro* developing explants under laminar flow was helpful in reducing explants wetness. Various treatments to prevent tissue vitrification are now being studied.

Introduction of multinodal long shoot segments into culture, and retaining some of the leaves, seem to be important for shoot proliferation and axillary bud development.

References

Ben-Jaacov, J., and Jacobs, G., 1986. Establishing *Protea, Leucospermum* and *Serruria* via *in vitro*. Acta Horticulturae 185: 39-52.

Eliovson, S., 1983. Proteas. Macmillan South African Publ. Ltd., Johannesburg.

George, E.F., and Sherrington, P.D., 1984. Plant Propagation by Tissue Culture. Handbook and Directory of Commercial Laboratories. Exegetics Ltd., Eversley, Basingstoke Hants. RG27 OQY, England.

Murashige, T., and Skoog, F., 1962. A revised medium for rapid growth and bioassay with tobacco tissue culture. Physiol. Plant. 15: 473-497.

Perez, J.A., 1990. Propagation by leaf bud cuttings of *Leucadendron* "Safari Sunset", *Leucospermum cordifolium, Leucospermum patersonii* and *Protea obtusifolia*. Acta Horticulturae this volume.

Rugge, B.A., Jacobs, G., and Theron, K.I., 1989. Factors affecting bud sprouting in multinodal stem segments of *Leucospermum* cv. Red Sunset *in vitro*. J. Hortic. Sci. 65: 55-58.

Figure 1 - Sprouting of normal axillary buds of *Protea obtusifolia* 40 days after the introduction of multinodal shoot segments on half-strength MS medium containing 1 mg.l^{-1} BAP and 2 mg./l^{-1} GA_3.

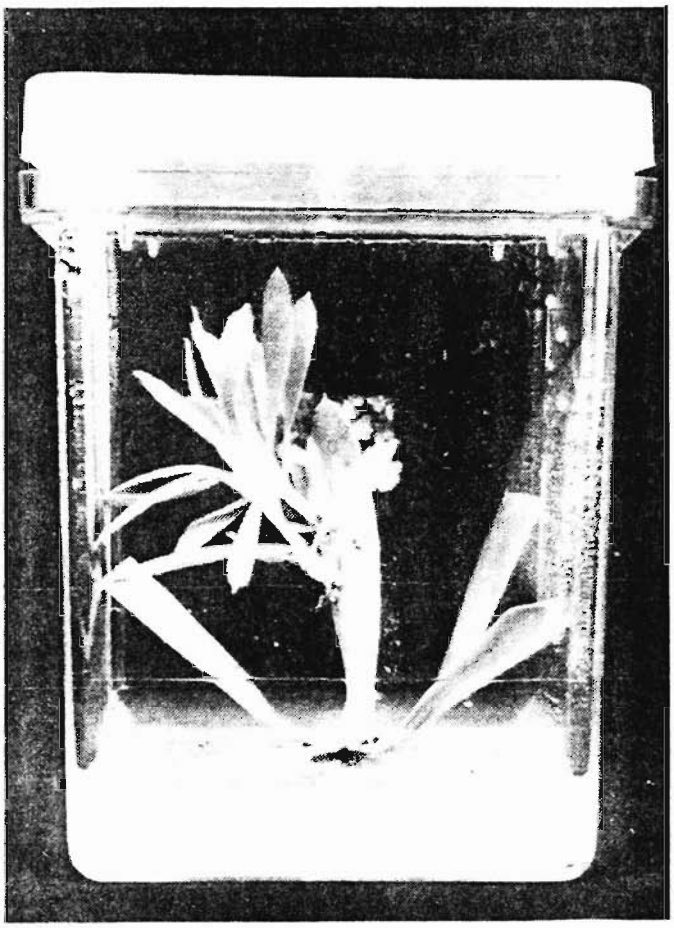
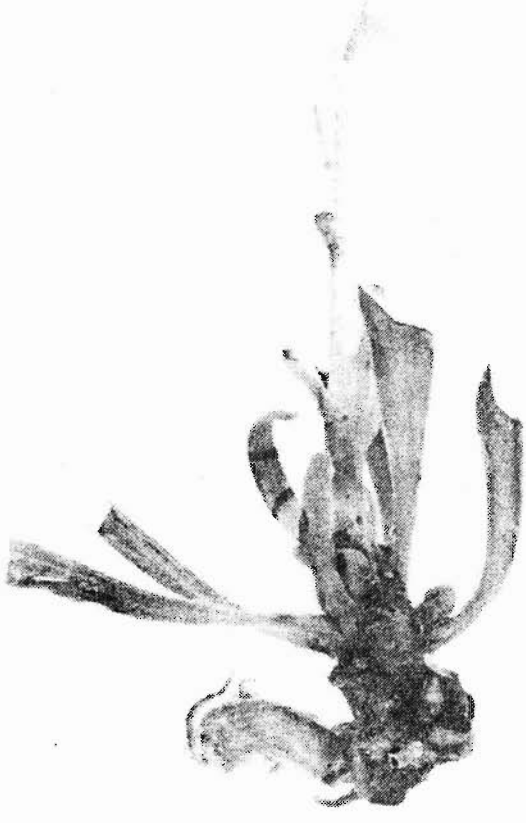


Figure 2 - Apical shoot elongation and abnormal vitrified axillary bud sprouting (bottom left) of *Protea obtusifolia* 40 days after introduction of a multinodal shoot segment into half-strength MS medium containing 1 $mg.l^{-1}$ BAP and 2 $mg.l^{-1}$ GA_3.



HARDENING AND *IN VIVO* ESTABLISHMENT OF MICROPROPAGATED *GREVILLEA* AND *LEUCOSPERMUM*

E. Tal
A.R.O. N. R&D Project
Misgav Exp. Station
M.P. Misgav 20-179
Israel

J. Ben-Jaacov, A.A. Watad
Dept. of Ornamental Horticulture
A.R.O. The Volcani Center
P O Box 6,Bet Dagan 50-250
Israel

Abstract

Studies were carried out to determine the optimal developmental stage of *in vitro* propagated *Grevillea* 'Roundo' and *Leucospermum cordifolium* for transplanting to outdoor conditions. In addition suitable environmental conditions were evaluated.

Longer plantlets with three to five nodes survived better than short ones. Under fog conditions a higher percentage of *in vitro* rooted plants became established than unrooted shoots but under mist the opposite results were observed in *Grevillea* 'Roundo'.

1. Introduction

Weaning of micropropagated material has been a major concern for many years. Problems of stomata opening, photosynthesis and unsuitable root system are considered to be the main limiting factors (George and Sherrington 1984; Zimmerman 1988). Developments in recent years of facilities which allow control of humidity, light, temperature and level of medium wetness improved the rate of weaning success of many species, especially woody ones.

Grevillea 'Roundo' and *Leucospermum cordifolium* both members of the Proteaceae family, are known to need a high light intensity for rooting and growth (Tal *et al.*, submitted 1991). This is probably the main reason for the lack of success in establishing these species in the conventional low-light high humidity chamber.

2. Material and Methods

Five environmental conditions were used with both species: mist, three levels of fog, and a high humidity chamber. Temperature, humidity, light intensity and water flow of the different environments are summarized in table 1.

The fogging machine used was an 'Agritech' (Virginia) centrifugal fogging machine for ventilated high humidity propagation as described by Milbocker (1988) and Harrison-Murray *et al.* (1989). It is controlled by wet and dry thermometers and a thermostat. The arrangement of the different environments is shown in figure 1.

2.1 *Grevillea* 'Roundo': Effect of root stage and plantlet height.

In vitro rooted and unrooted (callused) plantlets were divided into two groups: those up to 0.5 cm in height and those >0.5 cm in height, in a preliminary experiment, and hardened in "Fog 2" (figure 1) for 5 weeks. The same experiment was subsequently repeated in five weaning environments and percent success was recorded after 6 weeks.

2.2 *Leucospermum cordifolium*

The same experimental design as above was applied for *L. cordifolium* using plantlets of four different developmental stages: *in vitro* rooted and unrooted plantlets, both in two shoot lengths: < 1.5 cm and > 3cm. The plants were placed in six weaning environments (Fog 1, 2 and 3; mist; dry mist; closed chamber) at two shade levels (14 Klux and 8 Klux). The additional (sixth) environment was dry mist in which the mist bursts were operated every 45 minutes for 8 seconds.

3. Results

3.1 *G.* 'Roundo'

In vitro-rooted material produced a much higher percent of success in the fogging environments (excluding Fog 1) than the unrooted material, resulting in 13% success of plantlets which were unrooted, 80% with short roots and 88% with long roots, but in the mist environments the unrooted plantlets achieved a higher success rate (figure 2). The wettest environment (fog 1) was the poorest for weaning.

3.2 *L. cordifolium*

The driest fog environment ("Fog 3") produced the overall best results (table 2). Lowering the light intensities reduced remarkably the rate of success in all environments (table 3).

Using taller plantlets (rooted or unrooted) gave much better results than the short plant material (table 2) and *in vitro*- rooted material was superior to unrooted (table 2).

4. Discussion

Relatively high light intensities in the weaning facility improved the rate of success of both species. Similar findings have been reported for micropropagated palms (George and Sherrington 1984). The fact that the combination of high humidity and relatively dry medium is good for weaning of micropropagated material has also been documented elsewhere (Marks *et al*, 1985). The use of plantlets >2cm seemed to be better for *G.* 'Roundo' and essential for *L. cordifolium.*

In vitro rooting was better than *in vivo* rooting, although the rates of success for tall unrooted material of L. *cordifolium* were commercially acceptable. The fact that unrooted material of *G.* 'Roundo' performed better than *in vitro* rooted material in the mist house (contrary to the results in "Fog 2 and Fog 3") may have been due to the wetter medium of the mist house (as in "Fog 1") which caused root rot.

References

George, E.F. and Sherrington, P.W., 1984. Plant propagation by tissue culture. Handbook and Directory of Commercial Laboratories. Exegetics Ltd., Eversley, U.K.

Harrison-Murray, R.S., Howard, B.H. and Thompson, R., 1988. Potential for improved propagation by leafy cuttings through the use of fog. Acta Hort. 227.

Marks, T.R., Wiltshire, S.E., Shepherd, H.R. and Beckham, C.F., 1985. Rooting and weaning of microcuttings. Rep. E. Malling Res. Stn. 73.

Milbocker, D.C. 1988. Ventilated high humidity propagation. Proc. Int. Plant Prop.

Soc. 38: 576-579.
Tal, E., Ben-Jaacov, J., and Watad, A.A., 1991. Micropropagation of selected *Leucospermum cordifolium*: effect of antibiotics and GA_3. Submitted to Acta Hort.
Zimmerman, R. H., 1988. Micropropagation of woody plants: post tissue culture aspects. Acta Hort. 227:489-499.

Table 1 - Weaning environments measured at different times during the day.

Hour of day	Mist	Fog 1	Fog 2	Fog 3	Humidity Chamber
		Temperature (°C)			
0900	26	34	34	34	28
1400	27	33	33	33	28
1700	26	32	32	32	26
		Humidity (% RH)			
0900	85	95-98	95-98	95-98	-
1400	75	95-98	95-98	95-98	83
1700	80	95-98	95-98	95-98	-
		Light (lux)			
0900	5250	5500	5500	5500	-
1200	10125	10100	10100	10100	900
1500	6500	6500	6500	6500	-
		Water mm/h			
1100	1	4	0.8	0.25	1.2

Table 2 - Effect of weaning environment, plantlet length and rooting stage on establishment (% success) of *Leucospermum cordifolium*.

	Short plantlets (<1cm)		Tall plantlets (>1.5cm)	
	in vivo roots	*in vitro* roots	*in vivo* roots	*in vitro* roots
Fog 1	0	0	10	17
Fog 2	10	27	20	60
Fog 3	42	78	70	85
Mist	23	40	55	67
Dry Mist	0	10	0	40
Humidity Chamber	0	0	0	10

Table 3 - Overall effect (mean of all treatments) of light intensity on establishment (% success) of *Leucospermum cordifolium* (best treatment in parentheses)

	14000 Lux	7000 Lux
Fog 1	6.8 (17)	0 (0)
Fog 2	29.2 60)	16.7 (32)
Fog 3	68.7 (85)	42.2 (69)
Mist	46.2 (67)	25.2 (48)

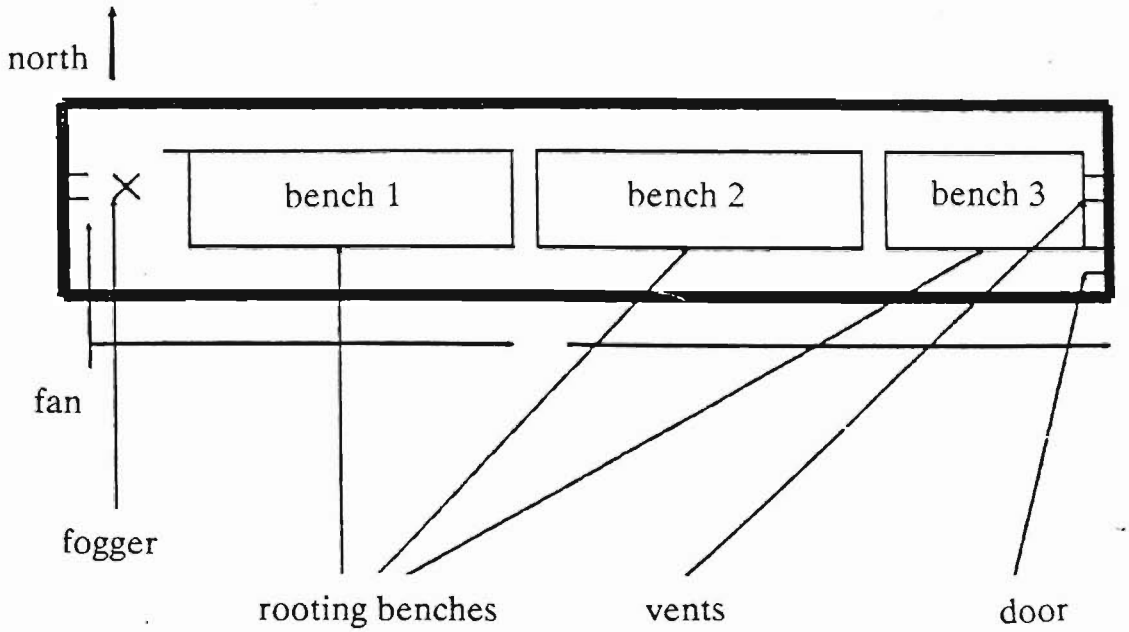


Figure 1 - Fogging tunnel at Misgav Experimental Station

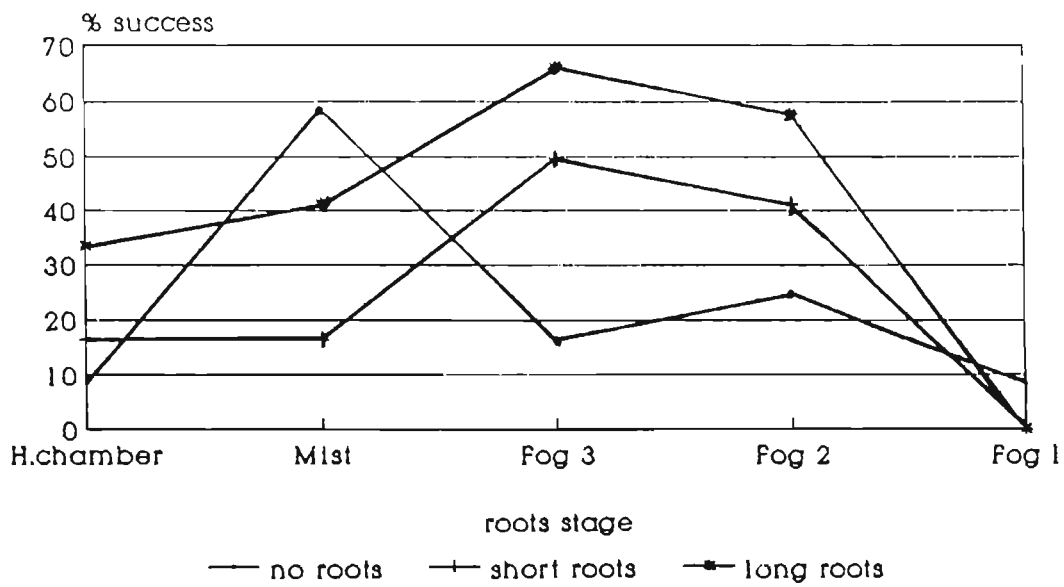


Figure 2 - Effect of root stage and environment on weaning success of *Grevillea* "Roundo"

GRAFTING TECHNIQUES AND THE USE OF ROOTSTOCKS IN *LEUCADENDRON*, AND OTHER PROTEACEOUS PLANTS

J. Ben-Jaacov, A. Ackerman, S. Gilad, R. Carmeli, A. Barzilay and Y. Shchori
Department of Floriculture
A.R.O. The Volcani Center
Bet Dagan 50-250
Israel

Abstract

Breeding can be simplified if grafted plants are used, since the rootstock and the scion plants may be improved separately.

Proteaceae production under Israeli conditions is limited mainly by soil-root problems, namely: high pH of the soil, diseases and nematodes. Grafted plants, using selected, resistant rootstocks can overcome these problems. In addition grafting is being used as a method for vegetative propagation.

Grafting has been accomplished successfully and a large number of grafted *Leucadendron* plants have been produced. It is somewhat more difficult, or useful, with other Proteas.

With *Leucadendron* the clonal *L. coniferum* selection L. "Orot" was tested. As scions *L. discolor* and *L.* "Safari Sunset" was used. *L.* "Orot" seems to be a very suitable rootstock for the above scions under our conditions.

1. Introduction

Some Leucadendrons, Leucospermums, Proteas and Banksias have been grown commercially in Israel since the early 1970's. Production, however, has been limited, mainly because of difficulties which are related to soil problems: too high soil pH, too high soil phosphate and the abundance of soil borne diseases. The climate in Israel seems to be very suitable for many proteaceous plants (Ben-Jaacov *et al*, 1989). About five years ago the hybrid "Safari Sunset" was introduced to Israel. In a few locations it grows well: - on some soils of the Golan Heights and on some of the well-aerated light soils of the coastal plain. Some farmers have developed a method of cultivating "Safari Sunset" in volcanic ash trenches (Ben-Jaacov, 1989). Quality of flowers is excellent (prices up to $0.35 per stem) and the yield is good. These plantations however need special care - daily irrigation and fertilization. The investment is very high and the life span of the plantation is limited. Some Leucadendrons grow very well in Israel and individual plants (*L. muirii*) have survived for over 20 years. The present study describes the technologies developed for grafting several commercial cultivars of *Leucadendron* on selected *Leucadendron* rootstocks. The report includes initial observations on the performance of several scion/rootstock combinations under field conditions.

The idea to use rootstocks in *Proteaceae* has been suggested as early as 1966 by Rousseau. But, even though several studies were carried out on the subject (Jacobs, 1981) there is still no use of rootstocks in commercial Proteaceous cut flower production. There is a need for using rootstocks and grafted plants under Israeli conditions, not only in the genus *Leucadendron* but also in other proteaceous genera. Most Banksias grow well in Israel and the main reason for grafting is vegetative propagation. The reason for grafting *Protea* and *Leucospermum* is the possibility of using resistant rootstocks.

2. Methods and Materials

2.1 Plant Material

We used *L.* Orot (a local clonal selection of *L. coniferum*) as a rootstock and *L. discolor* and *L. Safari Sunset* as scions.

With Banksias we tested *B. integrifolia* and *B. ashbyi* as rootstocks and *B. coccinea* and *B. ashbyi* as scions. In the genus *Protea* we used *P. obtusifolia* and *P. susannae* as rootstocks and *P. neriifolia. P. cynaroides* and some selected cultivars as scions. In the genus *Leucospermum* the main rootstock tried was *L. patersonii* with few observations also on *L. conocarpodendron*. As scions, cultivars of *L. patersonii*, selected clones of *L. cordifolium* and some *L cordifolium* x *L. lineare* hybrids were used.

2.2 Methods of propagation and grafting

Propagation of *L.* Orot was done by cuttings: stem segments, 10cm long and 2-3 mm in diameter were taken from 2 year old *L.* "Orot" plants grown in 35 litre plastic containers, containing 75% tuff: 25% peat growth medium. Leaves were stripped off the bottom half of the cuttings. The bases of the cuttings were dipped for 10 seconds in aqueous solution of 4000 ppm K-IBA. The cuttings were stuck in 1 inch-cell plug trays ("speedling") containing rooting medium of 50% peat: 50% styrofoam. Temperature at the base of the cuttings was kept at 24±2°C. The rooting was done in a temperature controlled (max. : 25°C winter, 32 °C summer, min. : 11°C) glasshouse. The cuttings were kept under intermittent mist (on 20 sec., off 20 min). Rooting of 55-75% occurred 8 weeks after planting, when the rooted cuttings were repotted into 5 cm plastic pots.

Grafting was done 3-6 months after repotting when the plants were actively growing. Stem segments, 5 cm long with 2 fully developed leaves were used as scions. Top-cleft grafts and side grafts were tied with plastic strips and covered with white plastic bags. Most successful grafts started sprouting 2-3 months after grafting. Similar methods were used with other Proteas.

2.3 The plantation

Soil and soil preparation: The experimental field used is located at the Department of Floriculture, A.R.O. The Volcani Center, Bet Dagan. The soil is sandy loam, rich in lime agregates. The pH is 8.2 at 10 cm depth. The soil was ploughed well to a depth of 40 cm, treated with methyl bromide and leached well before planting.

Planting was done in May 1989. Plants are watered with on-line (3/meter) 2 litre/h drippers. Watering is done once a week providing 2 litre/plant/day. Every irrigation includes 100 g/m^3 ammonium sulfate, 30 g/m^3 potassium nitrate 2.5 g/m^3 Fe Chelate (Sequestreen) and 30 ml/m^3 microelements complex ("Korateen").

3. Results and Discussion

3.1 Grafting

Only initial results of graft take have been summarized. In grafting *L. discolor* and *L.* "Safari Sunset" on L. "Orot" in late fall, winter and mid spring, 40-100% graft take was achieved. Early March seems to be a generally favourable date for grafting of all combinations. There is still no sufficient data to indicate superior rootstock/scion combinations.

3.2 Growth response under field conditions

The preliminary experiment included 2 groups of plants established in adjacent rows. In one group the plants were in the following order:

1) "Orot"
2) "Safari Sunset" on its own roots
3) "Safari Sunset" on "Orot"
4) "Safari Sunset" on its own roots
5) "Safari Sunset" on "Orot"

In the second group "Safari Sunset" was exchanged by *L. discolor*. Now, a year after planting all four plants of "Safari Sunset" and *L. discolor* grown on their own roots are chlorotic and have not grown at all. The grafted plants and the "Orot" rootstock plants are growing very well (reaching a height of about 1 meter). The growth is at least as good and even better than "Safari Sunset" plants of identical age grown on their own roots in the best plantations in the country (in volcanic ash), in artificial medium and on low pH sandy loam of the coastal plane. Grafted *L. discolor* plants grow well and produce very dense bushes, denser than *L. discolor* grown on its own roots in sandy soils.

Since it is difficult to grow leucadendrons in most soils in Israel, the above technology may make it possible to produce successfully *L.* "Safari Sunset" and *L. discolor* in a much wider range of soils. In addition, the proposed method may make it possible to grow in Israeli soils a much wider range of *Leucadendron* cultivars. Continuation of the present study and further testing of other scion/rootstock combinations may lead to specifically selected scion/rootstock/soil combinations.

3.3 Results obtained with other proteaceous genera

With *Leucospermum* grafting is very easy. We have not, however, identified the most suitable rootstock. *L. patersonii* seems to be the best - it is very lime (high pH) tolerant. However, it has a relatively short life span under Israeli conditions and is sensitive to nematodes.

We were successful in grafting only a few Banksias. Scions of selected *Banksia ashbyi* clones were grafted successfully on one year old *B. ashbyi* seedlings. Rate of grafting success was very low (10-15%) but these grafted plants grow well in our experimental plantation.

In the genus *Protea* best results, in both grafting take and growth performance, were achieved with *Protea obtusifolia* as a rootstock. We have not encountered any incompatibility problems and even *P. cynaroides* was grafted successfully on *P. obtusifolia* (short term observation).

References

Ben-Jaacov, J., Ackerman, A., Gilad, S. and Shchori, Y., 1989. New approaches to the development of proteaceous plants as floricultural commodities. Acta Hort. 252: 193-199.

Jacobs, G., 1981. Vegetative propagation of Proteas - recent developments. In: P. Mathews, Growing and marketing Proteas : 7 - 18. Proteaflora Enterprises Pty. Ltd., Melbourne, Victoria, Australia.

Rousseau, G.G., 1966. Proteas can be grafted. Farming in South Africa 42 (6) : 53-55.

CONTROL OF GROWTH AND FLOWERING IN *BANKSIA ASHBYI*, *LEUCOSPERMUM PATERSONII* AND *LEUCADENDRON* "SAFARI SUNSET"

Israela Wallerstein and Ada Nissim
Dept. of Ornamental Horticulture
A.R.O. The Volcani Center
P.O. Box 6, Bet Dagan 50-250
Israel

Abstract

The patterns of growth and flowering of *Leucospermum patersonii*, *Banksia ashbyi* and *Leucadendron* "Safari Sunset" under natural growth conditions in Israel are almost the same. In all cases active vegetative growth takes place during spring and summer, cessation of elongation at the end of summer, and inflorescence initiation during autumn. During autumn sprouting, but not elongation of axillary buds, can be forced by pruning in *Banksia* and *Leucadendron*.

The ability of a meristem to initiate an inflorescence depends on the shoot growth rate during spring and summer. Based on shoot growth data it appears that inflorescence initiation in these plants is controlled by the long-short day sequence. This suggestion was proved to be correct in *Leucospermum patersonii*, grown under phytotron controlled conditions. In this case long-days not only influenced the rate of vegetative growth before inflorescence initiation, but also activated the axillary buds, so that the initiation could occur under short-day conditions. Short-days are needed further during the development of the inflorescence primordia. Inflorescence development in *L. patersonii* and *B. ashbyi* can be accelerated by moderate temperatures, especially during the primordial stage. The spread of the *B. ashbyi* flowering over a period of 6 months is due to the stage of inflorescence development at the beginning of winter.

The effects of benzyl adenine and gibberellic acid on *Leucospermum* inflorescence development and on *Banksia* shoot growth are reported and concidered from the standpoint of controlling the flowering period.

1. Introduction

Leucospermum patersonii, *Banksia ashbyi* and *Leucadendron* "Safari Sunset" are three species of the Proteaceae grown commercially in open plantations in Israel. The growth characteristics of *L. patersonii* and *B. ashbyi* were described previously (Wallerstein, 1989a; Wallerstein, 1989b, respectively). Shoots of the two species sprout in spring (March-April), elongate during summer, stop elongating at the end of summer (August-September), initiate inflorescence primordia in autumn (September-October), have inactive axillary buds during winter (October-March), and flower in early spring (February-March) in *Leucospermum* or during winter and spring (December-May) in *Banksia*. Control of the flowering period could be divided roughly into two major processes: control of inflorescence initiation and control of further flower development.

Even though inflorescence initiation in *L. patersonii* and B. *ashbyi* occurs under the short-days of autumn, the ability of the meristem (apical in *Banksia* and axillary in *Leucospermum*) to initiate an inflorescence depends on the previous effect of long-days on shoot growth and cessation of elongation (Jacobs, 1985; Wallerstein, 1989a,b). This is indicated also by the fact that inflorescence initiation occurs in autumn but not in spring, in spite of the similarity in day-length and temperatures

between the two seasons. Long-days are essential for the prefloral stage of inflorescence initiation in *Leucospermum* while further initiation occurs under short-day conditions (Wallerstein, 1989a).

The rate of development of inflorescence primordia in *Banksia* and in *Leucospermum* depends on temperature, being accelerated by moderate temperatures (Wallerstein, 1989a). It is suggested that early initiation of inflorescence, within the inductive period, may promote the development of the inflorescence under the moderate temperatures of autumn. Because critical vegetative mass, reached under long-day conditions, is essential for inflorescence initiation, advancing the sprouting time in the preceeding spring and the further elongation period seems to be important for early initiation in autumn.

We studied the effect of benzyl adenine (BA) and gibberellic acid (GA_3) on early sprouting and shoot development in *B. ashbyi* and on inflorescence development of *L. patersonii*. We also followed the effect of pruning period on shoot development and inflorescence initiation in *Leucadendron* "Safari Sunset".

2. Material and methods

Plants of *L. patersonii* propagated from cuttings, and of *B. ashbyi* propagated from seeds, were grown outdoors in 10 litre plastic pots containing volcanic tuff: peat mixture (1:1, v/v), using commercial irrigation and feeding practices. Two-year-old *Leucospermum* plants initiated inflorescences at the beginning of October. On November 24, the inflorescence buds were less than 0.5 cm in diameter. At this stage the plants were sprayed three times, at 14-day intervals, with Promalin (containing equal concentrations of BA and GA_3), BA or GA_3 (100, 250 or 400 ppm) plus 0.1% Tween 20. Each treatment included ten plants. Observations included number of inflorescence buds per shoot, number of flowering shoots per plant and number of axillary vegetative shoots per main shoot at the time of flowering.

Three-year-old *Banksia* plants were treated with BA and GA_3 in the same way as the *Leucospermum* plants. The experiment started on November 25 and included plants grown outdoors as well as plants introduced into a greenhouse with minimum temperature of 18 °C. Each treatment included ten plants. The sprouting time, elongation and thickening of new shoots were followed.

Three-year-old *Leucadendron* "Safari Sunset" plants grown in a commercial plantation were pruned all year round, using different groups at the beginning of each month. The date of growth resumption after pruning was recorded and the elongation of the new shoots was followed until they reached their maximal length at the time of growth cessation. On February 1 the degree of inflorescence differentiation was scored as follows: 1. vegetative, 2. one whorl of primordial bracts, 3. complete inforescence primordium. Each treatment included five replicates of ten shoots.

3. Results

As indicated in table 1, 100 and 250 ppm of GA_3 advanced the main flowering date of *Leucospermum* by about one month, from the beginning of April to the beginning of March. The quality of the inflorescences sprayed with GA_3 was poor, due to their light colour and the irregular opening of the florets. Higher concentrations of GA_3 significantly reduced the number of flowering shoots per plant. Benzyl adenine, at all concentrations, reduced the number of flowering shoots per plant. The effect of BA, when applied as Promalin, overrided the effect

of GA_3 in Promalin.

Application of BA to *Leucospermum* shoots bearing inflorescence buds at an early stage of development caused simultaneous development of severeal inflorescences on the same shoot, a process which resulted in abortion of reproductive buds and their replacement by vegetative shoots (table 2).

Gibberellic acid and BA, at all concentrations, advanced the early sprouting of new axillary shoots of *Banksia* (table 3). The effect of GA_3 disappeared with time, while that of BA on the number of new sprouting shoots increased, and secondary new shoots appeared during March (but were not counted). Both BA and GA_3 accelerated the elongation of the new shoots but the effect of BA disappeared during March whereas that of 300 ppm GA_3 became stronger (table 4). The shoots which were forced to grow by BA were more vigorous and thicker than those treated with GA_3. When outdoors treatments are compared with greenhouse ones (table 5 vs tables 3 and 4), it can be seen that the main effect of BA and GA_3 remained the same but that in the greenhouse sprouting date was advanced by about one month in all treatments and the elongation rate of the new shoots was higher.

New shoots of *Leucadendron* "Safari Sunset" control plants sprouted in March, stopped elongating in July, and flowered during the following spring (table 6). Pruning plants at the beginning of each month between March and July, was followed by axillary bud sprouting one month later. The new shoots stopped elongating during August-September. Axillary buds of plants pruned between August and October sprouted two months after pruning and stopped elongating immediately after sprouting, indicating that elongation ceases between July and October while axillary buds are still able to sprout after pruning but not to elongate more that 3-5 cm. Plants pruned between November and February sprouted during spring when the control plants sprouted, indicating that the axillary buds are inactive during winter. The ability of the meristem to initiate an inflorescence seems to depend on time of growth cessation or maximal shoot length.

The earlier the time of growth cessation, the longer was the maximal shoot length reached and the higher was the stage of the inflorescence development at the beginning of February. Plants pruned later than June failed to flower during the following spring.

4. Discussion

The main pattern of growth activities found in *Leucadendron* "Safari Sunset" (table 6) was similar to that of *B. ashbyi* and *L. patersonii* (Wallerstein, 1989a,b). Their growth cycle included sprouting in the spring, cessation of shoot elongation in late summer, inflorescence initiation in autumn, cessation of axillary buds activity in winter and flowering in spring. The ability of the *Leucadendron* apical meristem to initiate an inflorescence depends on the date when elongation stops and/or on shoot length at that time. This kind of dependence seems to be common to the three species of the Proteaceae family and is probably the key to the control of their flowering period. The fact that axillary buds of *Leucadendron* can sprout but can not elongate during autumn is similar to the situation found in *Banksia* (Wallerstein, 1989b) and is probably and indication to the positive effect of long-days on shoot elongation, as is the case in *Leucospermum* (Wallerstein, 1989a).

Both BA and GA_3 can be used to advance the sprouting of new shoots in early spring. In the case of *Banksia*, BA is preferable because of its positive effect on

the number and vigour of the new shoots (tables 3,4 & 5). Long-days during early spring are important for early shoot elongation, so that (a) maximal length and cessation of elongation will be attained earlier than in the control, (b) inflorescence initiation will be expected at the onset of short-days, and (c) its further development will be possible under the moderate temperatures of autumn. Neither BA nor GA_3 can be used to accelerate the development of the inflorescence in *Leucospermum* (tables 1 & 2). Benzyl adenine caused simultaneous development of several inflorescent buds, which was followed by their abortion. The effect of GA_3 on abortion and rate of development was dependent on its concentration (table 1). The lowest concentration accelerated the flowering date without causing abortion but has a negative effect on colour and order of flower opening in the inflorescence, and the higher concentrations also caused abortion. The fact that reproductive buds of *Leucospermum* abort as a result of BA affecting their simultaneous development, should be taken into account when this growth regulator is used to accelerate the sprouting of *Leucospermum* vegetative buds in early spring.

References

Jacobs, G., 1985. *Leucospermum*. In: Halevy, A.H. (ed.) Handbook of Flowering. Vol. 3, pp. 283-286. CRC Press, Boca Raton, FL.

Wallerstein, I., 1989a. Sequential photoperiodic requirement for flower initiation and development of *Leucospermum patersonii* (Proteaceae). Israel J. Bot. 38: 25-34.

Wallerstein, I., 1989b. *Banksia ashbyi*. In: Halevy, A.H. (ed.) Handbook of Flowering. Vol. 6, pp. 87-92. CRC Press, Boca Raton, FL.

Table 1 - The influence of gibberellic acid (GA_3) and benzyl adenine (BA) on number of flowering shoots and on flowering date of *Leucospermum patersonii*.

Treatment and concentration ppm)	Number of flowering shoots per plant					
	Date of flowering					Total
	25/2	11/3	24/3	13/4	1/5	
Control	0.0	0.0	1.5ab	5.3a	0.5ab	7.3a
Promalin						
100	0.0	1.6b	0.6b	1.0b	0.7ab	3.9c
250	0.0	1.9b	0.0	0.0	0.0	1.9e
400	0.0	0.0	0.0	0.0	0.0	0.0
GA_3						
100	0.0	4.2a	1.3ab	1.3b	1.3a	8.1a
250	0.0	4.9a	1.0ab	0.5b	0.0	6.4b
400	0.0	1.2bc	0.6b	0.6b	0.1b	2.5de
GA						
100	0.0	0.3c	1.6a	0.6b	0.4ab	2.9d
250	0.0	0.0	0.0	0.0	0.0	0.0
400	0.0	0.0	0.0	0.0	0.0	0.0

Values within columns followed by different letters differ significantly (P = 0.05)

Table 2 - Effect of benzyl adenine on the development of vegetative or reproductive axillary buds in *Leucospermum patersonii*.

Benzyl adenine (ppm)	Inflorescence buds (no./shoot)	Flowering shoots (no./plant)	Axillary shoots (no./shoot)
Control	0.2d	7.5a	1.5c
100	1.2c	3.0b	8.3a
250	4.4a	0.0	10.0a
400	3.3b	0.0	10.0a

Values within columns followed by different letters differ significantly (P = 0.05).

Table 3 - Number of new shoots on *Banksia ashbyi* plants growing outdoors and treated with gibberellic acid (GA_3) or benzyl adenine (BA).

Treatment and concentration (ppm)	Number of new shoots on various dates								
	30/1	5/2	12/2	19/2	26/2	6/3	11/3	19/3	26/3
Control	0.0	0.0	0.4e	0.8e	1.4e	2.0c	2.8c	4.2c	5.8b
GA_3									
300	0.8a	1.0bc	1.0de	1.0e	1.0e	1.8c	3.8c	5.2c	6.2b
500	0.2a	0.2a	0.4e	0.4e	0.4e	1.8c	3.2c	4.6c	6.6b
1000	0.6a	0.6bc	1.2de	1.2e	1.2e	1.4c	1.4c	2.0c	3.0b
BA									
100	2.8a	3.0abc	4.8abcd	6.8bcd	9.6bc	13.6ab	15.2ab	16.2ab	18.0a
300	2.6a	4.4ab	6.4abc	9.0abc	11.6ab	15.0ab	17.8a	18.4a	20.6a
500	0.0	5.4a	8.8a	12.8a	16.4a	8.8a	22.6a	25.6a	28.4a

Values within columns follwed by different letters differ significantly (P = 0.05)

Table 4 - Length and diameter of new growing shoots of *Banksia ashbyi* plants growing outdoors and treated with gibberellic acid (GA_3) or benzyl adenine (BA)

Treatment and concentration (ppm)	Length of new shoots on various dates (cm)									Shoot diameter (mm)
	30/1	5/2	12/2	19/2	26/2	6/3	11/3	19/3	26/3	19/3
Control	0.0	0.0	0.2d	0.3c	0.7c	1.4bc	2.3bc	3.4bc	5.1cd	3.0ab
GA_3										
300	0.7a	1.2a	2.5a	4.2a	7.7a	14.1a	19.4a	23.5a	26.1a	2.4bc
500	0.1bc	0.5ab	0.2d	1.7bc	0.7c	5.0bc	7.8bc	10.4bc	12.9bcd	2.3bc
1000	0.3abc	0.3ab	0.8bcd	1.3bc	2.1bc	4.0bc	6.1bc	8.6bc	10.6bcd	2.3bc
BA										
100	0.2bc	0.7ab	1.6ab	2.9abc	3.3bc	5.7b	7.8bc	9.9bc	12.8bcd	3.0ab
300	0.3ab	0.8ab	1.6ab	2.5ab	3.5b	6.4b	8.4bc	10.5bc	12.6bcd	3.2a
500	0.0	0.7ab	1.3bcd	2.1bc	3.0bc	6.2b	9.3bc	11.9bc	16.3b	3.2a

Values within columns follwed by different letters differ significantly (P = 0.05).

Table 5 - Length and diameter of new shoots of *Banksia ashybi* plants grown in a greenhouse (18 °C minimum) and treated with benzyl adenine (BA) or gibberellic acid (GA_3).

Treatment and concentration (ppm)	Length of new shoots on various dates (cm)					Diameter (mm)	
	1/1	8/1	15/1	22/1	27/1	22/1	27/2
Control	0.8bc	1.5b	2.8e	4.5de	13.7b	3.0b	3.0a
GA_3							
300	4.0a	10.8a	20.0e	26.9a	51.5a	2.0c	3.7a
500	1.8abc	8.6a	18.1ab	27.2a	58.3a	2.3c	3.0a
1000	3.2ab	3.8b	12.9bc	16.6b	26.9b	2.0c	3.0a
BA							
100	0.9bc	2.4b	4.4de	7.0de	17.6b	3.0b	3.4a
300	0.9bc	2.0b	4.2de	7.0de	17.4b	3.6ab	3.8a
500	0.9bc	1.9b	4.3de	7.2de	21.7b	3.8a	3.4a

Values within columns followed by different letters differ significantly (P = 0.05).

Table 6 - The influence of pruning date on shoot growth and inflorescence initiation of *Leucadendron* "Safari Sunset".

Date of pruning	Date of growth resumption	Date of growth cessation	Maximal length of shoot (cm)	Degree of inflorescence differentiation
1987				
Control	15.3	1.7	50.0a	3.0a
March	1.4	2.8	40.4a	2.1b
April	1.5	2.8	35.1ab	1.5c
May	1.6	3.9	25.6ab	1.8bc
June	1.7	3.9	24.0b	1.5c
July	1.8	4.10	12.0c	1.1d
August	4.10	4.10	5.1d	1.0d
September	4.11	4.11	4.8d	1.0d
October	2.12	2.12	3.2d	1.0d
November	2.3	pruning		
December	2.3	pruning		
1988				
January	17.4	pruning		
February	17.4	pruning		

Values within columns followed by different letters differ significantly (P = 0.05).

FLOWERING AND VEGETATIVE GROWTH OF *PROTEA NERIIFOLIA* AND *PROTEA CYNAROIDES*

S.A. Dupee and P.B. Goodwin
Department of Horticulture
School of Crop Science
University of Sydney
Sydney, New South Wales 2006
Australia

Abstract

Differences in the flowering period and in the productivity of *Protea cynaroides* 'Long Leaf' occur between four commercial plantations in Southeastern Australia. The number of flowers varies from 45 flowers on 17 plants (2.65/plant) to 16 flowers on 5 plants (3.2/plant). The clone *Protea neriifolia* 'Salmon Pink' has a different bloom distribution and yield at each plantation. More blooms were produced at the two warmer plantations. Bloom production at each plantation on 25 plants varied from 26 to 112 during 1988 to 157 to 579 during 1989. The two species have different vegetative phases resulting in different seasons of flowering. Soil particle analysis and nutrient level influence root growth and shoot development.

1. Introduction

The process of flowering in *Protea* is dependent on a number of factors which influence the growth and development of root systems, vegetative shoots and flowering stems. Within the natural habitat of the south-western Cape Province of South Africa, *Protea cynaroides* grows mostly in moist coastal mountains at elevations from 200 to 1000m. These areas have wide daily temperature fluctuations (typical average daily range of 7°C to 24°C) with reliable annual rainfall of at least 400mm (Rourke, 1980; Vogts, 1977). This amount of rain may fall at any time during the season, resulting in no prolonged dry periods. *Protea cynaroides* always grows in acid, sandy soils derived from Table Mountain Sandstone or Witteberg quartzite (Vogts, 1982). Both the surface and subsoil require adequate drainage, and must be quick draining during heavy rains.

Vogts (1977) reported that the flowering period of each *P. cynaroides* variant is genetically stable. Thus when four or more variants are grown together in the same environment, their characteristic flowering times are retained. The variants are known by the names of their ecological areas; their characteristics are determined by the soil, microclimate and associated plants of each area. The Long Leaf variant of *Protea cynaroides* is vigorous and has a long flowering period (May to November) that may result in a production of 20 blooms per year (Vogts, 1977; 1982). The Long Leaf variant grows in foothills or coastal plains of the eastern and southern regions of South Africa. This variant has long narrow leaves, long stems and long involucral bracts, and is well suited for commercial cut flower production. After four or five annual growth flushes, a given stem terminates in a flower head (Rourke, 1980).

Protea cynaroides tends to grow in specific areas which have a sandy soil with ground water low in dissolved nutrients (Vogts, 1977). The erratic occurrence of such restricted soil types in the natural habitat seems to be the main determinant of the species' distribution. Soils on which *Protea cynaroides* grows contain at least 60% sand and less than 10% clay, pH of 3.5 to 4.5, electrical conductivity of 1250

or higher for the topsoil and low levels of phosphorous, potassium and sodium. Lamont (1986) reports that proteoid roots grow near the surface and concentrate where organic matter decomposes. The number of proteoid roots varies greatly with soil type, being greatest in sandy soils. Claassens (1986) noted that *Protea* respond to different levels of nitrogen and Parvin (1986) indicated that chlorotic symptoms appear with low levels of calcium and magnesium.

Protea neriifolia grows and flowers in South Africa in similar conditions to *Protea cynaroides*, along the coast up to 1300m. This species also prefers sandy soils but has a wider soil tolerance than other *Protea* species (Vogts, 1982). Initial growth of *Protea neriifolia* with good conditions can be rapid (Rourke, 1980). The flowering period is dependent on the locality, being from February to November.

Protea neriifolia grown in a summer rainfall area of South Africa flowers earlier than in the Cape, from January to July (Heinsohn and Pammenter, 1988). Shoot growth occurs from July to February, with a peak in September. Root systems of this species and others may spread to 3m with 60% or more of the mass in the upper 10cm of soil (Higgins *et al*, 1987).

The time of flowering of *Leucospermum cordifolium* has been shown by Jacobs and Honeyborne (1979) to have a linear relationship with daily mean temperature. Heat unit accumulation of 925 units with a base temperature of 5.8 °C in spring will result in maturation of approximately 90% of the buds at one location. In Hawaii, Criley, Parvin and Lekawatana (1990) found that flower development of *Leucospermum cordifolium* 'Vlam' is correlated to degree day accumulation with a 6°C base and solar radiation accumulation. Air temperature has also been shown to control the development of flower buds of *Strelitzia reginae* in four separate locations (Halevy *et al*, 1987). Temperatures below 13°C retarded leaf and flower development and over 17°C and up to 27°C increased development. The seasonality of flowering varied in each location and was dependent on the air temperatures of those sites.

The main objective of this study is to examine the variation in flowering between four field locations of cultivated *Protea cynaroides* 'Long Leaf' and a clone of *Protea neriifolia* in southeastern Australia. The phases of vegetative growth are being studied in relation to the period of flowering of both species. Soil conditions and root growth are being correlated with productivity of plants. This work will provide a basis for manipulating flowering time by means of varying the time of planting and of pruning; define the effect of air temperature on flowering periods; and enable the use of multiple sites to produce a uniform volume of cut flowers.

2. Materials and Methods

One year old clonal plants of *Protea neriifolia* 'Salmon Pink' and one year old seedlings of *Protea cynaroides* were planted in April 1987 at four commercial plantations along the southeast coast of Australia. The northernmost site is at Springbrook, Queensland, latitude 28° 14'S, longitude 153° 17'E. The southernmost site is located at Robertson, New South Wales, latitude 34° 35'S and longitude is 150° 37'. A total of 25 plants of both species were planted in a completely random design at each plantation. Individual plants were spaced at 2m on center in rows. Weed mat was used to control weeds.

Vegetative growth of both species was recorded for all plants at the Peats Ridge site, from winter (June) 1988 to May 1990. all large stems of at least 40 cm were measured on each plant of *Protea cynaroides*. The shoot length of *P. neriifolia* was

measured on ten healthy main branches, including any side shoot or bypass growth. The cumulative amount of growth was recorded for each month.

During June 1990 the root systems of five plants of each species were carefully removed from the ground. Fresh mass of above ground growth and roots were recorded. The number of proteoid roots clumps were counted and the distribution of the roots in the soil profile was noted. A soil analysis using replicate soil samples was completed to determine the physical and chemical properties of the soil at each plantation. Acidity was measured using 1:5 soil-water and with calcium chloride (.01M), to give measurements more relative to field conditions. Electrical conductivity was measured. Bulk density was sampled with 5 cm cores and the soil oven dried. Particle size analysis was completed using a hydrometer. Organic matter was measured by the modified McCloud Organic Carbon method using potassium dichromate. Nitrogen levels were determined by the Kjheldahl method. Phosphorous levels were recorded by the Colwell phosphate method. Soil cations were analyzed by an atomic absorption spectrophotometer with a 1:5 solution of silver thiourea.

The flowering of all plants at Springbrook, Peats Ridge, Kurrajong Heights and at Robertson was recorded using Julian days. Each flowering date for every inflorescence was recorded as the time when the inner bracts of both species separated at the top of the flower head. Length of flower heads and stem length were also recorded. The following climatic data at each plantation was recorded daily: maximum and minimum temperatures, amount of rainfall and number of rainy days. Temperature and rainfall records are summarised by months.

3. Results

3.1 Vegetative Growth

The amount of vegetative growth differed between the two years for *Protea cynaroides* at Peats Ridge as shown in figure 1. The data suggest two growth periods and two rest periods during the year. Figure 2 illustrates the vegetative growth of the 'Salmon Pink' clone. This clone appears to grow by multiple flushes in the spring. The total growth during late 1989 and early 1990 was considerable due mostly to many side shoots.

3.2 Root Growth and Distribution

The distribution of roots for *Protea cynaroides* is shown in table 1. Plants at Kurrajong Heights and Robertson show roots to be most numerous in the top 8 cm of the soil. Differences occur in proteoid root formation as well as in overall trunk diameter. Root systems of the plants at Peats Ridge were the most prolific and deepest.

Table 2 shows that the 'Salmon Pink' clone also had many surface roots and produced more proteoid roots than *Protea cynaroides*. Plants at Peats Ridge and Springbrook had twice the mass of those at Kurrajong Heights and Robertson.

Table 1 - Root distribution of *Protea cynaroides*

Site	Ave.No. Proteoid Roots	Ave.No. of Main Roots	Main Roots at Depth: 0-8 cm	8-16 cm	16-24 cm	24-32 cm	Trunk Diam. (mm)	Fresh Top Mass (kg)	Fresh Root Mass (kg)
Springbk	62	46	12	22	10	2	59	4.29	2.28
Peats Rd	73	53	13	11	14	15	86	4.90	2.27
Kurr. Hts.	49	25	13	7	4	1	58	0.96	0.41
Roberts.	61	29	11	9	6	3	43	1.42	0.76

Table 2 - Root distribution of *P. neriifolia* 'Salmon Pink'.

Site	Ave.No. Proteoid Roots	Ave.No. of Main Roots	Main Roots at Depth: 0-8 cm	8-16 cm	16-24 cm	24-32 cm	Trunk Diam. (mm)	Fresh Top Mass (kg)	Fresh Root Mass (kg)
Springbk	188	55	28	16	8	3	55	2.82	1.61
Peat Rd	156	38	23	5	5	5	58	2.54	1.73
Kurr. Hts.	59	28	19	6	3	0	34	1.07	0.78
Roberts.	133	34	15	15	3	1	34	1.22	0.77

3.3 Soil Analysis

When compared to the soil of the three other plantations, the soil at Kurrajong Heights has the highest electrical conductivity and levels of phosphorous, potassium, magnesium, calcium and sodium as shown in tables 3 and 4. The soil with the lowest level of nutrients was found at Peats Ridge. This soil has a high bulk density, low levels of clay and organic matter and a high percentage of sand. the nutrient levels are relatively low. The soil at Robertson has a reasonable fertility with a high level of organic matter. The soil tends to hold water and has a high cation exchange capacity. The surface soil at Springbrook has a high level of clay and organic matter with easonable nutrient levels.

The particle size analysis indicates that the Peats Ridge soil is a sand. The soil at Springbrook, Queensland is a loam. the soild at Kurrajong Heights is the most heavy, being a clay loam. The reddish soil at Robertson is a sandy loam.

3.4 Productivity of Flowers

The King Protea (*P. cynaroides*) at Peats Ridge produced the most flowers (table 5). Plants at this plantation had the most stems as well as the earliest blooms of the four sites. A few plants at Springbrook produced most of the flowers. Flowering was not yet complete on plants at Kurrajong Heights. All plants at Robertson had not yet initiated flower buds and stems appeared to be ready to set flowers in the next few months.

The clone *P. neriifolia* 'Salmon Pink' produced a prolific number of flowers in

the second season (table 6). During the first year there was significant growth and flowering at Springbrook. During 1989, plants at Peats Ridge produced copious quantities of flowers with good (long) stems. The plants at Kurrajong Heights produced a reasonable number of flowers over both years. In contrast, the number of flowers was lowest at Robertson for both years.

3.5 Seasonality of Flowering

During 1988 plants of *Protea neriifolia* 'Salmon Pink' bloomed heavily in April and May. the next year flowering became earlier (figure 4). For both years the production of a few flowers over winter and spring was due to side branch flowers.

Flowering at Peats Ridge occurred over the longest period (figures 3,4). The Robertson plantation has a good spread of flowering while Springbrook flowers develop quickly. In both years flowers at Springbrook and Peats Ridge matured in a glut. By contrast, plants at Robertson had the shortest flowering season and bloomed later than the other three plantations.

3.6 Differences of Climate

The monthly averages of the maximum and minimum temperatures are given in figures 5,6 and 7 (information is yet to be analyzed for Springbrook). Rainfall for the three other plantations is shown in figures 8,9 and 10. The average yearly temperatures, rainfall and number of rainy days are listed in table 7. Springbrook has the greatest number of cloudy days and the highest annual rainfall. Peats Ridge has the greatest number of sunny days and the lowest annual rainfall of 1207 mm.

During 1989, rainfall was heavy, especially in autumn (March). Monthly maximum temperatures were highest at Peats Ridge and lower at Kurrajong Heights than at Robertson, because Kurrajong Heights is farther from the ocean. Both Robertson and Kurrajong Heights have relatively cool winter minimum temperatures.

Table 3 - Soil properties of four *Protea* plantations.

Site	pH Water	pH $CaCl_2$	Elec. Cond.	Moist. Con(g/g)	Bulk dens.	% Sand	% Silt	% Clay	% O.M.
Springbk	4.8	4.2	99.9	.516	2.14	14	31	55	7.7
Peats Rd	5.3	4.3	19.6	.031	3.66	88	2	10	2.6
Kurr. Hts.	5.3	4.7	246.0	.224	2.99	39	25	36	10.6
Robertson	5.2	4.4	69.3	.716	1.96	52	17	31	11.5

Table 4 - Soil nutrient levels of four *Protea* plantations.

Site	CEC	%N	ppmP	ppmK	ppmMg	ppmCa	ppmNa	ppmFe	ppmMn
Springbk	65.2	0.11	15.8	74.3	64.7	210	30.2	70.5	5.5
Peats Rd	7.1	0.08	21.2	5.8	3.43	7.16	5.82	7.22	0.1
Kurr. Hts.	102	0.29	86.1	451	208	1313	35.2	13.6	10.8
Robertson	104	0.72	14.3	295	203	742	23.2	11.5	46.6

Table 5 - Flowering behavior of *Protea cynaroides* 'Long Leaf' 1990.

Site	Alt (m)	Mean Julian Days	First Bloom	Last Bloom	No. Flowers	Plants Flwrd	Ave. Stems/ Plant	Ave. Blooms/ Plant
Springbk	610	95	63	119	16	5	5.1	1.6
Peats Rd	280	80	56	119	45	17	7.4	2.0
Kurr. Hts.	580	-	113	-	18	12	3.4	0.8
Robertson	740	-	-	-	0	0	4.4	0

Table 6 - Flowering behavior of *Protea neriifolia* 'Salmon Pink' for 1988 and 1989 (Julian Days)

Site	First Bloom	Last Bloom	Mean of Blooms	S.E. Mean	No. of Flowers
1988					
Springbk.	77	234	124	3.21	112
Peats Rd	75	275	113	4.30	68
Kurr. Hts.	72	192	132	2.78	90
Robertson	192	293	232	5.81	26
1989					
Springbk.	55	255	93	1.78	296
Peats Rd	49	352	120	2.92	579
Kurr. Hts.	81	300	121	1.45	351
Robertson	88	190	141	1.86	157

Table 7 - Average yearly temperatures, rainfall and number of rainy days for four *Protea* planting sites. A: maximum mean °C; B: minimum mean °C; C: mean rainfall mm; D: mean no. of rainy days.

Site	Jan	Feb	Mar	Apr	May	Jun	Jul	Aug	Sep	Oct	Nov	Dec	Total
Springbrook													
A	25.6	25.4	24.6	22.8	19.9	17.7	17.1	18.3	20.3	22.5	22.4	25.6	22
B	17.3	17.2	16.1	14.2	11.3	9.2	7.9	8.6	10.4	12.9	14.8	16.2	13
C	436	472	482	254	235	191	152	97	95	173	215	260	3062
D	16	16	19	14	12	9	8	8	9	12	12	14	149
Peats Ridge													
A	26.3	25.6	24.4	22.2	18.1	15.2	15.3	16.8	19	21.7	23.5	26.4	21.2
B	16.3	16.4	15	12.2	9.4	6.9	5.6	6.6	8.5	11	12.7	15	11.3
C	131	169	138	84	113	119	52	73	52	88	87	101	1207
D	11	11	10	8	8	9	7	8	8	10	10	9	109
Kurrajong Heights													
A	23	22.3	20.3	16.7	13	10.1	9.1	11	13.9	16.9	20.2	22.2	16.6
B	12.7	12.8	11.5	8.8	6.1	3.7	2.4	3.3	5.1	7.6	9.9	11.6	8
C	162	173	169	122	103	125	88	78	73	93	101	125	1412
D	13	12	13	10	9	9	9	9	9	10	11	12	126
Robertson													
A	25.3	24.6	22.6	19.5	15.5	12.5	11.7	13.5	16	19.1	21.4	24.3	18.8
B	13	13.2	11.3	7.1	4.3	2.3	0.7	2	4.1	6.9	9	11.1	7.1
C	152	161	192	152	147	181	129	110	93	122	107	119	1665
D	12	11	12	10	9	10	9	9	9	10	10	10	121

4. Discussion

This study clearly shows that differences exist between flowering and bloom production at four field locations, of the clone *Protea neriifolia* 'Salmon Pink' and *Protea cynaroides*. Flower number is particularly variable between the four sites of coastal south-eastern Australia. The King Protea grows and flowers significantly differently at each plantation. Further data on stem size and time and blooming of flowers are needed to determine the reasons for this occurrence. The period of flower initiation is after a flush when new flower buds develop.

Protea neriifolia 'Salmon Pink' produces the best flowers at Peats Ridge, apparently because of a combination of the sunny, warm temperatures, excellent drainage characteristics of a sandy soil and low fertility levels. The cool temperature delays flower bud development at Robertson. This clone is productive at Kurrajong Heights but poor soil characteristics reduce the quality of the flowers.

Location of the plantation clearly determines the volume and seasonality of flowering of the two *Protea* species studied. Vegetative growth is more prolific in areas of warmer temperatures and greater pruning is therefore required.

Continued research will demonstrate more clearly the differences in flowering period, particularly in *Protea cynaroides*. With a greater understanding of the factors which influence root and shoot development, the number and quality of flowers can be predicted. Manipulation of flowering may be possible, especially by

delaying shoot development for buds to develop over winter and spring.

Acknowledgements

The authors gratefully acknowledge the assistance of E. Smith, V. Holubinskyj, M. Warmann, and C. Meltzer with the data analysis. Plants were grown in co-operation with R. and B. Flanders, M. Flockhart, D. Tranter and C. Walker. This research was funded by the Australian Rural Credit Development Fund.

References

Claassens, A.S,. 1986. Some aspects of the nutrition of proteas. Acta Hort. 185: 171-179.

Criley, R.A., Parvin, P.E. and Lekawatana, S., 1990. Flower development in *Leucospermum cordifolium* 'Vlam' Acta Hort. 264: 61-70.

Halevy, A.H., Criley, R.A., Besemer, S.T., Van de Venter, H.A., McKay, M.E. and Kawabatu, O., 1987. A comparison of flowering behaviour of *Strelitzia reginae* at four locations as affected by air temperature. Acta Hort. 205: 89-96.

Heinsohn, R.D. and Pammenter, N.W., 1988. Seasonality of shoot growth and flowering in the fynbos shrub *Protea neriifolia* cultivated in a summer rainfall area. S. Afr. J. Bot. 54 (5): 440-444.

Higgins, K.B., Lamb A.J. and Van Wilgen, B.W., 1987. Root systems of selected plant species in mesic mountain fynbos in the Jonkershoek Valley, southwestern Cape Province. S. Afr. J. Bot. 53: 249-257.

Jacobs, G. and Honeyborne, G.E., 1979. The relationship between heat unit accumulation and the flowering date of *Leucospermum* cv. Golden Star. Agroplantae 11: 83-85.

Lamont, B., 1986. The significance of proteoid roots in proteas. Acta Hort. 185: 163-170.

Parvin, P.E., 1986. Use of tissue and soil samples to establish nutritional standards in protea. Acta Hort. 185: 145-153.

Rourke, J.P. 1980. The Proteas of Southern Africa. Purnell & Sons Cape Town. Pp. 47-125.

Vogts, M.M., 1977. *Protea cynaroides* (L) L. Intensive cut flower production. Part II. Farming in South Africa No. B10/1977.

Vogts, M. 1982. South Africa's Proteaceae: know them and grow them. C. Struik P/L. Cape Town 82-227.

Figure 1 - Average vegetative growth for each month of 20 *Protea cynaroides* plants at Peats Ridge, N.S.W.

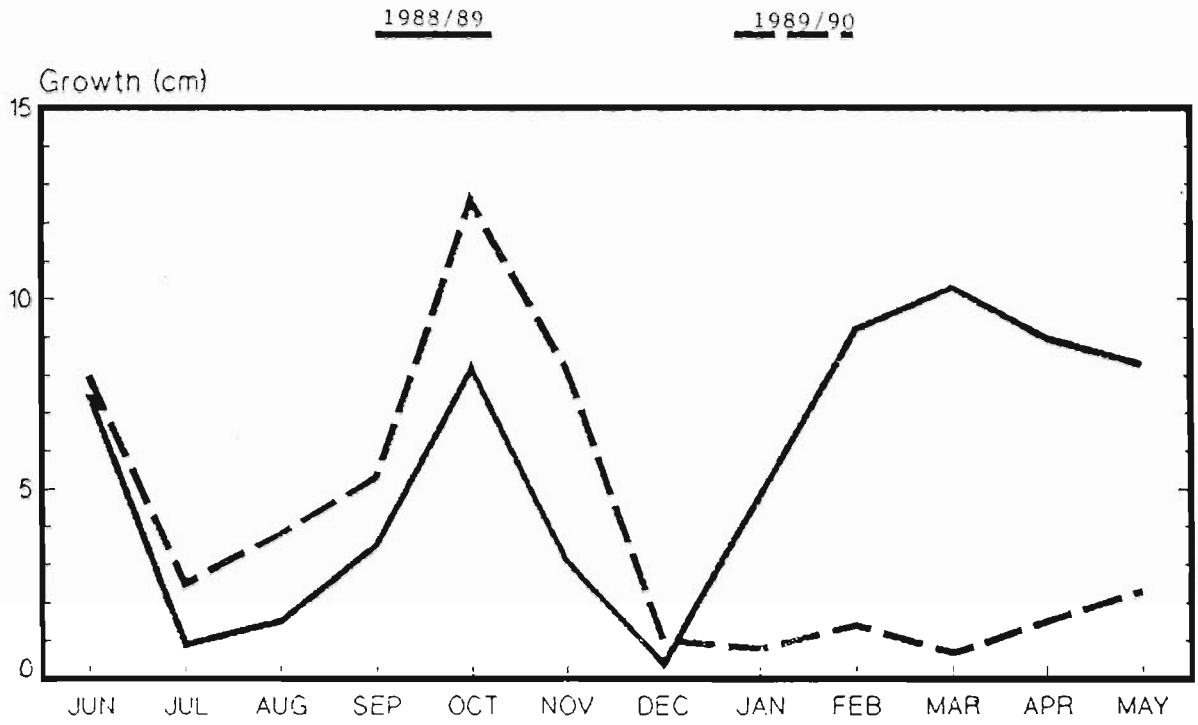


Figure 2 - Average vegetative growth for each month of 20 *Protea neriifolia* plants at Peats Ridge, N.S.W.

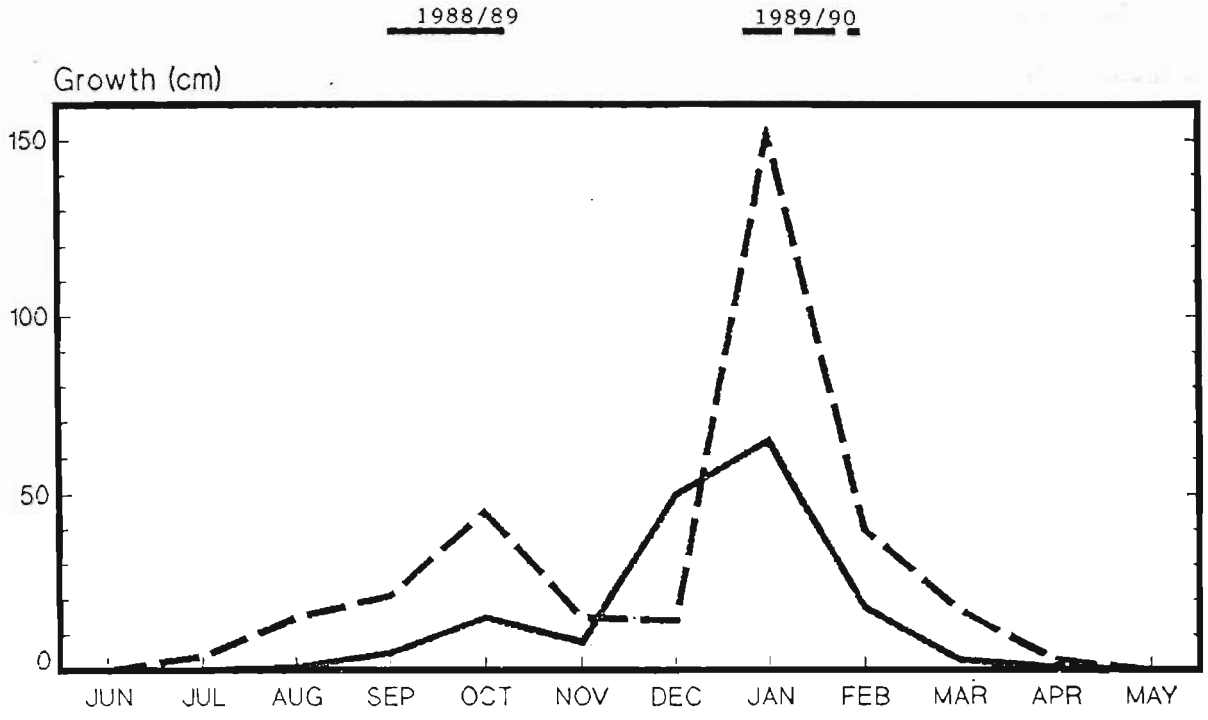


Figure 3 - Flowering behaviour of *Protea neriifolia* 'Salmon Pink' - 1988

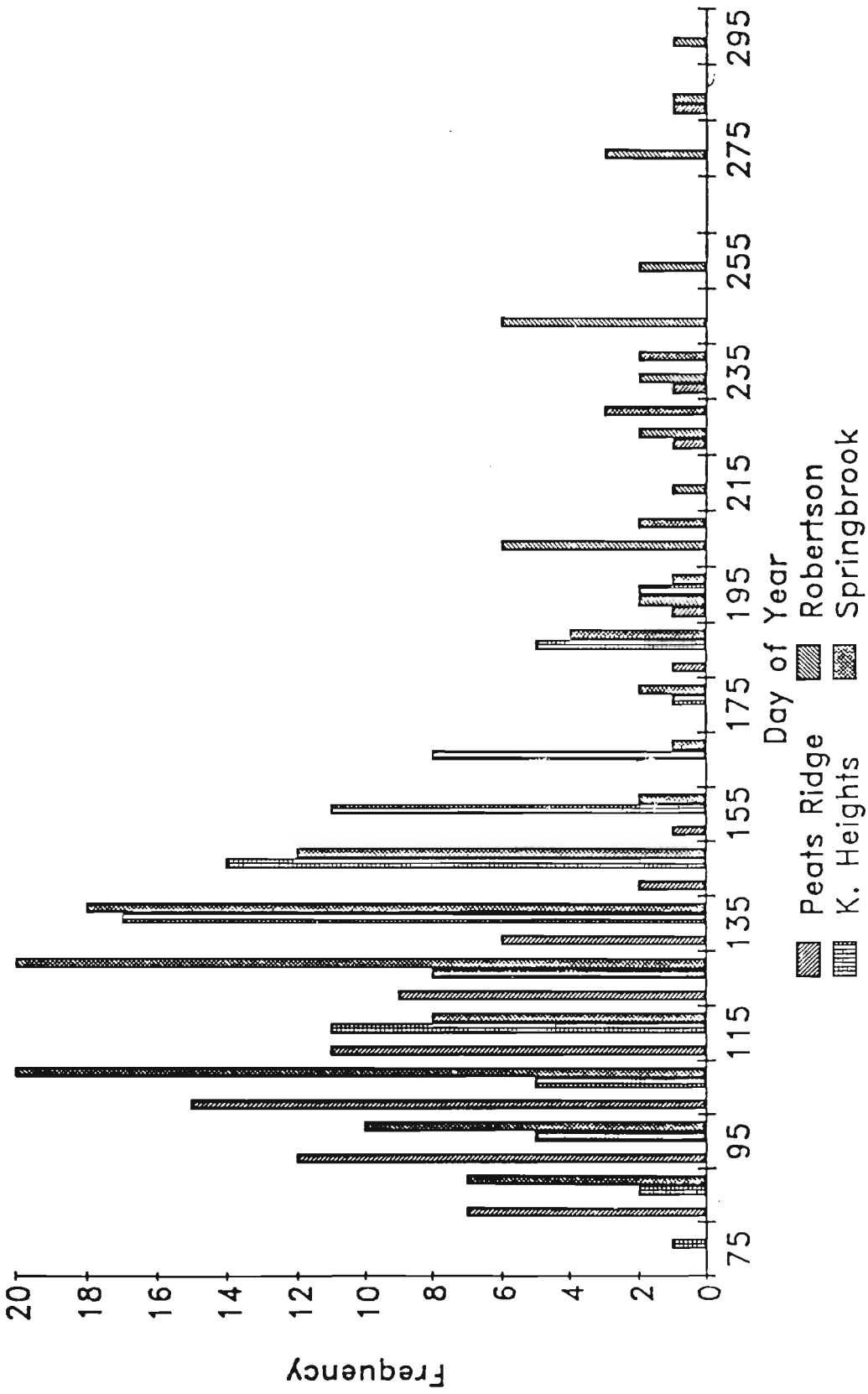


Figure 4 - Flowering behaviour of *Protea neriifolia* 'Salmon Pink' - 1989

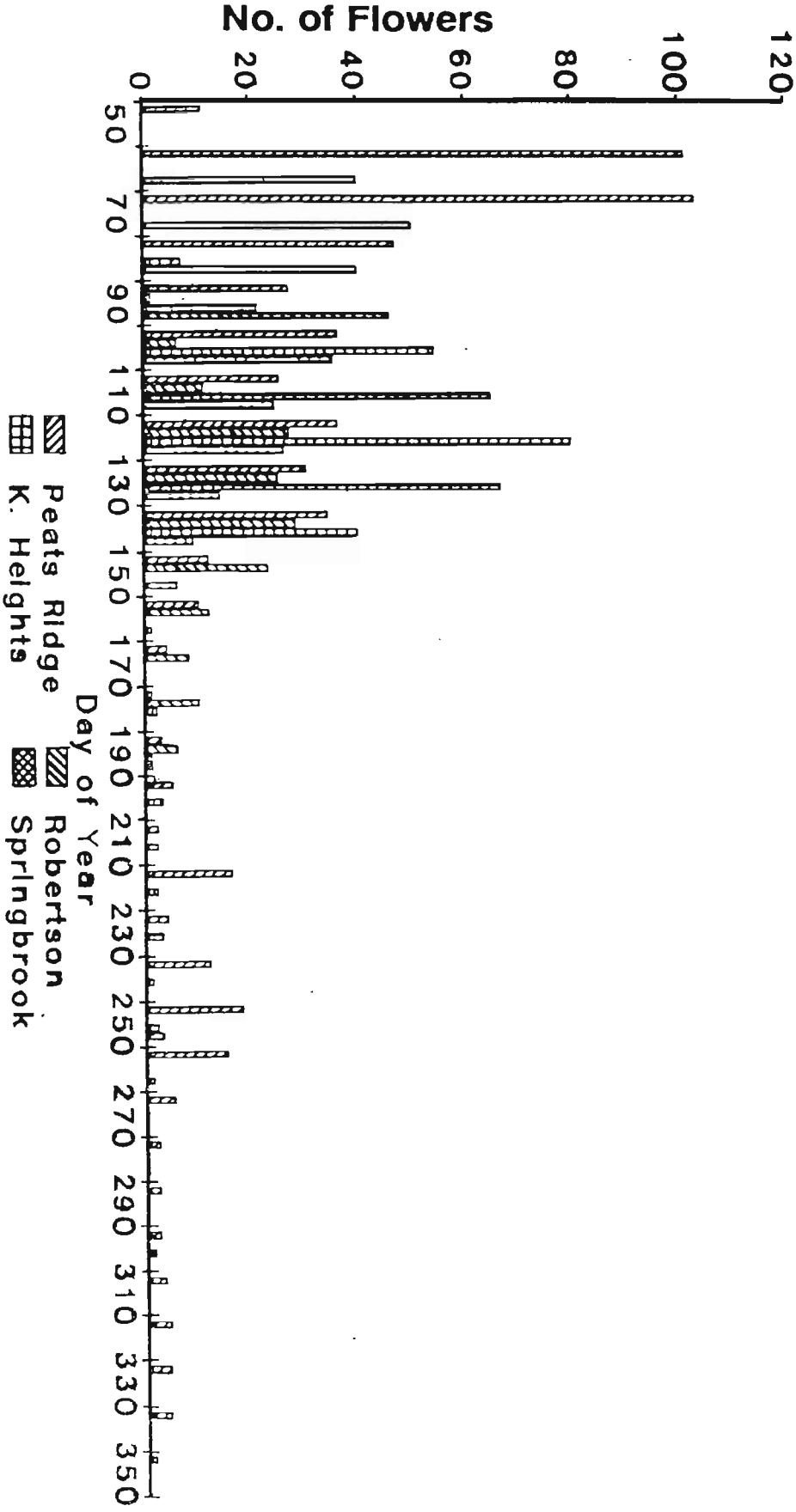


Figure 5 - Meteorology for Peat's Ridge 1989 - temperature

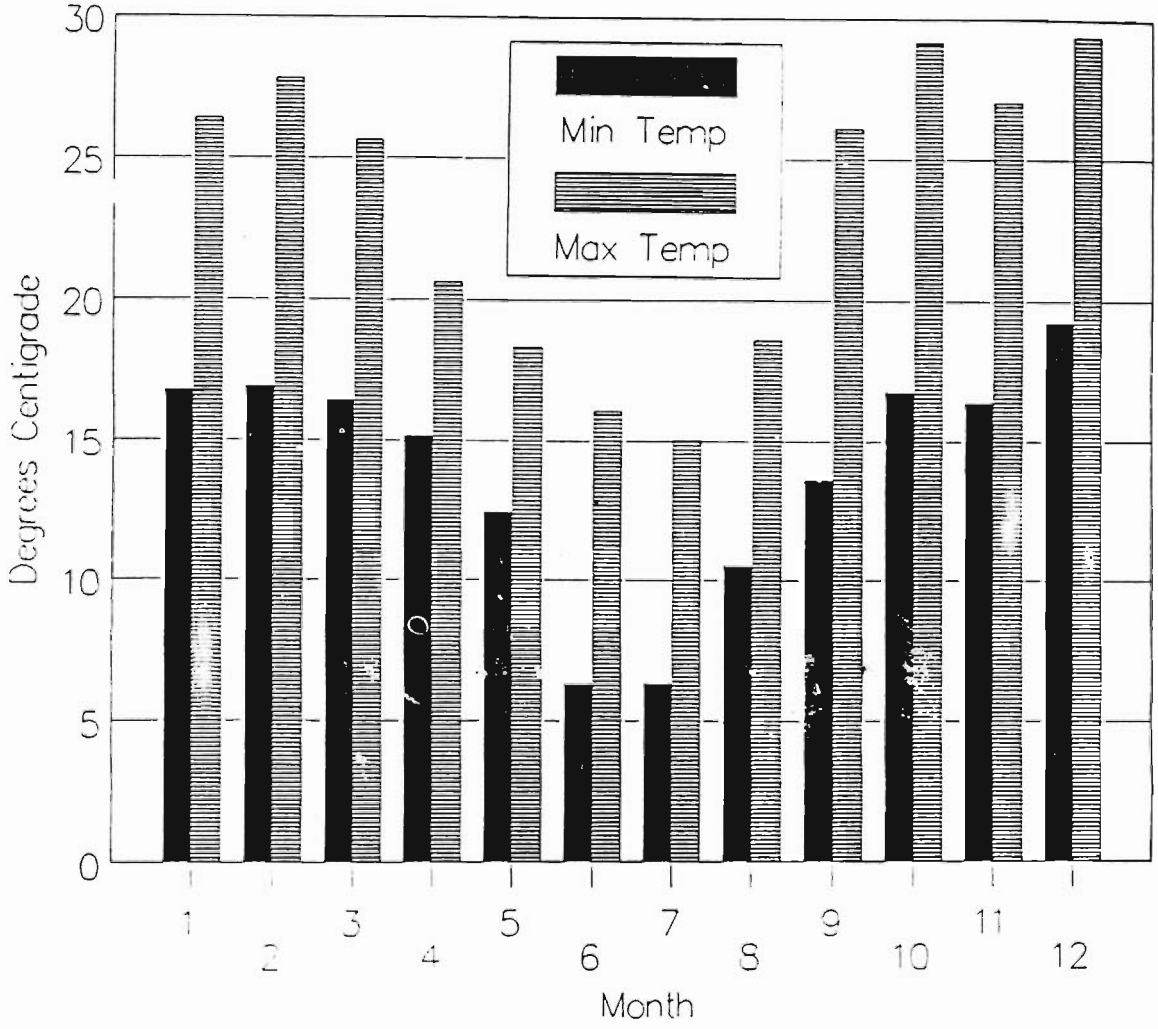


Figure 6 - Meteorology for Kurrajong Heights 1989 - temperature

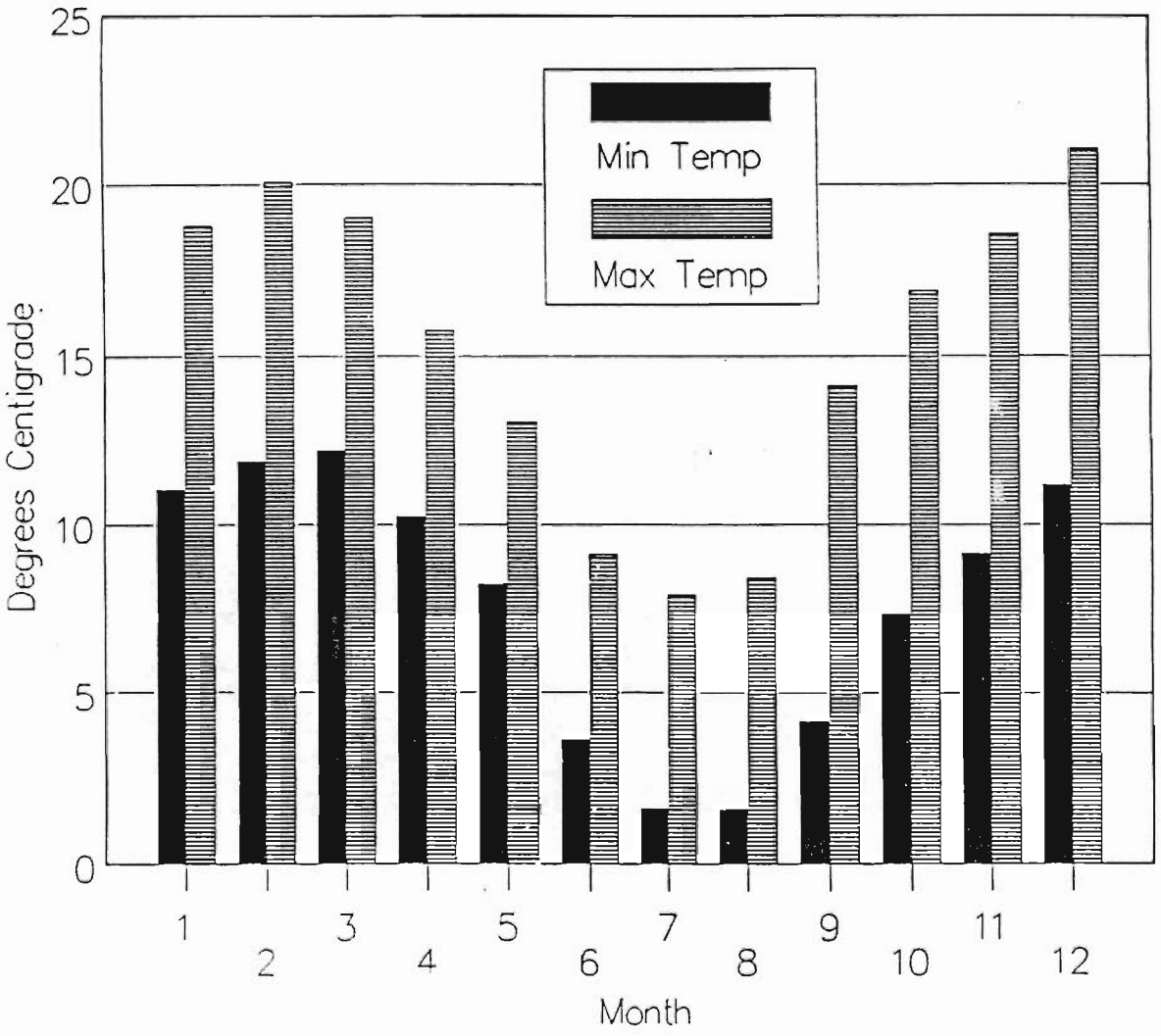


Figure 7 - Meteorology for Robertson 1989 - temperature

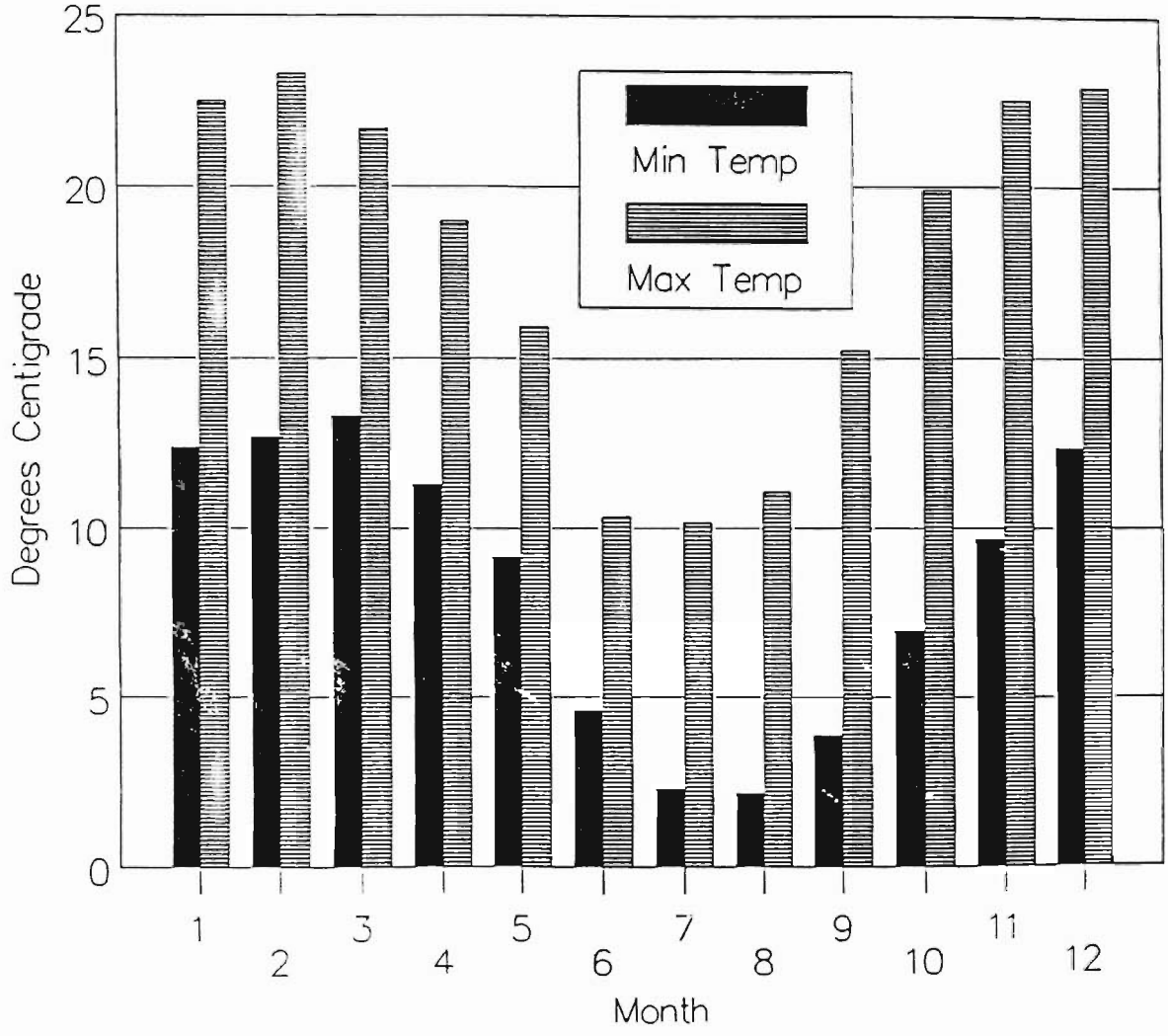


Figure 8 - Meteorology for Peat's Ridge 1989 - rainfall

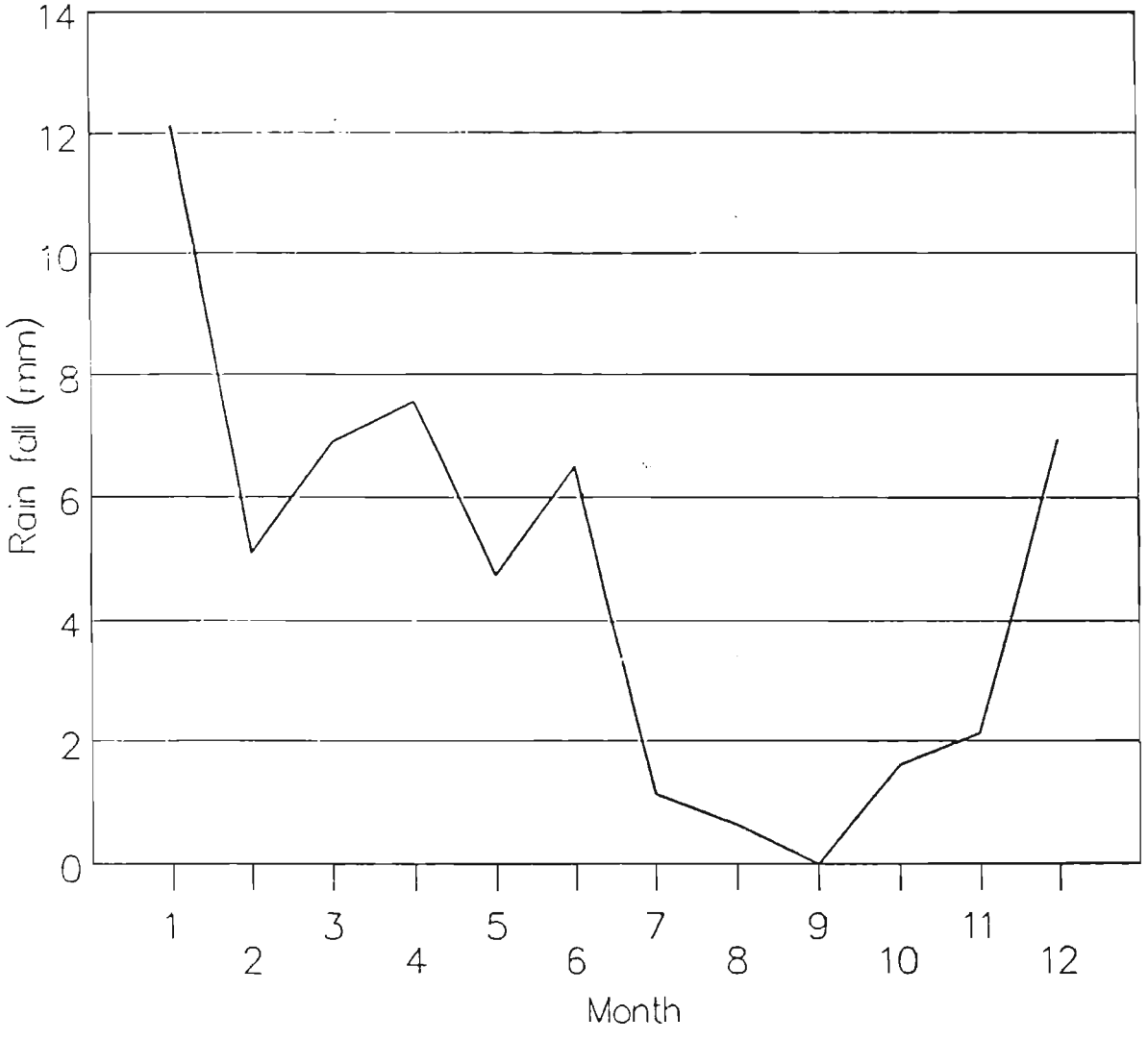


Figure 9 - Meteorology for Kurrajong Heights 1989 - rainfall

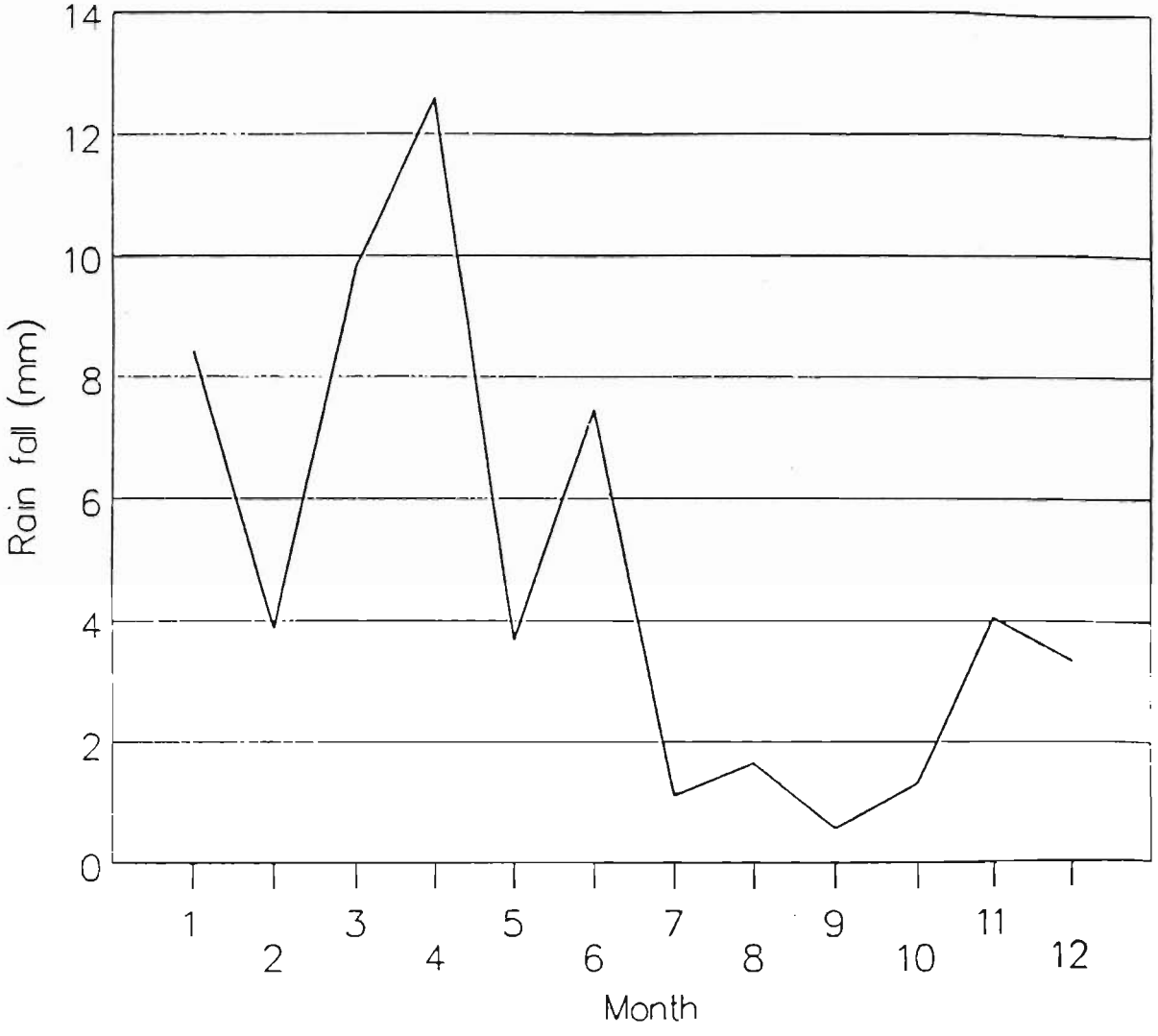
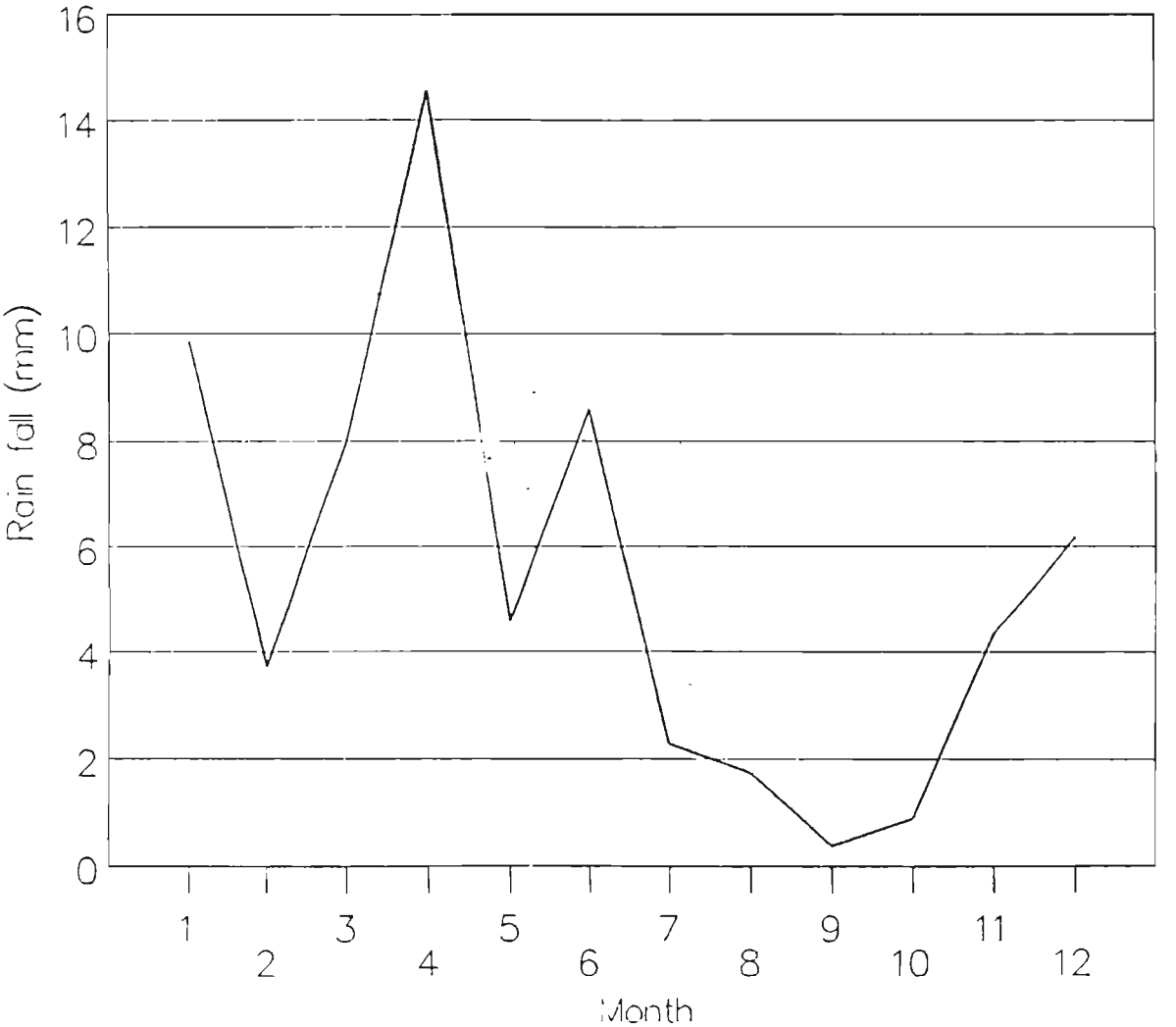


Figure 10 - Meteorology for Robertson 1989 - rainfall



EFFECT OF GIBBERELLIC ACID ON SHOOT GROWTH OF *LEUCOSPERMUM* R.BR.

Daniel G. Malan
Fynbos Research Section
VOPRI, Private Bag
Elsenburg 7606
South Africa

Gerard Jacobs
Department of Horticulture
University of Stellenbosch
Stellenbosch 7600
South Africa

Abstract

The commercial value of a hybrid *Leucospermum* selection (probably a cross between *L. conocarpodendron* x *L. cordifolium*) is limited due to inherently short flower stems. A single spray with gibberellic acid (GA_3) at 500 mg.dm^{-3} 10 to 12 weeks after pruning, when new axillary shoots were 100 to 170 mm long, resulted in marked increase in the number of shoots longer than 30 cm, as compared to the control plants.

1. Introduction

Leucospermum conocarpodendron has short flower stems (Rourke, 1972). This characteristic is transmitted to some of the progeny of *L. conocarpodendron* x *L. cordifolium* and limit the commercial value of such hybrid selections. Napier *et al* (1985) reported that the shoot growth of a *L. conocarpodendron* x *L. cordifolium* hybrid was stimulated by gibberellic acid (GA_3). However, the cost of GA_3 made the spray treatment uneconomical.

This paper reports on the concentration, the minimum number of applications and the optimal time of GA_3 application, required to reach acceptable production levels of marketable flowers to warrant commercial cultivation of a *L. conocarpodendron* x *L. cordifolium* hybrid.

2. Materials and Methods

The experiments were carried out in a 3-year-old clonal plantation of a *L. conocarpodendron* x *L. cordifolium* hybrid on the farm Protea Heights, in the Stellenbosch district of the Republic of South Africa. The climate is characterised by cool, wet winters and hot, dry summers. The annual rainfall varies between 600 to 700 mm. The plants were spaced 0.75 m apart with 1.75 m between rows, were clean cultivated and were not irrigated or fertilized.

The plants were pruned on 20 August (late winter, southern Hemisphere). Except for four or five vigorous shoots, all shoots were removed at the point of inception. The remaining shoots were topped to leave a short stump of ca 200 mm. All new shoots which developed from axillary buds on these stumps were allowed to grow. Plants were sprayed with a water solution of GA_3 1 g/10 g) to which Agral^R (1 drop per litre) was added as a wetting agent. Plants were sprayed to drip-off with a handspray.

GA_3 concentration and number of applications: GA_3 was applied in multiples of 100 mg.dm^{-3} from 100 to 700 mg.dm^{-3}. Plants were treated once (on 21 August), twice (on 21 August and 20 Sept.) or three times (on 21 August, 20 Sept. and 22 Oct.). Unsprayed plants were included as a control. The main treatment (GA_3 concentration) was replicated four times using nine plants per treatment. Sub-treatments (number of applications) were applied to three plants in a split plot

design.

Shoot length was measured on 15 May the following year when inflorescences had started to develop. All the shoots on two of the bearers of each plant were measured. One shoot per plant, of which the length was close to the average shoot length for the plant, was selected on 30 June and the number of nodes per shoot determined by counting the number of bract-like leaves at the base of the shoot and foliage leaves. The average internode length was determined by dividing the shoot length by the number of nodes.

Time of GA_3 treatment: A single GA_3 (500 $mg.dm^{-3}$) spray was applied to plants 6, 8, 10, 12 or 14 weeks after pruning. Untreated plants were included as controls. The length of the longest shoot on each plant was measured on the day the plants were sprayed. Treatments were replicated 5 times with 2 plants per treatment in a randomized complete block design.

Shoot length was measured on 15 May. On 30 June one shoot was selected as described previously. The shoot length and the number of nodes per shoot were determined. The area of each leaf was measured on a control shoot and on shoots treated 10 or 14 weeks after pruning, using a portable area meter (Lambda Instruments Corp., Model LI-3000).

3. Results and Discussion

Shoot length increased linearly with increasing GA_3 concentration (figure 1). The rate of increase was greatest with three GA_3 applications (figure 1). Node number increased linearly with GA_3 concentration (figure 2) and number of spray applications (figure 3). The increase in shoot length resulted primarily from the increase in node number (figure 2). Internode length was affected only slightly (data not presented). The number of marketable shoots longer than 30 cm increased linearly with concentration (figure 2) and the number of GA_3 sprays (figure 3).

The optimum time for a single GA_3 application was 10 weeks after pruning when the longest shoots on the plants were approximately 10 cm long (table 1). Leaves on basal nodes on shoots of *L. conocarpodendron* x *L. cordifolium* were small and reached a maximum size at nodes 15 to 20. At higher node positions leaf size decreased again (figure 4). When GA_3 was applied 10 weeks after pruning more nodes with intermediate size leaves were formed (figure 4) which accounted for the increase in shoot length. Delaying GA_3 spray treatment until 14 weeks after pruning stimulated the formation of a new shoot growth flush. This is indicated by the presence of an undesirable series of small leaves followed by a growth cycle bearing larger leaves (figure 4, figure 5). These shoots were shorter than shoots treated at 10 weeks (table 1).

We conclude that a single GA_3 spray concentration not lower than 500 $mg.dm^{-3}$ is adequate to obtain economically viable flower yields on this *L. conocarpodendron* x *L. cordifolium* hybrid. The spray should be applied 10 to 12 weeks after pruning, when the largest shoots on the plant vary from 100 to 700mm. Longer shoots can be obtained with higher and/or more GA_3 applications, but would not be economically viable.

Acknowledgements

The South African Nature Foundation and the South African Protea Producers and Exporters Association are thanked for financial support.

References

Napier, D.R., Malan, D.G., Jacobs, G. and Bernitz, J.W., 1985. Improving stem length and flower quality of *Leucospermum* with growth regulators. Acta Hort. 198: 67-74.

Rourke, J.P., 1972. Taxonomic studies in *Leucospermum* R. Br. Cape Town and Transvaal Printers, Cape Town, South Africa.

Table 1 - Effect of GA_3 (500 mg.dm^{-3}) treatment, at different intervals after pruning, on the shoot characteristics of *L. conocarpodendron* x *L. cordifolium* hybrid. Values are the average of 5 replicates with 2 plants per replicate.

Date of Application	Weeks after pruning on day of treatment (cm)	The length of longest shoot/plant length (cm)	Final average shoot	Number of nodes/ shoot bearers	Number of shoots longer than 30 cm/2
Control	22,42a	39,1a	1,1a[z]		
3 Oct.	6	3,64	25,03ab	41,2a	3,5a
16 Oct.	8	5,09	24,23a	40,3a	3,0a
30 Oct.	10	10,41	34,33c	46,5b	10,3b
13 Nov.	12	17,00	33,13c	58,6c	8,5b
27 Nov.	14	21.70	29,52bc	63,4c	9,2b

[z]Values in a column not followed by the same letter differ significantly. Tukey's H-test (P 0,05)

Figure 1 - Effect of GA_3 at different concentrations applied 1, 2 or 3 times to plants, on shoot length, of a clonal selection of a *L. conocarpodendron* x *L. cordifolium* hybrid. Values are the average of 4 replicates with 3 plants per treatment. : 1 application, []: 2 applications, ... : 3 applications.

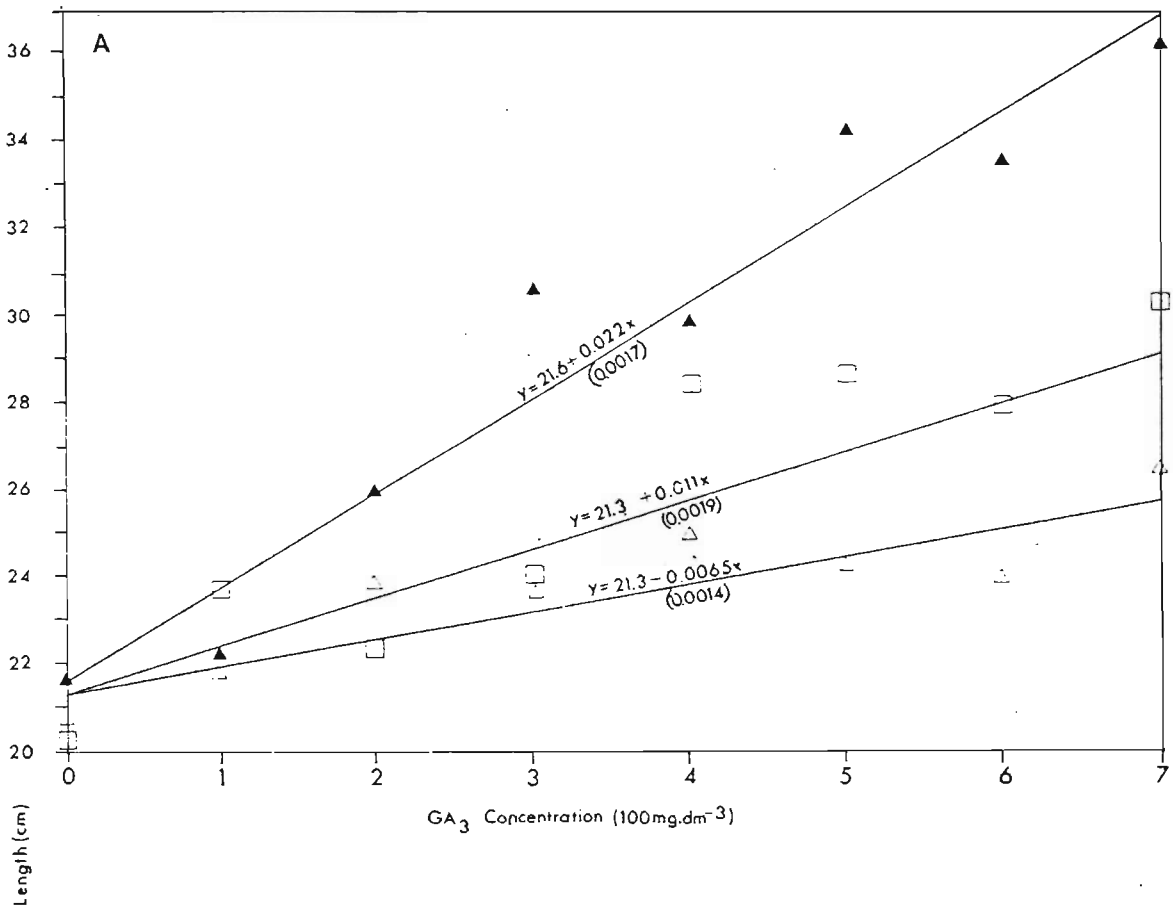


Figure 2 - Effect of GA_3 spray concentraction on the number of nodes per shoot and the number of marketable flower stems of a clonal selection of a *L. concocarpodendron* x *L. cordifolium* hybrid. Values are the mean of 9 plants.

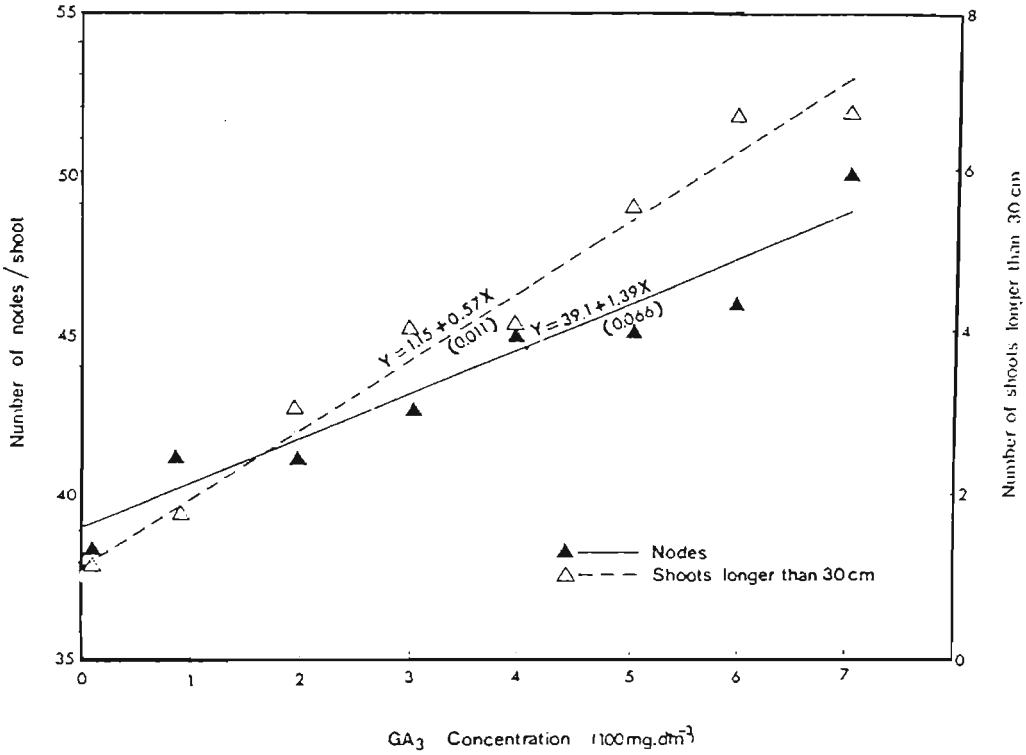


Figure 3 - Effect of number of GA_3 applications on the number of nodes per shoot and number of marketable flower stems of a clonal selection of a *L. concocarpodendron* x *L. cordifolium* hybrid. Values are the mean of 9 plants.

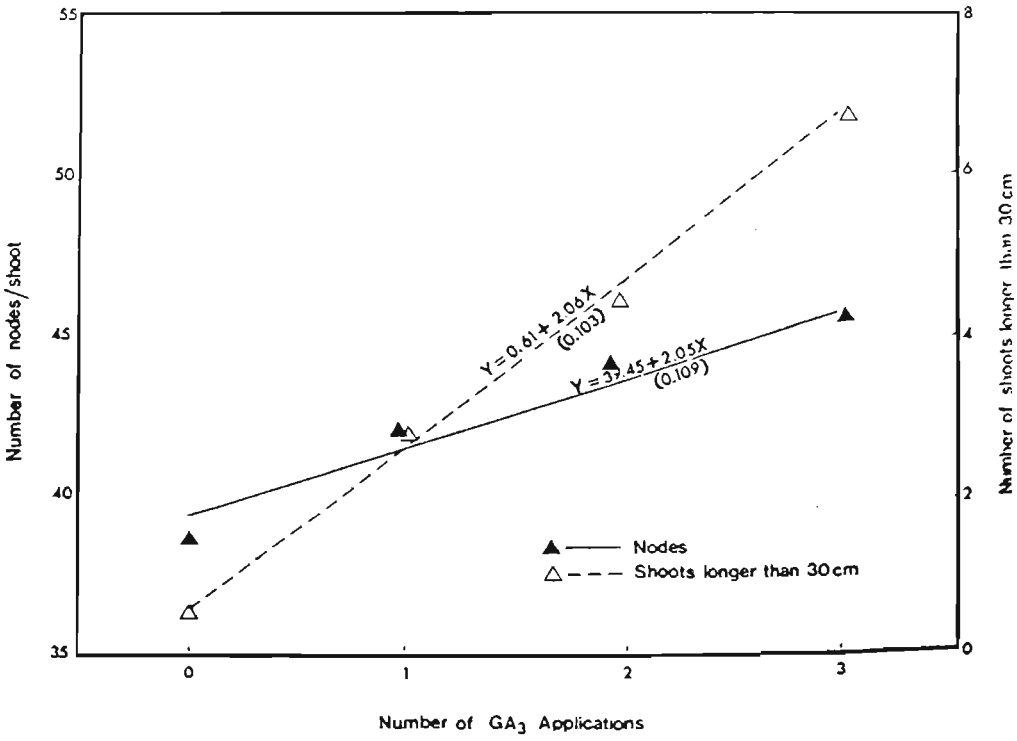


Figure 4 - The leaf area per leaf in relation to node position on a normal shoot of a *L. conocarpodendron* x *L. cordifolium* hybrid and of shoots treated with GA_3 (500 mg.dm^{-3}) 10 and 14 weeks after pruning.

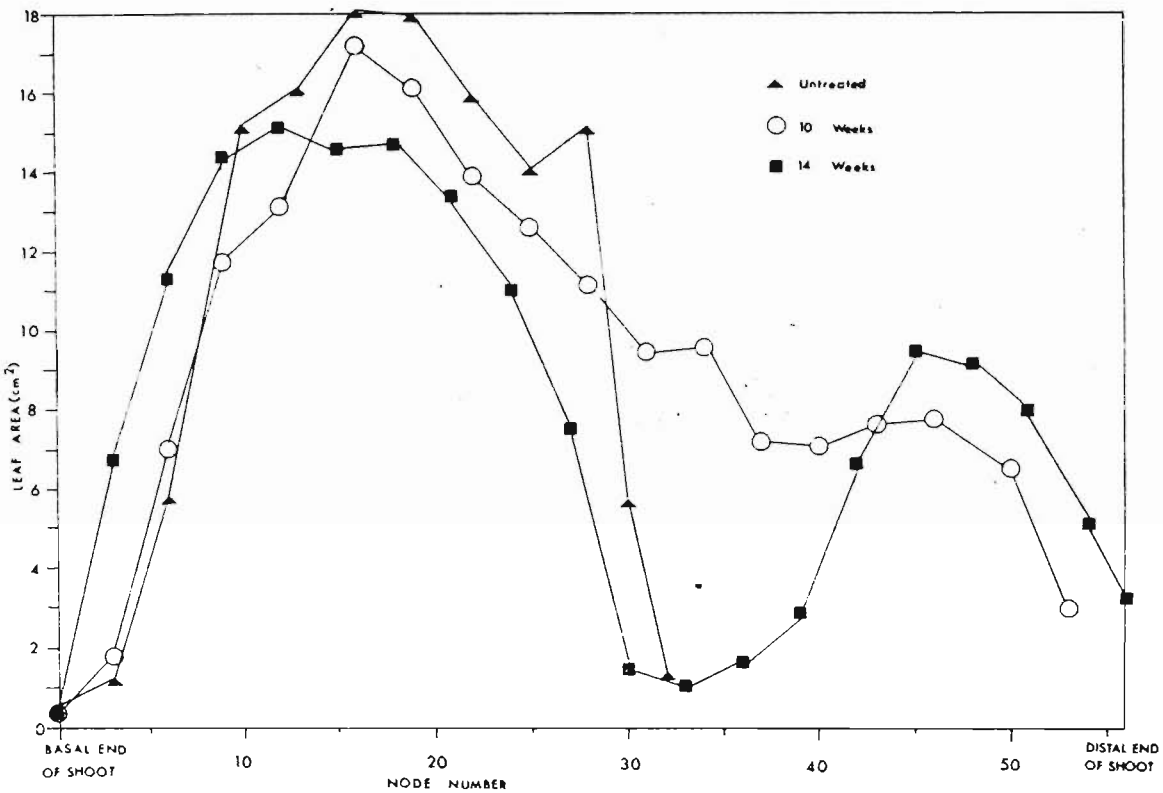


Figure 5 - *L. conocarpodendron* x *L. cordifolium* shoot illustrating the "bare neck" and rosette-like growth produced in reaction to GA_3 (500 $mg.dm^{-3}$) treatment 14 weeks after pruning

COOPERATIVE PRODUCTION OF PROTEA FLOWERING POT PLANTS SELECTED FOR RAPID PRODUCTION

G.J. Brits
Fynbos Research Unit
VOPRI
Private Bag
7607 Elsenburg
South Africa

E. Tal
A.R.O. Northern R & D
Misgav Experimental Station
Mobile Post Misgav 20-179
Israel

J. Ben-Jaacov and A. Ackerman
Dept. of Ornamental Horticulture
A.R.O., The Volcani Center
P.O. Box 6
Bet Dagan 50250
Israel

Abstract

An overview of current production technology and likely future research developments is presented. A newly developed technique is described to produce a well-branched *Leucospermum* cutting on the mother plant in which the side shoots have both a good wide angle of growth and the desired thickness to produce a flower. Branched cuttings harvested during flower initiation resume flower development after rooting, producing flowering plants after 6-8 months. This "rapid production" technique requires material selected specially for propensity to flower following rooting. A series of Proteaceae clones has been identified which yield a high percentage superior flowers when rapidly produced. These include selections of *Leucospermum* with large heads (*L. cordifolium, L. lineare, L. tottum*); small, multiple heads (*L. mundii, L. oleifolium*); Leucadendrons (*L. discolor, L. salignum*); and Serrurias (*S. aemula, S. florida*); and especially interspecies F_1-hybrids of these. The potential for cooperative production of pot plants for the European autumn and winter markets is discussed.

1. Introduction

The concept of protea flowering pot plants originated ca. 1975, when it was noticed that many selections, when rooted from standard 20 cm cuttings, tended to produce apparently normal flowers (inflorescences) after rooting, within the same flowering period as untreated shoots. A selection programme was subsequently started by V.O.P.R.I. (Vegetable and Ornamental Plant Research Institute) to identify proteaceous items that showed a propensity to yield flowers following the rooting of cuttings (Brits, unpublished progress reports).

In further work this process of prompting autumn rooted cuttings of some of the woody, slow growing Proteaceae species to flower in the ensuing spring, was labeled "rapid production" (Ben-Jaacov, 1986). Flowering pot plant technology advanced significantly following the initiation by Israel of a commercial development programme for Proteaceae (Ben-Jaacov *et al*, 1989).

The process of rapid production basically entailed a) harvesting of cuttings during flower initiation (Ben-Jaacov *et al*, 1986; Brits, 1989b), resulting in the suspension of inflorescence development; b) rooting of cuttings and c) resumption of inflorescence development following rooting, which finally yielded a flowering, rooted cutting 6-8 months after harvesting (figure 1) (Brits 1989a).

In the early stages of production development the following parameters were studied, and corresponding selection criteria applied (Brits 1989b):

-type and size of inflorescence required
-flowering period of species and selections
-cutting type suitable for rapid production
-rootability of cuttings
-cutting ability to resume flower development following rooting
-semi-open lath house rooting system
-post-rooting handling of cuttings.

Very little has been published about the basic technological development of protea flowering pot plants. The main current as well as new production procedures are outlined below, with emphasis on research bottlenecks and potential future developments.

2. Type and size of inflorescence required

Protea shoots require a minimum thickness for a flower to develop (Brits, Jacobs & Steenkamp, 1986). *Leucospermum* "Caroline" shoots, for example, remain vegetative if thinner than ca. 3,5 mm (figure 2). In *Leucospermum* flower quality was shown to be correlated with shoot diameter (Jacobs & Minnaar, 1980; Napier, 1985). These results suggest that species normally capable of producing reasonably large flowers on relatively thin shoots could be more suitable for pot plant purposes, because plants will produce a flower even on a thin shoot - e.g. in *Leucospermum tottum*; also that species bearing conflorescences (multiple inflorescences grouped together terminallly) would be more suited, because even thin shoots will produce some flowers. These guidelines apply especially where branched cuttings are to be used. Branching treatments lead to the production of markedly thinner side shoots compared with the primary shoot.

Protea flowering types bearing smaller inflorescences were indeed found to be more practical to use as pot plants since they can be manipulated easier; they also appear more natural in the compacted, dwarfed format of a typical flowering pot plant.

Species flowering from axillary buds (e.g. *Leucospermum*, *Serruria*) are more suitable than those producing terminal inflorescences (e.g. *Protea*, *Leucadendron*). The reason for this is that the primary flower bud in rooted cuttings often aborts. In the case of the axillary flower bud type this problem can be overcome because the shoot produces secondary axillary flower buds which are inhibited but which resume their development upon deactivation of the primary bud (Jacobs & Honeyborne, 1978). In contrast an aborted terminal flower bud cannot be replaced.

3. Flowering period

Flowering period is extremely important in marketing. The season of optimal demand on European markets is that of autumn through winter (Sept. - March) with peaks around major holidays, e.g. Christmas and St. Valentine's Day. Flowering period of pot plant candidates is therefore an important selection criterion.

The rooting of cuttings delays the flowering of shoots by approximately 6-8 weeks, roughly the period required by the plant to establish a new root system. This is brought into account in the selection of pot plant types with a suitable flowering period (tables 1, 2, 3).

Other potentially useful methods to extend the flowering period of proteaceous plants include temperature regulation (selection of production areas at different altitudes) and the use of growth rooms, pruning practices (manual and chemical), light intensity and photoperiod regulation, as well as growth regulators.

4. Cooperative production in the southern and northern hemispheres

It would be useful to be able to extend the marketing period of flowering pot plants. Year round production would be the ultimate goal. The limitation of Proteaceae plants of strictly limited seasonal flowering periods can theoretically be overcome by means of daylength manipulations (Wallerstein, 1989; Malan & Jacobs, 1990). However the practicability of cooperative pot plant production in different hemispheres, utilizing naturally complementary flowering seasons (Parvin, Brits & Claassens, 1982) should also be investigated.

5. Cutting type

5.1 Complexity of cuttings

A flowering single-shoot protea cutting is apparently inadequate as a marketable product. Therefore primary shoots must be manipulated on the mother plant to branch in order to give an end product with a suitable degree of complexity. Alternatively multiple single-shoot cuttings, rooted separately and transplanted together in the same pot, is feasible (Tal, unpublished).

A technique has been developed to produce a well-branched shoot on *Leucospermum* "Ballerina" mother plants in which the secondary (side) shoots have both a good wide angle of growth and the desired thickness to produce a flower. Primary shoots are sprayed with ethephon 960 mg.l^{-1} during the early elongation stage of approximately 10 cm length. This results in the sprouting of an increased number of secondary shoots of shorter length and with a wider growth angle, compared with hand-pinched controls (table 4). The former is regarded as a more desireable shape for marketing. These results are consistent with the observed branching patterns of *Leucospermum* primary shoots from the terminal as opposed from subterminal shoot regions (Brits, Jacobs & Steenkamp, 1986).

5.2 Physiological stage of harvested cuttings

Cutting type also refers to the season of, or physiological development stage during harvesting. The importance of this factor in the flowering response of rooted *Serruria* cuttings is shown in table 3.

As was mentioned above, a problem commonly experienced with cuttings harvested during the flower initiation stage (semi-hardwood) is the abortion of the primary flower bud. Preliminary results suggest that the problem can possibly be overcome by harvesting cuttings earlier, during the active growth stage. This would allow rooting in late summer followed by normal initiation of flowers onto well-rooted cuttings (figure 3).

latter technique may require the heading back of softer, actively growing terminal shoot sections to harder parts subterminally (see "Cutting ability.." below). The scheme may require a slightly longer production period, of approximately 10 months

5.5 Compactness of branched cuttings

It difficult to control elongation growth of vigorous mother plants. It has been found that, in genera flowering from axillary buds, shoots headed back before or during flower initiation can initiate flower buds from undifferentiated axillary buds (e.g. in *Leucospermum* - Jacobs, 1983). It is therefore theoretically possible to head back undesireably long shoots in branched cuttings to the desired length. Branched primary shoots of *L.* "Ballerina" were indeed shaped on the mother plant in this way and were subsequently found to flower normally in pots (see "Cutting ability.." below).

A promising option appears to be the use of growth retardants to obtain a compact growth form (Ben-Jaacov *et al*, 1990). This could result in a) shorter shoots b) increased leaf number per unit length of shoot and c) increased thickness of shoots. Circumstantial evidence suggest that leaf number correlates positively with the ability of shoots to produce mature flower heads. Reducing internode length with growth retardants such as paclobutrazol, therefore, holds much promise for improving the cutting type required for successful production (see illustrative scheme - figure 4).

6. Cutting ability to resume flower development following rooting

Species and selections of proteas differ much in their ability to resume development of flowers following the rooting of cuttings. As was mentioned above, inflorescence development often is not only suspended but inflorescence buds are aborted as well. Some selections were identified which readily develop a secondary bud successfully into a normal flower. This tendency to resume inflorescence development, either from the primary or from secondary flower buds, is an important selection criterion for flowering pot plants (tables 1, 2 and 3).

Leucospermum "Ballerina" has an inherently strong propensity for flowering. It was found that even after repeated manipulations, this hybrid *Leucospermum lineare* could still develop a normal inflorescence. In one experiment the effects of sequential shoot tip pinching, shaping and BA application on "Ballerina" mother plants, followed by rooting, was compared with the effects of these treatments in *L.* "Tango". It was found that only "Ballerina" had the ability to tolerate sequential manipulations, resulting in approximately 80% of cuttings flowering in December as suitable flowering pot plants (figure 5).

7. Rooting ability and vigour of cuttings

Species and selections differ considerably in their ability to root in high percentages as required for pot plant production. Likewise the growth rate of roots, and subsequently of inflorescence buds, also differ. These differences are apparently genetic and relate to factors such as inbreeding depression and hybrid vigour (tables 1, 2 and 3).

8. Semi-open lath house rooting system

Economics dictate the use of the simplest possible system for the nursery production of flowering pot plants. The mild Mediterranean winter climates of the south-western Cape (and of Israel) and the xerophytic character of Proteaceae plants, allowed the development of a "semi-open" rooting system (Brits, 1986). Cuttings are rooted in a lath house or even in the open sun. An important further measure is the omission of bottom heating. Electricity accounts for the greater part of running expenses in conventional mist bed rooting of cuttings. Selection is currently therefore applied to find genotypes capable of rooting under relatively low ambient temperature (Brits, 1986).

Another important feature of semi-open rooting is the maximizing of light levels. Tal *et al* (1990) has demonstrated the requirement for relatively high light levels in rooting of micropropagated *Leucospermum*. The lowering of light levels can reduce flowering in *Leucospermum* (Napier & Jacobs, 1989). High lighting requirement thus probably underlies both successful rooting and flowering in pot plant cuttings. Early rooting, during the late summer season, as opposed to autumn, could therefore achieve the combined purposes of providing a longer rooting period as well as utilizing the relatively high ambient temperature of this season (Brits, 1986) - both these factors would be conducive to the rooting of cuttings without bottom heating. Early rooting will also utilize the high light levels of summer. New developments in flowering pot plant technology and breeding therefore presently concentrate on early rooting of cuttings under extensive conditions of minimum protection without bottom heating and under maximum natural light levels.

9. Post-rooting handling of cuttings

Very little progress has been made in evaluating planting media. These should be lightweight but at the same time have a sufficient water and cation retention capacity and must also comply with international phytosanitary requirements when used for the shipping of pot plants. Future research should also be directed towards effective fertilization of rooted pot plant cuttings. Results suggest that nutrient status (Claassens, unpublished) and light regime (Napier and Jacobs, 1989) co-determine the optimum physiological status of shoots for flower initiation and development.

The conditions required for air transport and shipping of potted plants, such as light and temperature, must be investigated. The shelf life of protea flowering pot plants has not been adequately studied.

10. Selection and breeding

Selection and breeding of specialized items, which can tolerate the various manipulations required in flowering pot plant production, is a basic strategy in the development of suitable items. Much of the present progress can be attributed to stringent selection procedures, for example of cv. "Ballerina" - see also tables 1, 2 and 3.

It appears that interspecies hybrids of proteas have an exceptional potential as flowering pot plants. These hybrids are not only usually highly heterotic but are also more floriferous and have an extended flowering period as well. This is indeed the first line of attack on the inherent problem of the slow growth rates of the woody proteaceous plants - which called for a "rapid production" technique in the first place.

11. Summary

Future developments in protea flowering pot plants depend on the resolution of the following bottlenecks:

1. Inefficient hybridization - interspecies crosses are needed for improvement of poor vigour and lack of floriferousness; and the creation of novelties.
2. Slow development rate of new cultivars - develop methods for rapid identification, evaluation and multiplication of the best selections, preferably by means of micropropagation.
3. Poor control over cutting production on mother plants:
   - improve branching methods of primary shoots;
   - increase compactness of branched shoots with growth retardants;
   - increase secondary shoot diameter with growth regulators;
   - optimize physiological condition of shoots for rooting and subsequent flowering.
4. Lack of technology for manipulating rooted cuttings:
   - study rooting parameters which influence flower quality and yield;
   - determine effective fertilization methods for improved quality;
   - study shipment parameters: media, temperature requirements, etc.;
   - increase the economy of production.
5. Poor knowledge of marketing - upgrade market testing of new products.

References

Ben-Jaacov, J., Shillo, R. & Avishai, M., 1986. Technology for rapid production of flowering pot plants of *Leucadendron discolor* Phill. & Hutch. Scientia Horticulturae 28: 379-387.

Ben-Jaacov, J., Ackerman, A., Gilad, S. & Shchori, Y., 1989. New approaches to the development of proteaceous plants as floricultural commodities. Acta Hort. 252: 193-199.

Ben-Jaacov, J., Ackerman, A., Gilad, S., Carmeli, R. & Barzilay, A., 1990. Development of *Leucospermum* as potted plant - control of vegetative growth with paclobutrazol on *L.* "Tomer". International Workshop on intensive protea cultivation, Neve-Ilan, Israel, Sept. 1990. Abstract.

Brits, G.J., 1986. The influence of genotype, terminality and auxin formulation on the rooting of *Leucospermum* cuttings. Acta Hort. 185: 23-30.

Brits, G.J., Jacobs, G. & Steenkamp, J.C., 1986. The pruning of proteas for cut flower production. Pamphlet Series Flowers and Ornamental Shrubs B.15, 1986. Printed by the Dept. of Agricuture and Water Supply, Pretoria, South Africa.

Brits, 1989a. Rapid production of Protea pot plants under winter lath house conditions. Annual Congress of the South African Society for Crop Production, Wild Coast, January 1989. Abstract.

Brits, 1989b. The production of protea pot plants. S.A. Nurseryman 8 (3): 42.

Jacobs, G., 1983. Flower initiation and development in *Leucospermum* cv. Red Sunset. J. Amer. Soc. Hort. Sci. 108: 32-35.

Jacobs, G. & Honeyborne, G.E., 1979. The relationship between heat unit accumulation and the flowering date of *Leucospermum* cv. Golden Star. Agroplantae 11: 83-85.

Jacobs, G. & Minnaar, H.R., 1980. Light intensity and flower development of *Leucospermum cordifolium*. HortScience 15: 644-645.

Malan, D.G. & Jacobs, G., 1990. Effect of photoperiod and shoot decapitation on flowering of *Leucospermum* 'Red Sunset". J. Amer. Soc. Hort. Sci. 115: 131-135.

Napier, D.R., 1985. Initiation, growth and development of *Leucospermum* cv. Red Sunset inflorescences. M.Sc. thesis, University of Stellenbosch, South Africa.
Napier, D.R. & Jacobs, G., 1989. Growth regulators and shading reduce flowering of *Leucospermum* cv. Red Sunset. HortScience 24: 966-968.
Parvin, P.E., Brits, G.J. & Claassens, A.S., 1982. Comparative flowering periods of South African Proteaceae a world apart. XXIst International Horticultural Congress, Hamburg, West Germany (1982). (Abstract).
Tal, E., Solomon, H., Ben-Jaacov, J. & Watad, A.A., 1990. Micropropagation of selected *Leucospermum cordifolium*: effect of antibiotics and GA_3. International Workshop on intensive protea cultivation, Neve-Ilan, Israel, Sept. 1990. Acta Hort.
Wallerstein, I., 1989. Sequential photoperiodic requirement for flower initiation and development of *Leucospermum patersonii* (Proteaceae). Isr. J. Bot. 38: 25-34.

Table 1 - *Leucospermum* selections evaluated for rapid production of pot plants: % rooting (14 weeks), % cuttings which flowered, and flowering period

Species	Flower type*	Identification	% root-ing	% in flower	Flowering period Aug	Sept	Oct	Nov
L. cordifolium	Single	T73 10 02	90	20				———
L. cordifolium	Single	T80 10 14	66	100				———
L. cordif. x L. tottum	Single	T80 06 10	100	5				—
L. cordif. x L. tottum	Single	T84 11 04	87	53				———
L. glabrum x L. tott. ("Scarlet Ribbon")	Single	T80 06 01	60	20				———
L. glabr. x L. lineare ("Tango")	Single	T84 01 16	100	70				———
L. lineare x L.tott. ("Ballerina")	Single	T76 01 16	90	100				———
L. mundii	Conflor.	T75 06 10	100	100				———
L. oleifolium	Conflor.	T75 06 13	100	30				———

*Single = solitary large inflorescence; conflor. (conflorescence) = multiple small inflorescences

Table 2 - *Leucadendron* selections evaluated for rapid production of pot plants: % rooting (14 weeks), % cuttings which flowered, and flowering period

Species	Identification	% rooting	% in flower	Flowering period			
				Aug	Sept	Oct	Nov
L.album	T77 01 06	75	-				
L.cordatum	T81 09 05	20	40	———	———		
L.discolor	T76 03 09 01	5	0				
L.discolor	T87 05 10	50	90			—	
L.discolor	T86 09 25	80	100			———	———
L.salignum	T77 07 11	±80	-				
L.salignum	T77 07 31	±80	-				
L.salignum	T77 07 34	±80	-				
L. laureolum x L.sal. ("Safari Sunset")	T84 07 09	100	20				———

Table 3 - Effect of rooting date on *Serruria* selections produced as pot plants: % rooting (14 weeks), % cuttings which flowered, and flowering period

Species	Identification	Rooting date	% rooting	% in flower	Flowering period			
					Aug	Sep	Oct	Nov
S.florida	Ex Protea Heights	87-03-19	97	70	———	———		
Do.	do.	87-04-15	95	40	—			
Do.	do.	87-05-08	91	0				
S.florida x S.rosea	T87 03 01	87-03-11	88	29	———	———	———	
Do.	do.	87-04-27	100	100		———	———	
S.florida x S.rosea	T87 03 03	87-03-11	86	29			———	
Do.	do.	87-04-27	66	66				———

Table 4 - Effect of primary shoot spraying with ethephon, and hand pinching,on average lateral shoot number, length (mm) and growth angle (degrees) in *Leucospermum* "Ballerina".

Treatment	Number	Length	Angle
Ethrel	7,3	106	37
Hand pinch	5,7	152	23

Figure 1 - Diagram of original concept of rapid production of flowering *Leucospermum* potted plants from rooted semi-hardwood cuttings.

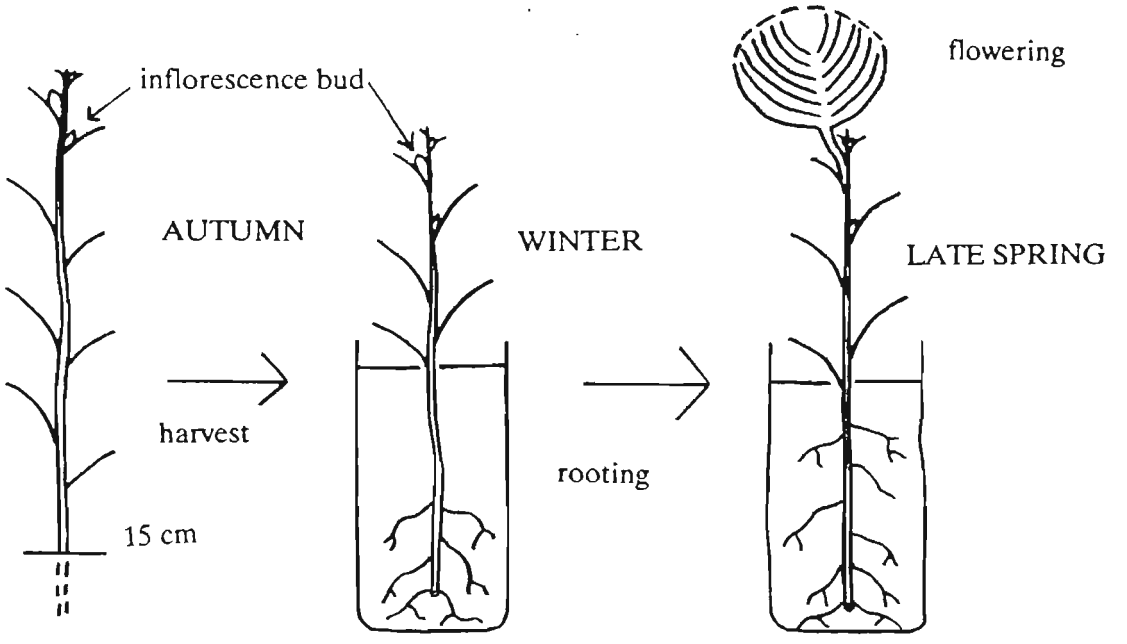
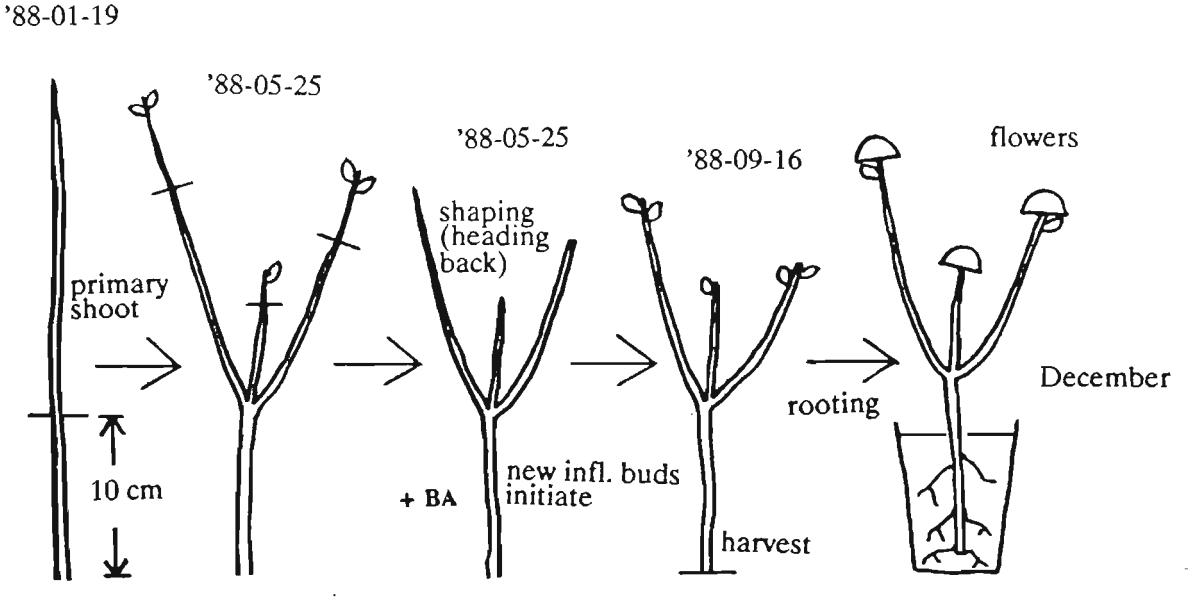


Figure 5 - Diagram of results of sequential manipulations performed on "Ballerina" (*Leucospermum lineare x L. tottum*) shoots, followed by rooting in early spring and resulting in branched pot plants flowering in December (southern hemisphere).



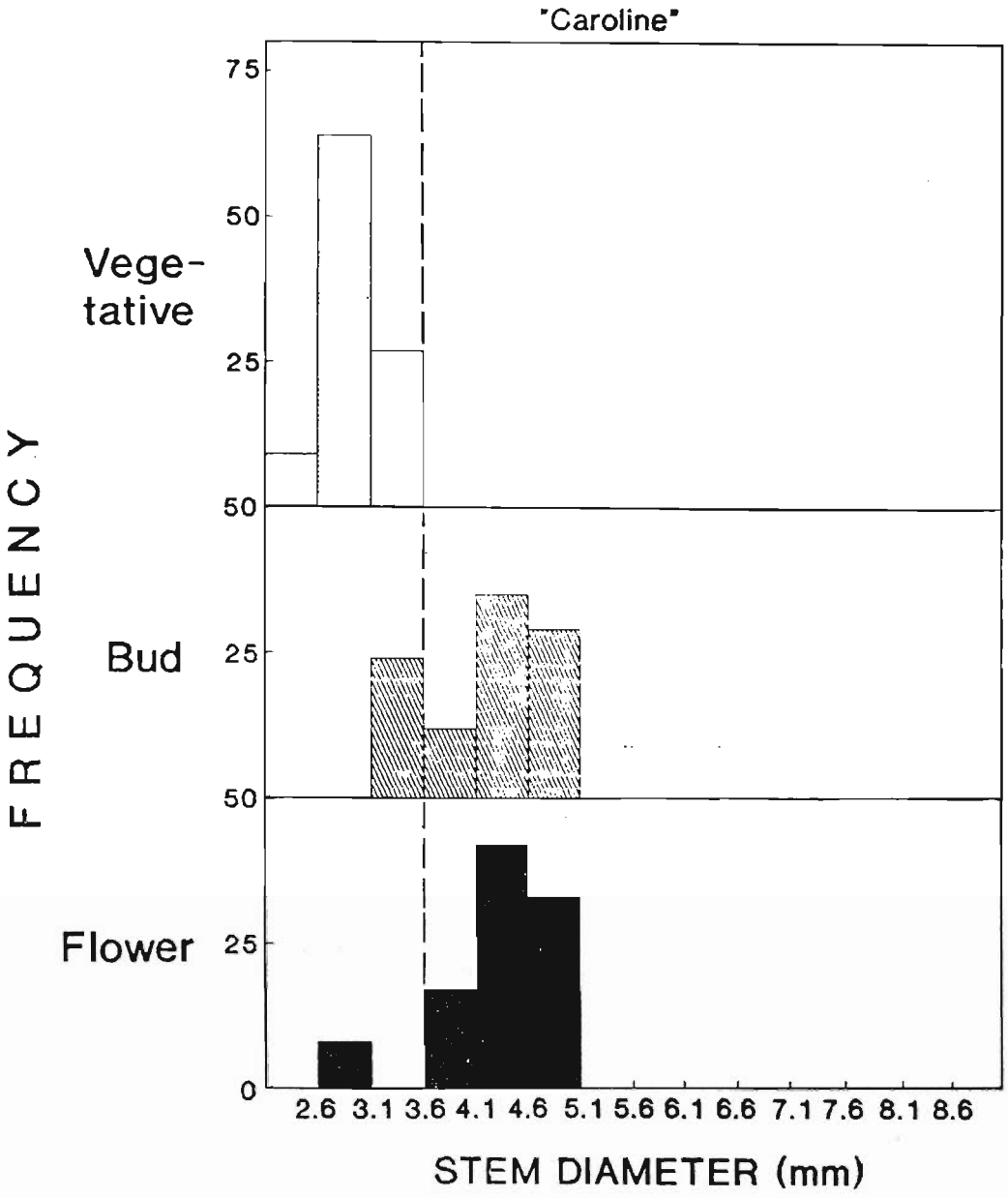
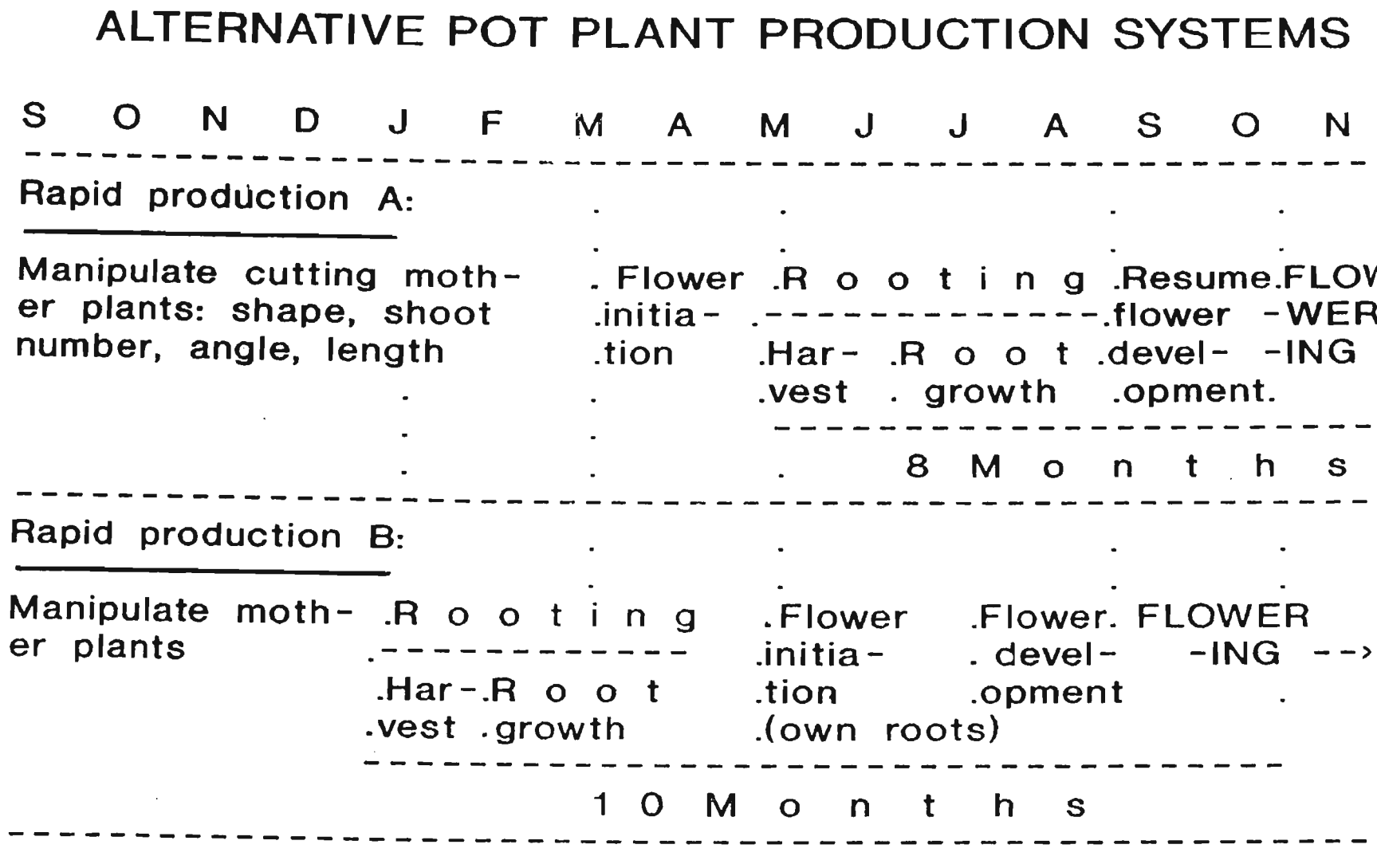


Figure 2– Percentage distribution of vegetative and flower stem diameters (buds or flowers) in *L. cordifolium x L. tottum* "Caroline" measured in autumn.

Figure 3 - Diagram of alternative seasonal rapid production systems for pot plants using cuttings harvested in different physiological stages (southern hemisphere).



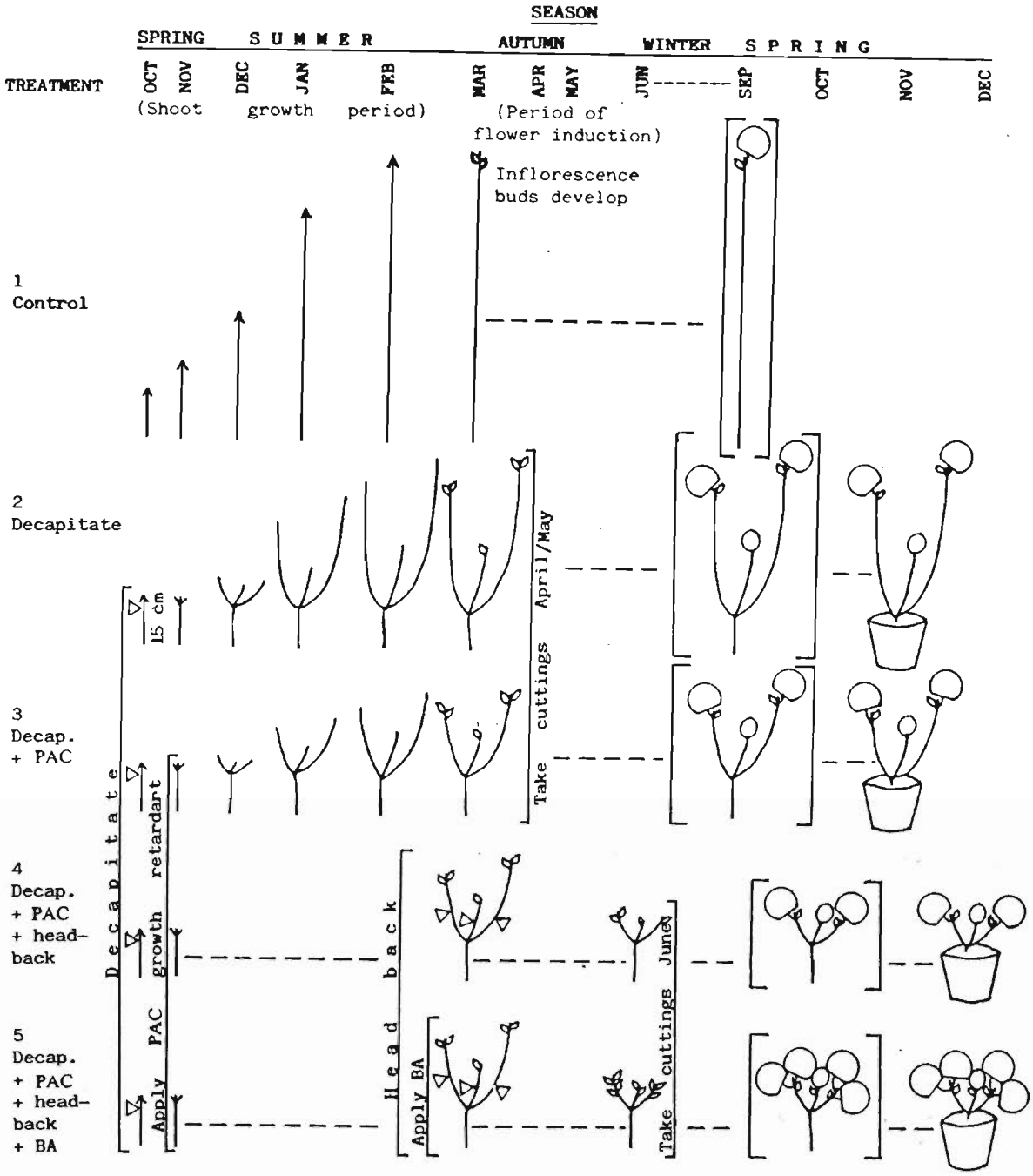


Figure 4 - Diagrams of basic production systems of *Leucospermum* and *Serruria* "rapid production" flowering pot plant techniques in southern hemisphere; seasonal manipulations are done on cutting mother plant primary shoots and on rooted cuttings and include, progressively, 1: control; 2: branching treatment; 3: growth retardation; 4: shaping of pot plant by heading back laterals; 5: benzyladenine treatment to increase no. of inflorescence buds in types bearing conflorescences; PAC = paclobutrazol.

ACTA HORTICULTURAE

Acta Horticulturae is used for publishing research and review articles.
Research reports of an oral or poster presentation should contain a commentary on a finished research project. A review article deals with knowledge obtained about a certain subject. Abstracts of copies of posters are not published.

As decided by the Council of ISHS, the proceedings of ISHS-symposia are published in a volume of Acta Horticulturae. The regulations for the publication of the proceedings are mentioned in a special Agreement, to be signed by the organizer of a symposium, by the institution involved and the Secretary-General of ISHS.
The ISHS-secretariat takes care of printing and publication.

The convener will be assisted by one or more editors, or by a scientific committee.
In case the organizers want to introduce refereed papers the authors should be informed in time about the system to be followed. In such a case the editorial board should be mentioned in the volume.

As to the financing of the proceedings reference is made to the special information for conveners.

Reprints

The orderform for reprints is enclosed with the writing-kit. The order of a minimum of 100 reprints can be forwarded to the ISHS secretariat. An invoice for the reprints will be sent to the authors.

Other ISHS Publications

- **Chronica Horticulturae,** the magazine of the Society which keeps members of ISHS informed of its activities and about other matters related to the objective of the Society.
- **Horticultural Research International,** the fourth edition which appeared in 1986, contains information on horticultural research in 63 countries, research institutes (some 1500), research workers (about 16650), main fields of interest, and an alphabetical list of research workers and places.
- **Proceedings of International Horticultural Congresses,** published by ISHS in Acta Horticulturae and obtainable from the ISHS secretariat.
- **Scientia Horticulturae,** published by Elsevier Publishing Company and sponsored by ISHS which has a co-responsibility for the appointment of the editorial board.
- **Elseviers Horticultural Dictionary,** a dictionary in 10 languages compiled by ISHS and published in close cooperation with Elsevier Publishing Company.

ISHS and Membership

The objective of the Society is the advancement of horticulture through improvement of international cooperation in science and technology. Besides through its members other means available to the society for the achievement of the objective are:
- the holding of **International Horticultural Congresses** at least every 4 years
- the setting up of:
- **Sections** dealing with groups of horticultural plants
- **Commissions** engaged in various scientific and technical aspects of horticulture
- **Working Groups** having interest in special horticultural science areas or activities within the Sections and Commissions.
- the organizing of **Symposia** on specific topics for scientists and other specialists
- the publishing of the proceedings of such symposia and other relevant publications.

The ISHS distinguishes three kinds of members:
- **Individual members** being natural persons engaged or interested in horticultural science or technology
- **Member organizations** being societies, associations, ministries, institutes, institutions, universities or departments of universities, organizations, agencies, firms or sections of any of these concerned with the advancement of horticulture.
- **Country members** being countries from where the annual dues for the country membership are paid and who nominate up to 3 individual members as members of the council, representing the scientific horticulture in the country concerned.

Advantages for members are:
- a reduced fee at each symposium organized by ISHS for each individual and for two representatives of a member organization
- the privilege to organize ISHS symposia or other scientific meetings (for organization members only)
- a free volume of Chronica Horticulturae per annum, containing 4 issues
- a discount on the price of all publications of Acta Horticulturae (20% for individual members, 30% for member organizations)
- a discount of 30% on the price of Horticultural Research International, HRI IV
- a discount of 10% on the subscription price of Scientia Horticulturae (for individual members only)
- a free copy of the list of members and of the directory of members of Sections, Commissions and Working Groups
- a reduced subscription fee for other ISHS publications.

The Society has 5 Sections, 9 Commissions and 63 Working Groups.
Members of Sections and Commissions are nominated by the national council members.
Members of Working Groups are nominated by the members of Sections and Commissions.

Section

- Fruit
- Vegetables
- Ornamental Plants
- Medicinal and Aromatic Plants
- Root and Tubercrops
- Viticulture

Commission

- Engineering
- Plant Protection
- Plant Substrates
- Urban Horticulture
- Protected Cultivation
- Post Harvest
- Education and Training
- Economics and Management
- Nomenclature and Registration
- Tropical and Subtropical Horticulture
- Biotechnology