Scaevola and Brachyscome flowering pot plants

Rodger Elliot
Outback Plants Pty Ltd

Project Number: NY01026

NY01026

This report is published by Horticulture Australia Ltd to pass on information concerning horticultural research and development undertaken for the nursery industry.

The research contained in this report was funded by Horticulture Australia Ltd with the financial support of Outback Plants Pty Ltd.

ISBN 0 7341 1182 7

Published and distributed by:
Horticultural Australia Ltd
Level 1
50 Carrington Street
Sydney NSW 2000
Telephone: (02) 8295 2300
Fax: (02) 8295 2399
E-Mail: horticulture@horticulture.com.au

HAL

Know-how for Horticulture™

Final Report for Horticulture Australia

Project No.: NY01026

Completion: 30/12/04

Project Title: Confidential *Scaevola* and *Brachyscome* flowering pot plants

Project Leader: **Rodger Elliot**

Principal Investigator: **Tony Slater**

Research Provider: **Department of Primary Industries, Victoria**

Victoria
The Place To Be

HAL
Know-how for Horticulture™

Final Report for Horticulture Australia

Horticulture Australia Project No: NY01026

Project Title: **Confidential *Scaevola* and *Brachyscome* flowering pot plants**

Project Leader: Rodger Elliot, Outback Plants

Principal Investigator: Tony Slater
Department of Primary Industries
Institute for Horticultural Development
Private Bag 15, Ferntree Gully Delivery Centre VIC 3156

Ph (03) 9210 9222
Fax (03) 9800 3521

Report Prepared by: Mary-Anne Blakemore and Tony Slater
Department of Primary Industries
Institute for Horticultural Development
Private Bag 15, Ferntree Gully Delivery Centre VIC 3156

Project Officer: Samantha Jones

Research Provider: Department of Primary Industries Victoria

Acknowledgement: The authors wish to fully acknowledge the funding support from Outback Plants, Horticulture Australia and the Department of Primary Industries Victoria.

Date: December 2004

Contents

Media Summary

The international market for flowering pot plants is always looking for new product lines and a large number are released each and every year. For a number of years Australian native plants have been grown as container plants and flowering pot plants for the European, Asian and American markets. *Scaevola* and *Brachyscome* are two genera that have gained significant portions of these markets. A limitation to the expansion of these two plant products in the markets is the limited colours that are currently available.

Recently, new colour forms have been identified that could be used in breeding programs to greatly expand the colour lines that could be developed for these products. Breeding programs for *Scaevola* and *Brachyscome* were designed to increase the range of colours and forms from available parent material. The reproductive and floral biology of these genera were examined to enable the reproductive barriers that currently exist to be overcome.

Technical summary

The international market for flowering pot plants is always looking for new product lines and a large number are released each and every year. For a number of years Australian native plants have been grown as container plants and flowering pot plants for the European, Asian and American markets. *Scaevola* and *Brachyscome* are two genera that have gained significant portions of these markets, and there are 18-20 million units of each genera sold annually. A severe limitation to the expansion of these two genera in the markets is the limited colours that are currently available for these products.

Recently, new colour forms have been identified that could be used in breeding programs to greatly expand the colour lines that could be developed for these products. Breeding programs for *Scaevola* and *Brachyscome* were designed to diversify the range of colours and forms from available parent material. The reproductive and floral biology of these genera were examined to enable the reproductive barriers that currently exist to be overcome. Once these barriers are removed, the establishment of a market orientated breeding program to produce diverse flower colours and forms would provide the opportunity to increase the markets for these crops.

The *Scaevola* breeding program examined the floral biology of the species and cultivars included. *Scaevola* possess a secondary pollen presentation system and strong self-incompatibility. Understanding the timing of emasculation and pollination were critical to the success of the breeding program.

Scaevola were found to successfully fertilise however seed development appeared to abort before seed could mature. Embryo rescue techniques were employed to germinate immature seed. Further research is required to optimise the germination percentages through timing of harvest. Preliminary studies indicate that harvesting of immature fruit as early as a few days after pollination may result in the best germination percentages although further research is required to confirm this finding.

Brachyscome were found to cross readily and hybrid seed could be successfully germinated using standard seed raising procedures. Both controlled and open pollination trials produced seed and many hybrid plants were assessed and showed promise for the export pot plant industry, however after assessment of the hybrids none were chosen for trialing overseas.

Introduction

The international market for flowering pot plants is very large. This market is always looking for new product lines and a large number are released each and every year. For a number of years Australian native plants have been grown as container plants and flowering pot plants for the European, Asian and American markets.

Scaevola and *Brachyscome* are two genera that have gained significant portions of these markets, and there are 18-20 million units of each genera sold annually. The market in Europe sells 9-10 million units, there are 7-8 million sold in the USA and other markets such as Japan are worth 1-2 million units annually (Confidential market information). A severe limitation to the expansion of these two genera in the markets is the limited colours that are currently available for these products. Recently new colour forms have been identified that could be used in breeding programs to greatly expand the colour lines that could be developed for these products.

For breeding programs in *Scaevola* and *Brachyscome* to be successful, the reproductive and floral biology of these genera must be understood to overcome the reproductive barriers that currently exist. Once these barriers are removed, the establishment of a market orientated breeding program will produce lines that can significantly increase the markets for these crops.

Materials & methods

Plant material

The *Scaevola* and *Brachyscome* selections and cultivars used in the hybridisation program were chosen from commercially sourced cultivars and species, and natural populations.

The *Scaevola* varieties were included in the breeding program based on commercially desirable characteristics. These characteristics included compact growth habit, flower size, flower colour and the time from germination till flowering of the plant. Similarly, desired characters for parents in the *Brachyscome* breeding program included; compact growth habit, flower size, flower number, flower colour and attractive foliage.

Agronomy

Scaevola and *Brachyscome* species and cultivars were propagated in September-October each year using cutting material. Suitable material was taken from motherstock plants that were growing either in an unheated polyhouse or from plants growing in garden beds at a field site in Cranbourne. Vegetative (non-flowering) side shoots were used for *Scaevola*, and cuttings were approximately five to eight centimetres long, with the soft, growing tip removed. *Brachyscome* cuttings ranged in size; from approximately two centimetres for the smaller varieties, to around five centimetres for the larger varieties. All flower buds were removed. The cuttings were misted and the basal ends dipped in IBA powder (3000 ppm). The cuttings were then placed in seedling punnets containing a 50:50 peat and perlite cutting mixture. Punnets were placed on steel mesh benches, covered with clear lids and then with shade cloth. They were located in a polyhouse where they were watered every two days, or more frequently when required. During the warmer weather, the punnets were placed on corflute before being covered with clear lids to reduce evaporation but still provide adequate drainage.

The polyhouse used for the propagation and initial cultivation was a large, unheated structure. The walls were raised on warm days to reduce temperature and improve ventilation.

Once cuttings had developed roots (approximately 4 - 8 weeks for *Scaevola*, 4 - 6 weeks for *Brachyscome*), they were potted into 75mm tubes containing a native plant potting mix (Biogrow) and Osmocote® (18:2.6:9.1) was applied at a rate of approximately half a teaspoon per plant. Plants were then grown under 50% shadecloth for a few days to harden off, then placed on open benches in the polyhouse. Irrigation was provided daily via an automated overhead irrigation system. After the plants had grown to fill the tubes, they were potted into 150mm pots and fertilised with approximately one teaspoon of native Osmocote® (as above).

Established plants in 150mm pots were transported to the Department of Primary Industries, Knoxfield where they were placed in a glasshouse with good air movement and temperatures controlled within the ranges of 15 - 26º C. Light levels were maintained between 180 and 460 µmol m^{-2} s^{-1} by an automatic screen on a 10 minute delay. The plants were irrigated daily via drippers placed in each pot. Plants were pruned at least once per year to encourage flowering and maintain healthy growth and fertilised every 4 – 6 months with native Osmocote® (as above).

Pests and diseases

The overall cultivation conditions in the mistbeds, glasshouse, nursery and field areas were monitored visually on a weekly basis for pest and disease problems. Sticky traps were also used to monitor for pests which could then be identified. The areas were kept as pest and disease free as possible using appropriate insecticides and fungicides as required.

Crop Health Service at IHD, Knoxfield were used to identify unfamiliar pests and diseases.

Scaevola breeding

Reproductive biology

Floral structure and development

To optimise the timing of pollinations, flowers of the main *Scaevola* species and cultivars for the breeding program were investigated under a dissecting microscope. Buds and flowers at different stages of development were examined to gain an understanding of the reproductive structures and their development in these species and the sequence and timing of critical stages. Drawings and notes were taken with anthers, indusium, stigma, style and ovaries observed for changes.

Pollen dehiscence and the pollen presentation system were examined and different stages of development were noted. These were observed to establish the ideal stage of floral maturity to perform emasculations prior to controlled pollinations. The female components of the flower (gynoecium) were observed to identify the location of the stigmatic surface.

Pollen viability and stigma receptivity

Fresh pollen grains from *Scaevola aemula* 'Purple Fanfare', *S.* OB#1 and *S. ramosissima* pink were germinated on microscope slides coated with a thin layer of a 1% agar solution containing 10% sucrose and 0.008% boric acid. Pollen grains were scattered on the slides using a small paintbrush and the slides placed in sealed petri-dishes containing a saturated tissue. The dishes were incubated at room temperature for approximately one hour and then examined under the microscope for the presence or absence of pollen germination and pollen tube growth.

To determine the period of stigmatic receptivity, stigmas of different developmental stages were excised and examined for visual changes. The stigmas were then stained with histochemical stain (Toluidine Blue) to identify areas of the stigmatic surface where the cuticle was broken. Freshly excised styles and stigmas were stained for 1 minute by placing a drop of the staining solution on a microscope slide. The stain was then rinsed from the tissue with water. Excess water was removed carefully with paper towel. The stigma was then examined under a dissecting microscope.

Stigma location and receptivity were also determined by observing pollen germination and growth on the stigma surface. Open pollinated flowers from *S. aemula* 'Purple Fanfare' grown in garden beds at Cranbourne were harvested at four different stages of development as described in Table 1.

Table 1. Floral developmental stages of *Scaevola*

Stage	Floral appearance
1	Indusium not yet full length, petals not yet open.
2	Indusium fully extended, pollen presented on indusium.
3	Stigmatic lobes protruding, no pollen left on indusium.
4	Stigma fully extended past indusium, petals senescing.

Four flowers from each stage were collected and the gynoecium of each flower was placed in a 2:1 solution of ethanol and lactic acid for two hours to fix the specimen. The stylar tissue then autoclaved at 121° C in a 10% sodium sulphite solution for 15 minutes. The tissue was placed on a microscope slide and a drop of decolourised aniline blue was added to stain callose in the pollen tube walls. A coverslip was then pressed onto the specimen to flatten and spread the tissue over the slide surface. The slides were allowed to stain for 30 minutes, and then examined using fluorescence microscopy.

Pollen storage
Pollen was collected for desiccation and storage trials from two *Scaevola* species, *S. albida* 'Jacob's White' and *S. aemula* 'Purple Fanfare'. Three desiccation methods and four temperature storage treatments were compared. For each species, the pollen was treated in one of three ways; no desiccation, desiccated for three days, and desiccated for seven days prior to storage. The four storage temperatures tested were 3°C, -20°C, -75°C and snap freezing with liquid nitrogen for four minutes, followed by storage at -75°C. The pollen samples were all stored for two weeks.
After the two weeks storage, pollen was removed and placed on agar slides to germinate. There were three replicates for each desiccation and storage temperature combination. Each slide was scanned to determine the spread of pollen grains, and then 20 grains on each slide were counted for the presence or absence of a pollen tube. The length of the pollen tube relative to the width of the pollen grain was also observed.

Embryo rescue
Embryo rescue techniques were investigated as a method to germinate developing seed prior to abortion of the maturing fruit. Selected *Scaevola* flowers were emasculated in the bud stage and covered with a small paper bag to exclude insect pollinators. The best buds for emasculation were located one or two buds above the last open flower. Emasculation was performed by carefully opening the corolla along the line of separation between the lobes on the adaxial surface of the bud. Once opened, the five anthers, either pre-anthesis or after anthesis but before deposition of the pollen on the indusium were removed using fine forceps. Between nine and twelve days later, when the stigma was protruding past the indusium, interspecific pollen was used to pollinate the flower. Pollen was placed on the stigmatic surface with a small paintbrush. Flowers were then re-covered with the bag and monitored weekly for the appearance of fruit development.

Immature fruit were harvested at different stages of development, from 12 to 30 days after pollination. Fruit were excised from the plant, and the flesh encasing the seed was removed. Seed were surface sterilised for one minute in 70% ethanol and then for approximately three minutes in 2% sodium hypochlorite containing 2 drops of Tween 20®. The seed were then rinsed in sterile distilled water.

Following surface sterilisation, one end of the seed was cut to penetrate the seed coat and expose the tissue inside the seed. The seed was then placed into petri dishes containing *Scaevola* multiplication tissue culture media, ensuring contact between the exposed seed tissue and the media surface.

The petri dishes were covered in aluminium foil to exclude light, and placed in a temperature controlled growth room maintained at 22° C. The dishes were monitored regularly for changes in appearance of the seed.

Different types of media were investigated to identify the suitable conditions for embryo rescue. A trial was designed to establish the effect of growth hormones on Murashige and Skoog media. Half strength MS media was prepared and divided in half with 0.1 mg zeatin/L added to one half and 1.0 mg each of zeatin, BAP and kinetin in the second half. Embryos were plated onto the media under sterile conditions and the plates observed.

A second trial investigated a new multiplication media preparation (Lunghusen's formulation). Again the media was divided in two, half containing 0.5mg Benlate/L for control of fungal contamination, while the other half contained no Benlate. Embryos were plated onto the media under sterile conditions and placed in a temperature controlled growth room maintained at 22° C and observed.

Breeding program
Season one:
An overall breeding program was designed to incorporate all the *Scaevola* varieties collected. Four varieties of *Scaevola aemula*, two varieties of *S. albida* and one variety each of *S. striata*, *S. ramosissima* and *S. enantophylla* were incorporated into the program. The number of controlled pollinations to be performed for each variety was prioritised.

Scaevola flowers were emasculated by the method described in the section on embryo rescue and bagged to exclude undesirable pollen. Pollen of the desired parent was placed on the stigma after the flowers had developed. The flowers were rebagged and the fruit allowed to develop.

Fruit formed were allowed to remain on the plant until mature, ie. the fruit was allowed to abscise from the pedicle naturally, without mechanical assistance. The bags used to cover the pollinated flowers prevented the dispersal of mature fruit. Seed was extracted/allowed to release naturally from the fruit.

Season two:
Hybridisation for the second flowering season placed emphasis on parental combinations controlled pollinations that were not completed in the first season.

Seed germination
Each seed was surface sterilised as per the procedure described in the section on embryo rescue. Before the seed were placed into half strength MS tissue culture medium, a small part of the seed coat was removed under the dissecting microscope. Petri dishes were then placed under lights to allow germination. When seeds germinated and seedlings were at the second leaf pair stage, they were removed from tissue culture and potted into seed raising mix in 2.5 cm tubes and fertilised with native Osmocote® (18:2.6:9.1). The tubes were then placed into a mistbed in a controlled glasshouse environment. When plants filled the tubes they were repotted, fertilised with native Osmocote® (as above) and transferred to glasshouse benches.

Mutation breeding
The effect of irradiation treatment on *Scaevola* to induce point mutations was investigated. Two *Scaevola* cultivars in 75mm tubes, *S. albida* 'Royal Pink' and *S. aemula* OB1B were taken to Steritech, Dandenong, Victoria where they were irradiated at 5 different levels 25, 50, 75, 100, and 150 Gray to determine the optimal exposure to a source of radiation required to induce mutation. The plants were transported back to Outback Plants at Cranbourne and placed on steel benches in a polyhouse with overhead irrigation every second day. After two weeks the plants were potted up into 250mm pots, fertilised with native Osmocote® (as above) and returned to the polyhouse. They were liquid fed periodically with Vital® 5ml /L. The plants were monitored weekly over the following 6 months.

Evaluation of hybrids
Hybrids resulting from the breeding program were assessed visually against S. 'Purple Fanfare', S. 'Fandancer', S. 'Royal Pink' and S. 'Jacob's White'. The criteria included foliage type/shape, flower colour, fan diameter, ray length and general habit and vigour.

Brachyscome breeding

Reproductive biology

Floral structure and development

To optimise the timing of pollinations, flowers of the main *Brachyscome* species and cultivars for the breeding program were investigated under a dissecting microscope. Buds and flowers at different stages of development were examined to gain an understanding of the reproductive structures and processes in these species and the sequence and timing of critical stages. Drawings and notes of floral structures were made with anthers, stigma, style, and ovaries clearly identified.

The pollen presentation system was examined and flowers at different stages of development were dissected to determine the timing of pollen dehiscence and pollen dispersal. This was necessary to establish the ideal stage of floral maturity to perform controlled pollinations.

The female components (gynoecium) of individual florets were dissected and observed to identify the location of the stigmatic surface.

To determine the location and period of stigmatic receptivity, stigmas of different developmental stages were excised and examined for visual changes. Visual markers such as papillae on the surface of the stigma were examined.

The stigmas were then stained with a histochemical stain (Toluidine Blue) as described in the section on *Scaevola*.

Hybridisation program

The breeding schedule concentrated on 4 superior parental types and planned to perform over 1500 controlled pollinations using 15 parents.

Season one:

Of the four female parental types selected for the program, three were 'mini' *Brachyscome*, and one was a 'jumbo' *Brachyscome.* Mini *Brachyscome* refers to the group of species and cultivars with many small flowers and a fine, compact foliage, whereas jumbo *Brachyscome* have large flowers and foliage. The breeding program prioritised the selected parental types and reciprocal crosses were included in the program. The small size of the flowers prohibited emasculation. Two techniques were employed to perform controlled pollinations. The first involved selecting flowers where at least half the florets on the capitulum were open and dusting the whole capitulum with donor pollen. The second technique involved targeting the female-only ray florets. Individual florets were observed using magnifying glasses and pollen placed on the lobes of the stigma using forceps or fine paintbrush. Flowerheads were then covered with paper bags and were monitored weekly to determine fruit development and maturity.

Season two:

Observations made during the first breeding season indicated that several of the female parents set minimal, if any, fruit. As a result, the emphasis of the second season's breeding program was on those varieties and cultivars that produced the most fruit. The cultivars that set little or no fruit were still used as pollen donors.

Seed harvest

All seed and fruit produced were harvested when the receptacle had enlarged and the fruit were dry and ready to disperse. Capitula were cut from the plant and the seed manually extracted.

Seed germination
Seed were sown in seedling punnets containing Debco® Seed Raising Mix and covered lightly with fine grade vermiculite. The punnets were placed on steel mesh benches in a polyhouse and were irrigated via an automated micro irrigation system. Once seed had germinated, Multigard® Snail and Slug Bait was applied. Plants were grown on and potted up into 75 mm pots with native plant potting media (Biogrow) and fertilised with native Osmocote® (18:2.6:9.1).

Evaluation of hybrids
Hybrids resulting from the breeding program were assessed using eight criteria. The criteria included foliage type/shape, stem length, stem colour, flower colour, bud colour, flowerhead diameter and general habit and vigour. In addition, the presence or absence of chlorosis in the seedlings foliage was noted. The seedlings that showed characteristics such as short flower stems, compact growth and desirable flower colour were kept for further assessment.

Open pollination
An open pollination program was designed to utilise the efficiency of natural pollinators such as honeybees and hoverflies. Selected species and cultivars were planted 30cm apart in 5 beds that were 2m by 20m. For each cultivar, all capitula were harvested from every second plant in the bed. The harvested material was collected in a bucket then transferred into bread trays lined with paper. The material was allowed to dry for approximately one week before being sieved to clean the seed. The seed was then treated as per the seed germination technique described above.

In addition to this trial, seed was collected at Outback Plants Cranbourne from plants grown in garden beds or pots in polyhouses. Seed from these plants was collected and germinated as per the seed germination technique described above.

Mutation breeding
The effect of irradiation treatment on *Brachyscome* to induce point mutations was investigated. Five *Brachyscome* cultivars in 7.5cm tubes, *B.* Jumbo Mauve, *B.* 'CJ City Lights', *B.* 'Metallic Blue', *B.* 'Mauve Delight', *and B.* Maureen #2 were taken to Steritech (Dandenong Victoria), where they were irradiated at 5 different levels 25, 50, 75, 100, and 150 Gray to determine the optimal exposure to a source of radiation required to induce mutation. The plants were transported back to Outback Plants at Cranbourne and placed on steel benches in a polyhouse with overhead irrigation every second day. After two to three weeks the plants were potted up into 25cm pots, fertilised with native Osmocote® (18:2.6:9.1) and returned to the polyhouse. The plants were monitored weekly over the following 6 months.

Results & Discussion

Cultivars collected

Scaevola cultivars

Plant material was collected from 2 genera and included 1 species of Goodenia, and 4 species of *Scaevola*. In total 13 selections were obtained. Cultivars collected were *Goodenia ovata, S. aemula* 'Purple Fanfare', *S. aemula* 'Fandancer', *S. aemula* OB#1C, *S. aemula* OB#1white sport, *S. aemula* OB #6, *S. albida* 'Jacob's White', *S. albida* 'Royal Pink', *S. enantophylla, S. striata* 'Pink Perfection', *S. striata* 'Misty Blue', *S. ramosissima* pink and *S. ramosissima* low blue/mauve.

Brachyscome cultivars

Brachyscome plant material collected included 4 species and 19 cultivars. In total, 24 selections were obtained. Cultivars collected were *B. angustifolia* lemon, *B. angustifolia* mauve, *B. angustifolia* pink, *B.* Barker #6, *B.* Barker #14, *B.* 'Bright Eyes', *B.* 'City Lights', *B.* 'CJ Bicolour', *B.* Cowes Pink, *B. curvicarpa*, *B.* Darlings Downs pink, *B.* 'Fern Leaf White', *B. formosa* mauve, *B. formosa* 'Pillaga Posy', *B.* Barker #1, *B.* Jumbo Mauve, *B.* Jumbo Yellow, *B.* 'Lavender Mist', *B.* 'Mardi Gras', *B.* Maureen #2, *B.* 'Mauve Delight', *B.* 'Mauve Mystique', *B.* 'Metallic Blue', *B.* Mini Yellow syn. *B.* 'Lemon Twist', *B.* 'Misty Yellow', *B.* 'Misty Lilac', *B.* 'Misty Pink', *B.* 'Moonlight' syn. *B.* 'White Delight', *B.* 'Purple Myst', *B. segmentosa*, *B.* 'Strawberry Mousse', *B.* 'Sunburst, *B.* 'Sunset', and *B. tatei*. The plant material collected from natural populations and nurseries were generally propagated readily by cuttings. The majority of cultivars were easily cultivated.

Pest and diseases

White flies, aphids, root-eating larvae, caterpillars, slugs and snails, pythium and powdery mildew needed to be controlled in the glasshouse. Fungal problems were controlled using Fongarid® systemic fungicide. Leaf eating caterpillars were controlled by Dipel® HG Bio-insecticide. White flies and aphids were controlled using Pyrethrum sprays. Root eating larvae were controlled using Gesapon® 800. EcoGrow® "Fungus gnatem" (beneficial nematodes) was also applied.

Scaevola breeding

Reproductive biology

Floral structure and development

Flowers in *Scaevola* are characteristically radial symmetrical during the bud stage with flowers becoming zygomorphic as the corolla tube splits open to form the typical 'fan' shape of the genus. The genus typically has 2 ovules but sometimes has one or four locules (and ovules) depending on species. A secondary pollen presentation system exists where pollen from the anthers is captured in a cup-like structure at the top of the style called an indusium. Pollen is shed and collected through co-ordination of elongation of both the anther filaments and style prior to the flower opening. The indusium folds over the mass of pollen protecting it until the flower opens. Flowers spend approximately 5 days in the male phase before entering the female phase for 4 days. The growth of the stigma forces the pollen from the indusium. It was noted that soon after hand pollination, many flowers would begin to senesce through abscission of both the style and corolla. Howell (1995) also noted that flowers would senesce after nine days if left unfertilised but would senesce within 24 hours if fertilisation took place.

Scaevola spp. appear to be self-incompatible as self-pollen that remained on the stigmatic surface either did not germinate or was arrested before the pollen tube entered the stigmatic pore. According to Howell (1995) this is a result of two barriers preventing self-fertilisation in *Scaevola*; physiological and genetic. Only dehydrated pollen is considered mature and can adhere to the

stigmatic surface and germinate. Pollen collected in the indusium during bud development remains hydrated and will not germinate. Any self pollen which then dehydrates and comes in contact with the stigma can germinate, but development is arrested very soon after.

Pollen viability and stigma receptivity

The timing of floral development meant that the one or two buds above the last open flower on a stem were at the optimum stage for emasculation to take place. Stigmas excised and examined under the microscope indicated that the stigma was receptive from the time it protrudes through the indusium. Stigmas treated with Toluidine Blue only stained an area of the surface in the older flowers. Flowers pollinated nine days after emasculation resulted in stigma abscission 24 to 48 hours after pollination, indicating fertilisation had taken place. Observations after pollination confirmed fruit development. This was observed in a number of cultivars although some required up to 12 days between emasculation and pollination for optimal results.

Stigma receptivity was examined through stylar squashes of open pollinated flowers at different stages of development. Four distinct stages of stigmatic development were identified and only in stage 3 and 4 was pollen germination observed and therefore the stigma receptive (Table 2).

Table 2. Pollen germination on open pollinated stigmas of different maturities

Stage	Observations
1 Bud closed, indusium not extended 1-2 days pre opening	Phloem and xylem visible along style. Pollen grains not found on stigma.
2 Flower open, indusium fully extended, pollen on indusium	Phloem and xylem visible. Pollen grains present but no sign of germination on stigma.
3 Stigmatic lobes protruding past indusium, no pollen on indusium	Pollen grains on stigma – many arrested on papillae on indusium. One pollen tube visible along style.
4 Stigma fully extended past indusium, petals senescing	Arrested pollen grains visible on stigma, pollen tubes along full length of the style.

Howell 1995, states that fertilisation is optimal during the female phase of the flower with fertilisation approaching 100%. However fertilisation rates of 80% may be achieved during the male phase. This may be due to of the longevity of *Scaevola* pollen and the necessity of the pollen to dry out before being capable of germinating, thus pollination carried out with pre-dried pollen during the male phase could result in germination when the stigma becomes receptive.

Pollen Storage

The in-vitro pollen germination trial confirmed that pollen was viable in *S aemula* 'Purple Fanfare' and *S. albida* 'Jacob's White' at 33% and 48% respectively (Table 3). The pollen germination trial indicated that germination was low in in-vitro, although adequate to fertilise the 2 ovules in each flower with a low number of grains applied to receptive stigmas.

Table 3. *In vitro* Pollen Viability

Pollen	Grains counted per slide	No. slides observed	Total grains germinated	Germination percentage
S. aemula 'Purple Fanfare'	20	4	27	33.75%
S. albida 'Jacob's White'	20	3	29	48.33%

The pollen samples collected from *S. aemula* 'Purple Fanfare' and *S. aemula* 'Jacob's White' that were prepared and tested for the pollen storage trial indicated 3 days desiccation followed by storage at -20°C to be optimal. There was a difference in germination percentages between the two cultivars trialled, however both displayed the best results under the same conditions (Table 4).

Table 4. Pollen storage trial summary statistics

Pollen	Pretreatment	Storage temp (2 weeks)	No. pollen grains counted	No. pollen grains germ'd	% germ.	Pollen tube length*
S. 'Purple Fanfare'	fresh - 0 days	3°C	60	5	8.3	10
S. 'Purple Fanfare'	fresh - 0 days	-20°C	60	0	0.0	
S. 'Purple Fanfare'	fresh - 0 days	-75°C	60	0	0.0	
S. 'Purple Fanfare'	fresh - 0 days	liq N to -75°C	60	1	1.7	10
S. 'Purple Fanfare'	desicc 3 days	3°C	60	9	15.0	3-20
S. 'Purple Fanfare'	desicc 3 days	-20°C	60	18	30.0	3-20
S. 'Purple Fanfare'	desicc 3 days	-75°C	60	5	8.3	10-20
S. 'Purple Fanfare'	desicc 3 days	liq N to -75°C	60	6	10.0	5-10
S. 'Purple Fanfare'	desicc 7 days	3°C	60	5	8.3	20-50+
S. 'Purple Fanfare'	desicc 7 days	-20°C	60	2	3.3	10-20
S. 'Purple Fanfare'	desicc 7 days	-75°C	60	3	5.0	10-50
S. 'Purple Fanfare'	desicc 7 days	liq N to -75°C	60	9	15.0	3-50
S. 'Jacob's White'	fresh - 0 days	3°C	60	7	11.7	6-20
S. 'Jacob's White'	fresh - 0 days	-20°C	60	2	3.3	3-6
S. 'Jacob's White'	fresh - 0 days	-75°C	60	3	5.0	5-10
S. 'Jacob's White'	fresh - 0 days	liq N to -75°C	60	2	3.3	20
S. 'Jacob's White'	desicc 3 days	3°C	60	9	15.0	5-20
S. 'Jacob's White'	desicc 3 days	-20°C	60	27	45.0	3-10
S. 'Jacob's White'	desicc 3 days	-75°C	60	17	28.3	3-10
S. 'Jacob's White'	desicc 3 days	liq N to -75°C	60	8	13.3	3-10
S. 'Jacob's White'	desicc 7 days	3°C	60	5	8.3	20-50+
S. 'Jacob's White'	desicc 7 days	-20°C	60	5	8.3	5-50
S. 'Jacob's White'	desicc 7 days	-75°C	60	5	8.3	5-20
S. 'Jacob's White'	desicc 7 days	liq N to -75°C	60	4	6.7	3-10

* Pollen tube length is expressed as a factor of the diameter of the pollen grains

Embryo rescue

A proportion of the developing seed from the hybridisation program was harvested while immature and placed into tissue culture. This premature harvest and *in vitro* culture of the developing embryos was done to prevent embryo abortion prior to full seed development, and resulted in

growth of hybrids. It was found that harvesting the seed prior to the hard seed coat being formed resulted in better germination rates due to the difficulties experienced in excising the fragile embryo from the seed coat. The hard seed coat tended to form at approximately 3 weeks in *S. aemula* cultivars and at almost 4 weeks in other species.

The half strength Murashige and Skoog media with hormone treatment gave poor germination. Seeds that did germinate exhibited poor growth and often died. A second multiplication media (Lunghusen's formulation) resulted in much better germination and subsequent growth of *Scaevola* embryos and seedlings. Most of the hybrids obtained from the program were germinated on this media.

Hybridisation program

The breeding program used the main desirable forms for overseas markets, introducing variations in flower colour and size from various species and cultivars.

The flowering response of some cultivars, and pollen availability severely limited the number of crosses that could be achieved during the first season. The season was extended in the second season using pollen storage to be used later in the season, and artificial lighting to extend day length and promote flowering for longer.

In total, 218 controlled pollinations were conducted using 4 species and 7 parental types. Flowers were monitored carefully for signs of fertilisation and fruit development. Developing fruit was collected from the crosses performed, and this included inter-specific hybrid seed, which was not expected due to previously documented incompatibility between *Scaevola* species.

Forty-five open pollinated seed, thirty-five intra-specific hybrid seed and six inter-specific hybrid seed resulted from the breeding program and were placed in tissue culture. The seedlings that germinated were potted on, hardened off, and are growing under glasshouse conditions.

A summary of the breeding activities, the seed that germinated and the number of resulting seedlings that grew large enough to be placed in the mistbed and eventually potted on are provided in Table 5.

Table 5. Summary of activities in Scaevola breeding program.

Female	Male	Emasculated	Pollinated	Total harvested	No. seed germinated	No. Mistbed	No. Mature
S. ‘ Purple Fanfare’	S. ‘ Pink Perfection’	14	14	1	0		
S. ‘ Purple Fanfare’	S. ‘ Purple Fanfare’	12	10	0			
S. ‘ Purple Fanfare’	S. ‘Jacobs White’	20	20	0			
S. ‘ Purple Fanfare’	S. OB #1C	12	5	0			
S. ‘ Purple Fanfare’	S. OB #6	12	9	1	0		
S. ‘ Purple Fanfare’	S. r low blue/mauve	12	8	0			
S. ‘ Royal pink’	S. ‘ Pink Perfection’	1	1	1	1	1	0
S. ‘ Royal pink’	S. ‘ Royal pink’	4	4	4	1	1	1
S. ‘ Royal pink’	S. ‘Jacobs White’	15	13	13	2	2	2
S. ‘ Royal pink’	S. OB #1C	1	1	1	0		
S. ‘ Royal pink’	S. OB #6	1	1	1	0		
S. ‘Fandancer’	S. ‘ Jacobs White’	22	22	0			
S. ‘Fandancer’	S. ‘ Purple Fanfare’	12	9	0			
S. ‘Fandancer’	S. ‘ Royal pink’	8	8	0			
S. ‘Fandancer’	S. OB #1C	11	17	0			
S. ‘Fandancer’	S. OB #6	12	12	0			
S. ‘Jacobs White’	S. ‘ Fandancer’	1	1	1	0		
S. ‘Jacobs White’	S. ‘ Royal pink’	12	11	11	3	3	2
S. OB #1C	S. ‘ Pink Perfection’	12	12	0			
S. OB #1C	S. ‘ Purple Fanfare’	20	20	4	4	2	0
S. OB #1C	S. ‘Jacobs White’	6	6	0			
S. OB #1C	S. OB #6	20	19	0			
S. OB #6	S. ‘ Purple Fanfare’	12	7	1	0		
S. OB #6	S. ‘ Royal pink’	12	1	1	1	0	
S. OB #6	S. ‘Fandancer’	12	2	0			
S. OB #6	S. ‘Jacobs White’	11	10	0			
S. OB #6	S. OB #1 - white	12	1	1	0		
S. OB #6	S. OB #6	10	8	0			
	Totals	413	252	41	12	9	5

Codes for cultivars

S. ‘Purple Fanfare’ =	*S. aemula* ‘Purple Fanfare’
S. ‘Royal Pink’ =	*S. albida* ‘Royal Pink’
S. ‘Fandancer =	*S. aemula* ‘Fandancer’
S. ‘Jacob’s White’ =	*S. albida* ‘Jacob’s White’
S. OB #1C =	*S. aemula* OB # 1C
S. OB # 1 – white	*S. aemula* OB # 1 - white
S. OB #6 =	*S. aemula* OB #6
S. r low blue/mauve =	*S. ramosissima* low blue/mauve

Mutation breeding

Plants exposed to all irradiation levels displayed unusual development over the weeks following irradiation. No plants produced flowers and all eventually died in the 6 months post treatment.

Evaluation of hybrids

5 controlled pollination *Scaevola* hybrids have reached maturity and have been assessed. One hybrid shows promising characteristics. A description of each of the hybrids is given below in Table 6.

Table 6. Scaevola Hybrid and Parental Characteristics

Hybrid Name	Habit	Diameter of fan (mm)	Length of ray (mm)	Flower description	Flower colour RHS Charts	Foliage
S. 'Jacob's White' X S. 'Royal Pink' # 1	Spreading, semi-upright floriferous	19	9	Flowering towards end of branches. Pink petals with white base (25%). Floriferous	Purple 75C	Leaves stalked and ovoid. Vary in size, largest at base
S. 'Jacob's White' X S. 'Royal Pink' # 2	Spreading, semi-upright	20	13	Flowering towards end of branches. Deeper pink petals, paler at base. White 15%, Floriferous	Red Purple 70C	Leaves stalked and ovoid. Vary in size, largest at base
S. 'Royal Pink' X S. 'Jacob's White' # 1	Spreading, semi-upright, flowering sparse	16	11	Flowering towards end of branches. Soft pink petals with white base (10%)	Red-Purple 65A	Leaves stalked and ovoid. Vary in size, largest at base
S. 'Royal Pink' X S. 'Jacob's White' # 2	Very compact upright	N/A	N/A	N/A	N/A	Leaves stalked and ovoid. More uniform in size than others at this stage
S. 'Royal Pink' X S. 'Royal Pink' # 1	Upright	N/A	N/A	N/A	N/A	Leaves stalked and ovoid. Vary in size, largest at base
S. 'Royal Pink' OP # 1	Spreading, semi-upright, well bracnhed	N/A	N/A	N/A	N/A	Leaves stalked and ovoid. Vary in size, largest at base
Parents						
S. albida 'Royal Pink'	Spreading, semi-upright	17	9	Flowering towards ends of branches, Deep pink petals, small flowers	Red Purple 70C	Leaves stalked and ovoid. Vary in size, largest at base
S. albida 'Jacob's White	Spreading	21	13	Flowering towards end of stem, floriferous. White petals, larger flowers	White	Leaves stalked and ovoid. Vary in size, largest at base

All hybrids which have flowered and been assessed to date exhibit characteristics intermediate between the two parents, with variation in colour intensity, hue and the number and size of flowers produced. Two hybrids are compared to the parental material in Figure 1 below.

Figure 1. Comparison of two *Scaevola* hybrids with parents.

Clockwise from top left:
S. albida 'Royal Pink'
S. albida 'Jacob's White'
S. 'Jacob's White' X *S.* 'Royal Pink' # 1
S. 'Royal Pink' X *S.* 'Jacob's White' # 1

The embryo rescue work showed the most promise for the production of new forms of *Scaevola*, although with the limited time, the project had only limited scope to produce the first hybrids. Further work is needed on embryo rescue in *Scaevola* to produce a range of hybrids.

Brachyscome program

Reproductive biology

Floral structure and development

Brachyscome 'Mauve Delight' florets are arranged in a spiral or whorl on the receptacle. The outermost whorl of florets are reproductively female only and form the ray florets of the capitulum. The remaining disc florets are bisexual comprising five anthers fused longitudinally surrounding a cleft stigma. The stigma has papillose sterile terminal appendages and there is a single ovule in the ovary. Anthers often have apical appendages.

The ray florets have no stamens and the stigma splits completely to form a 'V' shape. The inner receptive stigmatic surface is yellow with the remaining style tissue white.

As the disc florets develop, pollen is released and the stigma is pushed through the cuff of anthers by the elongation of the style. The arms of the style open after the stigma has pushed through and protrudes beyond the senescing anthers. In some of the species examined, the style arms do not completely split but remain fused at the apex forming a 'chinese lantern' shape. The receptive stigmatic surface is inside the 'lantern'.

Hybridisation program

The flowering response of some cultivars, and pollen availability has limited the number of crosses that could be conducted. 1794 controlled crosses were conducted resulting in 1300 seeds. From these 31 plants developed to maturity and were assessed for commercial potential. The characteristics of the hybrids are described in Table 7.

Open pollination:

Open pollinated seed provides the opportunity to obtain a large number of seedlings although a lot of the seedlings are similar to the maternal parent indicating a large amount of self pollination is occurring. Several promising hybrids were produced from seed collected from flower heads that were open pollinated. Capitula from the 12 open pollination trial cultivars and from 135 cultivars and hybrids from various locations at Outback Plants were harvested. This resulted in 661 plants that developed to maturity and were assessed for commercial potential as at 30/6/04. Open pollination resulted in prolific seed production from many of the cultivars.

Mutation breeding

Plants exposed to all levels of irradiation displayed unusual development over the weeks following irradiation. No plants produced flowers and all eventually died in the 6 months post treatment.

Evaluation of hybrids

Of the 692 hybrids obtained, 661 were through open pollination and 31 from controlled pollination. They have been assessed for both commercial potential and for the presence of desirable breeding characteristics. A list of hybrids and their characteristics obtained through the controlled breeding program are given in Table 7. Two hundred and two hybrids were identified as showing promise from the open pollination program and were kept for further observations and assessment. A complete list of these hybrids and their characteristics are given in Table 8.

Table 7. Characteristics of hybrids from controlled pollination

Hybrid Code	Foliage type								Chlorosis		Stem length				Stem colour	Flower colour	Bud colour	Flower size			Vigour and habit
	seg	angst	LM	M#2	DD pink	JM	Cu	form	Yes	No	< 5 cm	5 - 10 cm	10 - 20 cm	> 20 cm				1 - 2 cm	2 - 3 cm	3 - 5 cm	
JM X SM #1	X					X				X		X	X		red older	white	white with pink tinge			>5	
LM X angst pink # 1			X						X				X		green						yuck!
LM X angst pink # 2			X							X			X		red older	pale mauve	dark mauve			X	stems too long
Seg x Cu # 6						X	X			X			X		green	very pale lemon	lime yellow		X		longish stems
Seg x Cu # 10						X	X			X			X	X	green	very pale yellow/cream			X		
Seg x Cu # 11							X			X			X		green	very pale yellow/cream			X		
Seg x Cu # 12							X			X			X		green	very pale yellow/cream	orange, yellow tips		X		
Seg x Cu # 7							X			X			X		green	deep yellow flowers fading	sunset		X		lots branching
Seg x Cu # 8							X			X			X		green	very pale yellow	orange, yellow tips		X		large flowers
Seg x Cu # 9							X			X				X	green	very pale yellow			X		large flowers but very long stems
Seg x JM # # 10						X				X			X		green	white/pink back			X		
Seg x JM # 1	X					X				X			X		red older	white, pink under	pink			X	
Seg x JM # 1						X				X			X		green	white	white			X	like B. segmentosa
Seg x JM # 11						X				X			X		green	white/pink back			X		
Seg x JM # 12	X					X				X				X	green	white, pink back				X	
Seg x JM # 13							X			X			X		green	very pale yellow			X		
Seg x JM # 14						X			X				X		green	purple	purple		X		very chloritic
Seg x JM # 15						X				X			X		red	white/pink back	dark pink			X	large flowers
Seg x JM # 2	X					X				X		X			green	white	white		X		short stems
Seg x JM # 3	X					X				X			X		green	white	white		X		small flowers

Hybrid Code	Foliage type								Chlorosis		Stem length				Stem colour	Flower colour	Bud colour	Flower size			Vigour and habit
	seg	angst	LM	M#2	DD pink	JM	Cu	form	Yes	No	< 5 cm	5 - 10 cm	10 - 20 cm	> 20 cm				1 - 2 cm	2 - 3 cm	3 - 5 cm	
Seg x JM # 4						X				X			X		green	white/pink back	pink		X		
Seg x JM # 5						X				X			X		green	white/pink back	pink		X		
Seg x JM # 6						X				X			X		green	white/pink back	pink		X		
Seg x JM # 7						X				X			X		green	white/pink back	pink		X		
Seg x JM # 8						X				X			X		green	white/pink back	white with pink tinge		X		
Seg x JM # 9						X				X			X		green	white/pink back				X	huge flowers, but long stems
Seg x JM # 2	X					X				X			X		green	white pink under	pink			X	
SM X Cu # 3			X	X						X			X	X	red tinge	pale yellow, green disc	pink and yellow			X	very open, straggly, flowers good
SM X Cu # 1				X						X			X		green	pink	pink sepals		X		great habit - rosette
SM X Cu # 2			X	X						X			X	X	red tinge	pale yellow, pink edge	orange/pink			X	very open, straggly, flowers good
SM X JM # 1				X						X		X	X		pink young then green	pink	deep pink		X		good foliage (rosette), floriferous

Angst pink = *B. angustifolia* pink
Cu = *B. curvicarpa*
DD pink = B. 'Darling Down's pink'
Form = *B. formosa*
JM = *B.* 'Jumbo Mauve'
LM = *B.* 'Lavender Mist'
M#2 = B. 'Maureen #2'
Seg = *B. segmentosa*
SM = *B.* 'Strawberry Mousse'

Table 8. Characteristics of promising hybrids resulting from open pollination

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
Angst pink OP # 32				X						X		X	X		Red	bright pink	salmon pink		X		well branched
Angst pink OP # 33				X						X			X		dark older stems	yellow	dark yellow tips		X		basal growth
Angst pink OP # 36				X						X		X			very dark purple	purple/pink			X		basal rosette
Angst pink OP # 37				X					X			X			green	mauve	pale cream				
Angst pink OP # 38				X						X		X			red	dark pink			X		unusual - vigorous growth
Angst pink OP # 39		X		X						X		X			green	dusky pink			X		
Angst pink OP #40				X						X			X		red	yellow, pink back	dark		X		thick stems, dark tinged leaves
Angst pink OP # 61		X								X		X			green	yellow fading	orange yellow tips		X		floriferous, self cleaning!
Angst pink OP # 64				X					X		X	X			green	dark pink (cerise)	lolly pink		X		very compact, some chlorosis
Angst pink OP # 66		X							X			X			orange	cream	pink, yellow tips			X	
Angst pink OP # 68				X					X			X			green	purple/mauve	dark pink	X			
Angst pink OP # 69				X					X			X			dark	pale yellow/cream			X		vigorous growth
Angst pink OP #70				X						X		X			green	pale pink		X	X		compact, unusual flower colour
Angst pink OP #72			X	X						X			X		orange	lavender-mauve	mauve			X	
Angst pink OP #72				X						X			X		orange	pale yellow	lime yellow		X	X	upright , branching
Angst pink OP #74				X						X?	X				green	pale pink	bright pink		X		compact growth, short stems
Angst pink OP #75				X						X		X			orange	pink mauve	pale pink		X		unusual colour, well branched
Angst pink OP #79				X						X?	X				green, red older	candy pink		X			very like B. angustifolia pink
Angst pink OP #80		X		X						X			X		red older stems	bright yellow, pink underneath	yellow		X		great colour

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
Angst pink OP #81				X					X?		X				green	candy pink			X		like M # 2, good growth
Angst pink OP #82				X					X?			X			red older stems	M#2 pink	M#2 pink		X		like M # 2, good growth
Angst pink OP #84				X						X		X			red older stems	pink	salmon pink		X		good branching, short stems
Angst pink OP # 92		X								X				X	red young, green	yellow	orange	X			Like B. 'Mauve Delight' flowers
Angst pink OP # 96					X					X			X		red older stems	pink	dark pink		X		may reflex?
Angst pink OP #95				X						X		X			green	pink		X			cup-shaped flowers, less reflexing
Angst pink OP # 100					X					X			X		orange, pink older	bright pink	pale	X			cup shaped flowers
Angst pink OP #102				X					X?			X	X		red older stems	yellow		X			shorter than other yellows
Angst pink OP # 103				X						X		X			red older stems	pink	white		X		good foliage
Angst pink OP #106					X					X		X			orange	pale pink	salmon pink		X		nice flowers, large disc
Angst pink OP # 107				X						X		X			green	pink			X		good foliage
Angst pink OP # 110				X						X			X		green	yellow	yellow		X		shorter than other yellows
Angst pink OP # 111					X					X			X		red older stems	pink			X		stems too long, foliage good
Angst pink OP # 117		X			X				X?			X			green	pink		X			very fine foliage and flowers
Angst pink OP # 118				X						X			X		red	mauve	purple		X		great foliage but open habit
Angst pink OP # 120		X		X					X?			X			green pink old	deep pink				X	very open habit
Angst pink OP # 125			X	X						X		X			green	pale mauve/pink			X		interesting foliage and plenty of buds
Angst pink OP # 126		X								X			X		red older stems	bright yellow	yellow	X	X		cup shaped flowers, great colour
Angst pink OP # 127			X							X			X		red/orange old	pink/mauve				X	
Angst pink OP # 128				X						X		X			green	pink/mauve			X		columnar growth, good foliage

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
Angst pink OP # 131			X							X			X		red older stems	deep mauve	pink/purple			X	cup shaped flowers
Angst pink OP # 135		X		X						X			X		orange	pale sunset	yellow	X			cup shaped flowers, rosette foliage
Angst pink OP # 137		X								X			X		red older stems	deep yellow	lime yellow		X		great colour
Angst pink OP # 139		X								X				X	orange	deep yellow	peach/yellow		X		reflexing but good colour
Angst pink OP # 143		X								X			X		orange	yellow tinged mauve	pink, yellow tips		X		
Angst pink OP # 145			X							X			X		pink older	pink, mauve	pink		X		cup shaped flowers
Angst pink OP # 148		X		X						X		X			Pale, red older stems	pale yellow, dark tips			X		great colour
Angst pink OP # 151				X						X		X	X		pale, red older stems	pink			X		great branching, no reflexing
Angst pink OP # 155				X						X		X	X		green, red older stems	mid pink			X		cup shaped flowers
Angst pink OP # 158		X		X						X		X			green, red older stems	pink		X			great habit, well branched
Angst pink OP # 160				X					X			X			green, red older stems	pink, dark centre			X		large flowers but chloritic
Angst pink OP # 161		X								X	X				green	pale pink	white	X			tight habit, short stems
Angst pink OP # 162				X						X		X			dark young, green	pale pink	salmon pink		X		tight habit short stems
Angst pink OP # 163		X								X?	X				dark young, green	bright pink	candy pink	X			suckering?
Angst pink OP # 165		X								X		X			green, red older stems	bright pink	salmon	X			good branching
Angst pink OP # 167			X							X			X		green, red older stems	dark mauve	dark mauve		X		great colour
Angst pink OP # 168		X		X						X		X			green, red older stems	pink	dark pink	X			good branching
Angst pink OP # 169		X		X						X			X		green, red older stems	pale yellow			X		green disc, very nice flower
Angst pink OP # 170		X		X						X				X	green	bright yellow	citrus yellow	X			rounded ray florets, cup shaped flowers

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	,5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
Angst pink OP # 171		X		X						X			X	X	green, orange older stems	bright yellow	citrus yellow		X		too big but great flowers
Angst pink OP # 172		X		X						X			X		red older stems	bright yellow	pink, yellow tips		X		vigorous
Angst pink OP # 174			X							X			X		dark older stems	pink mauve	pink tips			X	Lavender Mist type
Angst pink OP # 176				X					X?			X			green	pink	salmon pink		X		nice habit, rosette foliage
Angst pink OP # 177				X						X		X			green, red older stems	pink	dark pink		X		
Angst pink OP # 178				X					X?			X			green, red older stems	pink	white		X		
Angst pink OP # 179				X						X		X			green, red older stems	pink	white		X		some petal reflex
Angst pink OP # 180				X						X		X			green, red older stems	pink			X		
Angst pink OP # 182		X		X					X?			X			green	pale pink	white	X			
Angst pink OP # 183		X								X			X		green, red older stems	pink outer white inner			X		great flower colour
Angst pink OP # 185				X						X		X			green, red older stems	pink	pink		X		good habit
Angst pink OP # 187				X						X		X			green, red older stems	pink	salmon pink		X		great flower colour
Angst pink OP # 188		X		X						X		X			green, red older stems	dark pink			X		
Angst pink OP # 189					X					X		X			green	pink	pale pink	X			petals - some reflex?
Angst pink OP # 190		X		X					X?				X		green, red older stems	deep yellow	orange yellow tips		X		
Angst pink OP # 191		X		X					X?			X			green , red older stems	pink	pink	X			
Angst pink OP # 192				X						X		X			green, red older stems	pink			X		cup shaped flowers
Angst pink OP # 195		X		X					X			X			green, red older stems	pale yellow, green disc	yellow		X		habit not great, cup shaped flowers
Angst pink OP # 196		X		X						X			X		green	pale yellow fading	dusky pink		X		big flowers
Angst pink OP # 200		X		X					X?			X			green	cream	very pale yellow		X		

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
Angst pink OP # 201		X							X?			X			green	pale pink	very pale pink	X			suckering well, some reflexing of petals
Angst pink OP # 202				X						X		X			green	dark pink		X			short stems, rosette foliage
Angst pink OP # 203				X						X			X		green, red older stems	light pink	white		X		short stems, rosette foliage
Angst pink OP # 204					X					X			X		green, red older stems	light pink	white	X			
Angst pink OP # 205				X						X		X			green, red older stems	mid pink			X		
Angst pink OP # 211				X	X				X			X	X		green, red older stems	pale pink			X		
Angst pink OP # 212					X					X			X		green	mid pink		X	X		
Angst pink OP # 213				X						X		X			green, red older stems	dark pink	dark pink	X			
Angst pink OP # 214					X					X			X		green, red older stems	pale pink	musk stick pink		X		fine ray florets - reflexing?
Angst pink OP # 225				X						X			X		red older stems	pale pink	dark pink		X		
Angst pink OP # 228		X								X			X		green	pale pink	salmon pink		X		
Angst pink OP # 229			X							X			X		green	pale mauve/pink	mauve		X		
Angst pink OP # 244		X								X?		X			green	mid pink	bright pink	X			dwarf form
Angst pink OP # 245					X					X		X			green	very pale pink	salmon pink	X			tiny compact flowers
Angst pink OP # 246		X		X					X			X			red, bright	mid pink	M #2 pink	X			
Angst pink OP # 247		X		X					X			X			green	candy pink fading	M #2 pink		X		
Angst pink OP # 248		X		X						X		X			green	pale pink, dark at centre	Bright pink		X		
Angst pink OP # 249				X						X		X			dark	purple-pink	dark pink		X		
Angst pink OP # 250		X		X						X			X		green	mid pink fading	dark pink			X	
Angst pink OP # 251		X								X		X			some green, some red	purple pink	cerise		X		

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
Angst pink OP # 252		X		X						X			X		green	blue pink, dark centre	dark pink		X		
Angst pink OP # 253					X					X		X			green	white, pink tinge	salmon pink	X			no reflexing,
Angst pink OP # 254		X								X		X			red older stems	pale outer, darker centre		X			compact
Angst pink OP # 255		X								X		X			red older stems	mauve pink	pink		X		short stems, good colour
Angst pink OP # 272		X		X						X			X		red older stems	white, pink tinge		X	X		
Angst pink OP # 277		X		X						X				X	green	bicolour pink/white	yellow		X		
Angst pink OP # 278		X							X?				X		green	pale yellow, darker tips	sunset		X		
Angst pink OP # 285		X							X?			X			green	candy pink fading	salmon pink		X		
Angst pink OP # 286			X							X			X		dark older stems	mauve	mauve		X		very fine growth
Angst pink OP # 287		X								X		X			dark older stems	M# 2 pink	white	X			tiny flowers, lots of buds
Angst pink OP # 288				X						X		X			red older stems	candy pink		X			
Angst pink OP # 293				X						X		X			red older stems	bright pink	pink	X			cup shaped flowers
Angst pink OP # 294			X							X			X		green	sunset			X		very narrow ray florets (spidery)
Barker # 6 OP # 1				X						X		X			red, dark	candy pink	pink		X		nice foliage, good flower colour
Barker # 6 OP #4			X							X			X		dark	mauve	Mauve			X	dark foliage, almost black
Barker #6 OP # 3				X						X		X			green, red older stems	pink	Dark pink		X		cup shaped flowers
Cu OP # 3							X			X			X		green	Cu yellow	Cu yellow	X			kept for more compact habit
Cu OP # 4							X			X				X	green	Cu yellow	Cu yellow		X		kept for more compact habit
Cu OP # 5							X			X				X	green	Cu yellow	Cu yellow	X			kept for more compact habit
form mauve OP #1				X				X		X		X			green, red older stems	pink mauve	Pink		X		nice branching

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
form mauve OP #2								X	X?			X			green	deep pink/mauve	Pink		X		great growth, short stems
form mauve OP #3								X		X			X		green, red older stems	deep pink/mauve	Dark pink			X	pinched ray florets, large flowers
LM OP # 20			X						X?				X		red older stems	mauve	pale mauve			X	huge flowers, well branched
LM OP # 29			X							X		X			red tinge	pale mauve		X			basal foliage, compact
LM OP # 35			X							X			X		green	pale mauve				X	compact growth
LM OP # 39			X							X		X			orange	yellow with mauve			X		very unusual
LM OP # 41			X							X			X		green	mauve			X		
LM OP # 42			X							X			X		red older stems	mauve				X	huge flowers
LM OP # 44			X							X			X		green	mauve fading	Dusky pink		X		double flowers?, biccolour?
LM OP # 45			X							X			X		green	bicolour mauve/yellow	Dark pink		X		floriferous, double flowers, unusual colour
LM OP #15			X							X			X		green	yellow	dark with yellow tips			X	florets narrow
LM OP #16			X							X		X			green	pale yellow	Purple			X	
LM OP #17			X							X		X			green, dark older	dark yellow			X		compact
LM OP #18			X							X			X	X	green, dark older	Yellow (sunset)	purple yellow tips			X	young flowers purpley yellow
LM OP #43			X							X			X		orange	white				X	rosette foliage, large flowers
LM OP #60			X							X			X		red older stems	pale mauve			X		open habit
LM OP #69			X							X		X			green	white			X		
LM OP #70		X	X							X		X	X		green	white		X			good branching
LM OP #72			X							X	X				green	pale yellow, little mauve		X			very compact yellow
LM OP #75		X	X							X			X		green	cream			X		cup shaped flowers

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
LM OP #78			X							X		X			green, red base	mauve		X			good branching, Mauve Delight like flowers
LM OP #80		X	X							X			X		green, red base	cream and mauve fading			X		branching good
LM OP #82		X								X			X		green	pale mauve				X	looks like Metallic Blue, bigger flowers
LM OP #83			X							X		X	X		red older stems	almost white				X	huge flowers
LM OP #93		X	X							X		X			green	yellow fading to white	yellow	X	X		good branching, large disc, compact form
M #2 OP #2				X						X?		X			green, red older stems	bright pink	dark pink		X		
MG OP #1				X						X		X			green	bright yellow	green		X		
MG OP #15		X		X					?			X			dark	bright yellow			X		
MG OP #20		X								X				X	green	yellow	yellow	X			Very vigorous
MG OP #43				X					X			X	X		green	yellow	dark pink		X		vigorous
MG OP #54			X							X		X			red	purple	purple				
MG OP # 61			X	X					?			X	X		red	yellow	pink yellow tips	X			good branching
MG OP # 63				X					X			X			red	pink	dark pink		X		interesting flower colour and dark foliage
MG OP #67				X						X				X	dark older stems	yellow	yellow		X		many ray florets
MG OP #69				X						X			X		red	yellow	pink yellow tips		X		beautiful bud
MG OP #71				X						X			X		green, older stems red	yellow	yellow orange tips		X	X	shaggy florets, large flowers
MG OP #72		X		X						X			X		green	yellow	yellow	X	X		upright habit
MG OP #79				X					?				X		dark older stems	yellow		X			compact growth, short stems
MG OP #85				X					?			X			red		dark pink	?			vigorous growth
MG OP #90		X		X						X			X		dark	yellow	dark yellow tips		X		short stems but tall upright

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	, 5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
MG OP # 97				X						X			X		red	yellow	pink, yellow tips		X	X	nice flowers
MG OP # 99				X						X		X	X		dark green	yellow	pink	X			small flowers, upright branched growth
MG OP # 102				X						X			X		red	bright yellow			X		
MG OP # 103				X					X?			X			red older stems	pale yellow		X			
MG OP #104		X		X						X		X			dark	mauve/pink	dark		X		interesting flower colour
MG OP #110				X						X			X		green	pale yellow	lime yellow		X		very long upright growth, nice flower colour
MG OP # 112		X							X?		X				green	brilliant pink		X	X		very like Mardi Gras
MG OP # 118		X								X		X			green, red older stems	dark pink	salmon (dark)	X			good habit, floriferous
MG OP # 122		X							X?			X			red older stems	pink mauve	pink	X			habit good
MG OP # 123					X				X?			X			green	mauve, red disc	purple/pink	X?			fine foliage
MG OP # 124						X			X?					X	red older stems	pink/p[urple	white			X	huge flowers - like Shaw?
MG OP # 130				X					X?					X	red older stems	yellow			X		great colour
MG OP # 131				X						X			X		red older stems	yellow	yellow		X		good branching
MG OP # 132				X					X			X			red older stems	pink purple	dark pink	X			good flower colour
MG OP # 133		X							X			X			green	deep pink	pale salmon	X			
MG OP # 134				X					X			X			red older stems	candy pink		X			
MG OP # 138		X							X			X			red older stems	pink purple	white	X			good seed production
MG OP # 140		X							X			X			red older stems	candy pink	pale pink	X			
MG OP # 141		X		X						X			X		red older stems	yellow	yellow	X			
MG OP # 145				X						X			X		red older stems	pale yellow	pink yellow tips		X		

Seedling Code	Foliage type								Chlorosis		Stem Length				Stem colour	Flower Colour	Bud Colour	Flower Size			Vigour and Habit
	Seg	Angst	LM	M#2	DD pink	JM	Cu	Form	Yes	No	,5cm	5 –10 cm	10 – 20 cm	. 20 cm				1 – 2 cm	2 – 3 cm	3 – 5 cm	
MG OP # 146		X		X						X			X		red older stems	yellow	pink yellow tips		X		
OP Seedling # 3 OP#1				X						X		X			green, dark older	pink/purple	pink	X			very unusual flowers, great colour, good branching
OP Seedling # 3 OP#2		X							X		X				green, dark older	pale pink	pale salmon	X			
OP Seedling #3 OP #10		X								X			X		green	very pale sunset	dark pink, yellow tips		X		
OP Seedling #3 OP #11		X	X						X?				X		green	cream	B. ‘Sunset’		X		
OP Seedling #3 OP #12		X								X				X	green, red older stems	very pale B. ‘Sunset’	B. ‘Sunset’		X		
OP Seedling #3 OP #13								X		X		X			green	sunset, disc green	B. ‘Sunset’		X		nice flower colour
OP Seedling #3 OP #14								X		X		X			green, red older stems	dark mauve				X	massive flowers
OP Seedling #3 OP #4				X					X?				X		green, red older stems	bright pink	pink	X			cup shaped flowers
OP Seedling #3 OP #7		X		X						X	X				dark red older stems	pale pink		X			flowers mishapen, but producing seed
OP seedling #3 OP #3				X						X		X			pale, dark older stems	mauve/pink		X			very fine growth
Seg OP # 12	X									X			X		red older stems	mauve-pink fading to white				X	huge flowers, good branching
Seg OP # 19			X							X		X	X		red older stems	mauve			X	X	great flowers - very big!
Seg OP # 39							X			X			X		green	bright yellow fading	lime yellow		X		
Seg OP # 41	X					X				X		X			green	white			X		
Seg OP # 46			X							X			X		green	white, pink near centre				X	fine ray florets
Seg OP # 6						X				X			X	X	red older stems	mauve pink with pink back				X	very jumbo semi-double flowers
SHW # 20 OP #1			X						X?				X		dark older stems	pale mauve	dark mauve			X	cuttings taken
SM OP # 1				X					X			X			green	pale pink			X		

Angst pink = *B. angustifolia* pink, Barker #6 = B. ‘Barker #6’, Cu = B. curvicarpa, DD pink = B. ‘Darling Down’s Pink’, Form = B. formosa, JM = *B.* ‘Jumbo Mauve’
LM = *B.* ‘Lavender Mist’, MG = B. ‘Mardi Gras’, M#2 = B. ‘Maureen #2’, OP seedling = Open pollinated seedling,, Seg = *B. segmentosa,* SHW = Sam’s Hybrid White,
SM = *B.* ‘Strawberry Mousse’

Recommendations - scientific and industry

The international ornamentals industry is large and always looking for new products. The Australian flora provides a great opportunity for the introduction of new products as flowering pot plants, landscape plants and cut flowers. The flora of Australian plants is vast with close to 25,000 species that are foreign to the international ornamentals market. Targeted selection programs can identify species and varieties that can be introduced as new products, but the real opportunity lies in designing breeding programs specifically to combine the most suitable characteristics of these plants. These specifically designed breeding programs may take time to develop the new products, but the range of new products will be greater and have characteristics that are more suitable for their desired end use.

Our work has shown that there are reproductive barriers that will prevent the easy establishment of a horticultural breeding program in *Scaevola*. The timing of emasculation and pollination is important and the further refinement of the embryo rescue techniques is necessary for the full development of a breeding program for this group of plants.

A large number of the open pollinated *Brachyscome* hybrids were found to have interesting combinations of characters, further commercial evaluation of these hybrids will be required before their suitability for use as flowering pot plants will be known.

Bibliography of literature cited

Howell., G.J., 1995. Reproductive Biology and Horticultural Development of *Scaevola*. A thesis submitted for the Degree of Doctor of Philosophy at The School of Botany, University of Melbourne.