Understanding the Cropping Behaviour of Riberry (Syzygium luehmannii)

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Understanding the Cropping Behaviour of Riberry (*Syzygium luehmannii*)

By G M Sanewski

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Foreword

Commercial use of *Syzygium* in Australia is confined mainly to the landscape and home garden market but there is growing interest in processing of the fruit, particularly Riberry (*Syzygium luehmannii*). Currently there are four commercial growers with a total of over 8,000 trees. Several companies seek supplies and it is estimated that approximately 3-5 tonnes of fruit are processed per year. Supply is however inconsistent. Wild harvesting is used to supplement the lack of cultivated supply. Currently all processed product is sold within Australia but potential exists on the export market. Inconsistent cropping and a lack of understanding of cropping behaviour are limiting the expansion of this fledgling industry.

This study highlights the complexity of flowering biology in *Syzygium* and demonstrates how a basic understanding of a species’ fundamental biology is necessary for successful commercial cultivation. This report brings together useful information from previous international research on *Syzygium* as well as providing a basic understanding of flower biology, the nature of fruit set and seediness in riberry. Much of these findings have implications for the cultural management of riberry orchards to optimise fruit set and minimise seed set. It raises the possibility of avenues for genetic improvement.

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This report, an addition to RIRDC’s diverse range of over 2000 research publications, forms part of our Native Plant Products R&D program, which aims to facilitate the development of new rural industries based on plants or plant products that have commercial potential for Australia.

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Craig Burns
Managing Director
Rural Industries Research and Development Corporation
Acknowledgments

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Executive Summary

What the report is about

This report describes a short study into the flowering biology of Syzygium luehmannii (riberry). The findings provide a better understanding of pollination requirements, fruit set and the nature of seediness.

Who is the report targeted at?

The report provides information for growers and intending growers but will be particularly useful for those wishing to further study cropping behaviour and genetic improvement in Australian Syzygiums.

Background

The genus Syzygium is endemic to many countries and is exploited as a fresh fruit, spice and ornamental plant, the most notable being clove, S. aromaticum. Within Australia there is growing interest in processing of the fruit of riberry, S. luehmannii. There is however a poor understanding of flowering biology, particularly, the mechanisms behind fruit set and seediness. This study examined the flowering biology of the Glover’s Seedless variety of riberry. Of particular interest were the effect of pollination on fruit set and the nature of seediness. Seedless fruit is preferred by the industry.

Aims/objectives

The aim was to develop an effective hand pollination technique and use that technique to establish the effect of various pollination treatments on fruit set and seed development. Of particular interest was the requirement for pollination and the nature of seediness.

Methods used

A hand pollination protocol was developed based on published data for clove. Various hand pollination treatments and standard fluorescent microscopy techniques were then used to study the effects of pollination on fruit set and seed development. Seeds were examined and germinated to determine if polyembryony existed.

Results/key findings

Flowering biology in Syzygium can be complex with apomixis, parthenocarpy and polyembryony exhibited within the genus. ‘Glover’s Seedless’ is a variety of S. luehmannii originally selected because of it’s high percentage of seedless fruit. It is possible it is unrepresentative of the majority of wild trees of S. luehmannii.

‘Glover’s’ had a low rate of seed set but is at least partially self-compatible. The percentage of fruit with seed is however likely to increase if a genetically compatible species flowers nearby at the same time. Fruit set occurred parthenocarpically and through fertilisation. The majority of fruit is parthenocarpic. Fruit set under natural conditions was approximately fifteen percent. Sixty to eighty percent of fruit were seedless. Seedlessness is probably related to a high rate of ovule abortion.

Insect pollinators were not necessary for fruit set or seed set. Seed appears to be monoembryonic and mostly the result of selfing.

New plantings are best established from cuttings rather than seedlings as some genetic segregation is expected in seedlings.
Recommendations

It is recommended that growers avoid planting other species of *Syzygium* with a similar flowering time in close proximity to riberry if seedless fruit is preferred. Pollinator activity should not be encouraged where other *Syzygium* species are growing nearby.

Inter-specific compatibility seems high between *Syzygium* species. In addition many of the exotic species produce larger fruits than most Australian species. It might be feasible to explore the potential for genetic improvement of riberry to either increase fruit size or improve cropping consistency. At the very least it would be worthwhile comparing available selections of riberry in a replicated trial to ensure the industry is established with the best genetic material available.

The two commercial varieties, ‘Glover’s Seedless’ and ‘Vic’s Choice’ should be evaluated, along with a seedy type for yield potential. Semi-dwarfing Riberry rootstocks are also worth evaluating.

The effect of irrigation on flowering should be established.
Introduction

The genus *Syzygium* is endemic to many countries including South Africa, South America, South-East Asia and Australia. The genus is exploited as a fresh fruit, spice and ornamental plant. Notable species include the wax jambu (*S. samarangensis*) and clove (*S. aromaticum*). A list of some commercialised species is shown in Table 1. Within Australia, commercial use of *Syzygium* is largely confined to the landscape and home garden market but there is growing interest in processing of the fruit, particularly riberry, *S. luehmannii* Early indications are that reliable production will be the most significant constraint to the expansion of this industry.

<table>
<thead>
<tr>
<th>Species</th>
<th>Common name</th>
<th>Origin</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>S. luehmannii</em></td>
<td>Riberry, Small-leaf LillyPilly</td>
<td>Australia</td>
</tr>
<tr>
<td><em>S. fibrosum</em></td>
<td>Rainberry, Fibrous Satinash</td>
<td>Australia</td>
</tr>
<tr>
<td><em>S. samarangensis</em></td>
<td>Wax Jambu</td>
<td>SE Asia</td>
</tr>
<tr>
<td><em>S. cumini</em></td>
<td>Black Plum, Jamun, Java Plum</td>
<td>SE Asia</td>
</tr>
<tr>
<td><em>S. javanicum</em></td>
<td>Water Apple</td>
<td>SE Asia</td>
</tr>
<tr>
<td><em>S. jambos</em></td>
<td>Rose Apple</td>
<td>SE Asia</td>
</tr>
<tr>
<td><em>S. aromaticum</em></td>
<td>Clove</td>
<td>SE Asia</td>
</tr>
<tr>
<td><em>S. malaccensis</em></td>
<td>Malay Rose Apple</td>
<td>SE Asia</td>
</tr>
<tr>
<td><em>S. aquem</em></td>
<td>Bell Fruit</td>
<td>SE Asia</td>
</tr>
</tbody>
</table>

Flowering response

The riberry (*Syzygium luehmannii*) industry in Australia is still very young and little is known about cropping behaviour or suitable management strategies to encourage consistent fruiting. Some observations suggest an irregular cropping pattern or alternate bearing. Alternate bearing is associated with alternating years of poor flowering whereas pollination and fruit development problems can reduce yields in all years. The first step in understanding behaviour is to understand the flowering response.

*Syzygium* is a genus within the family Myrtaceae. Flowering strategies within Myrtaceae vary from mass flowering species with a short, highly synchronised period to steady state species that flower over a long period of up to 90 days (Lughadha and Proença, 1996). Mass-flowering behaviour appears common in *Syzygium*. *S. tierneyanum* is a mass-flowering rain-forest specie from Nth Qld (Lack and Kevan, 1984). Indian blackberry (*S. cumini*) is another mass–flowering species (Reddi and Ramgaiah, 1999). Riberry also appears to exhibit mass flowering. Horticultural selections of the closely related *S. australe* appear to exhibit a steady state response.

It has been postulated that species with abrupt and concentrated flowering such as mass flowering are responding to abrupt changes in environmental stimuli such as might be experienced at the transition between dry and wet seasons in monsoonal sites. As an example, the Brazilian rainforest species *Eugenia dysenterica* appeared to flower in response to a rise in humidity in a 2 year study of flowering behaviour (Proença and Gibbs, 1994).

While there are few studies of flowering reported for *Syzgium* species in Australia, there are reports for the broader family of Myrtaceae. Law *et al.* (2000) reports that of the 20 species of woody-fruited
Myrtaceae (principally eucalypts) studied over a 10 year period, most did not flower regularly from year to year. Pook et al. (1997) observed mass flowering only once in 15 yrs in Spotted Gum (Corymbia variegata) in coastal southern NSW. Flowering, particularly in mass flowering species within this genus, is therefore not necessarily regular. In the study by Law et al. (2000) several environmental variables were recorded and high rainfall in the summer-autumn period was the best correlated with heavy flowering in spring for most of the species. Low temperature during winter was also well correlated with flowering in some species. Keatley and Fletcher (2002) reported a significant relationship between temperature and rainfall and flowering in four species of eucalypts over 23 years.

Fire is also known to stimulate flowering in some Australian plant species with some others showing a high tolerance. However, fire was shown to reduce the intensity of flowering in Syzygium eucalyptoides and ‘Lady Apple’ (Syzygium suborbiculare), (Vigilante and Bowman, 2004).

The environmental stimuli to flowering in riberry are not known but other Syzygium species have been observed flowering in response to changes in water availability and the seasonality of flowering suggests temperature is most likely to be involved. Flooding over 30-40 days is used to induce early flowering in wax jambu (S. samarangense) in summer in southern Taiwan (Lin and Lin, 1992; Liao and Lin, 2001; Crane, 2004) and is used commercially for that purpose but only works in a low percentage of orchards (Shu et al., 1994). ‘Malay Apple’ (S. malaccensis) was observed to flower twice in a single year in Brazil, in March (wet season) and July/August (dry season) (Falcao et al., 2002). In India, ‘Water Apple’ (Syzygium aquem) has been observed to flower twice in a single year (Tarai and Kundu, 2008). In South Africa, flowering in ‘Surinam Cherry’ (Eugenia uniflora) was shown to be dependent on rainfall and temperature while fruit set was dependant on relative humidity (Preez and Welgemoed, 1994). In Australia, S. aquem, which usually flowers in spring, has been observed by the author to produce a minor second flowering in early summer in SE Qld after a hot, dry spring. Flowering in riberry usually occurs in August in SE Qld but has been observed to occur a second time but at a much lower intensity in November in Northern NSW after unseasonal heavy rainfall in spring (R. Glover, personal communication). A specific daylength does not therefore appear obligatory for riberry but temperature and water availability are likely to be involved.

In most tree species, adequate starch levels are required for the production of flowers, particularly mass-flowering species, like most of the Syzygium. Trees with insufficient starch levels may not flower heavily in that year and hence gradually move into an alternate bearing pattern. Limb cincturing or girdling is used in some fruit trees to allow starch to accumulate in the tree and encourage flowering. While adequate starch reserves can be a prerequisite to flowering, environmental factors are usually the trigger.

Alternate bearing can develop gradually as a response to an annual increase in yield that gradually exhausts the tree’s reserves. This results in a mix of trees in the orchard that are cropping or not cropping. Conversely, an abrupt alternation in all trees is more likely related to a single environmental event or cultural practice that promoted a very heavy or very light crop in all trees within the orchard in a particular year (Goldschmidt, 2005).

**Pollination and fruit set**

The flower biology of riberry has not previously been studied but it is known that some trees will produce some seedless fruit. The cultivated varieties ‘Glover’s Seedless’ and ‘Vic’s Choice’ are considered to have a low rate of seediness. Pollination mechanisms and the nature of seediness are not understood.

Published studies in other species indicate that self-compatible and self-incompatible species of Syzygium exist although the former appears the most common. Self-compatible species include S. tierneyanum and S. nervosum from north Australia (Hopper, 1980; Shapcott, 1998), S. cuminii from
India (Reddi and Rangaiah, 1999), S. rubicundum from Sri Lanka (Stacey, 2001), S. lineatum from Indonesia (Lack and Kevan, 1984), and S. samarangense, S. jambos, S. megacarpum, and S. formosum from Thailand (Chantaranothai and Parnell, 1994). There are no reports on the pollination mechanisms in riberry but there might be implications for fruit set and seediness.

Apomixis occurs within the Syzygium genus and is thought linked to polyembryony (Lughadha and Proenca, 1996; Chantaranothai and Parnell, 1994). The species S. cuminii (Kader et al., 2000; Krishnamurthy et al., 1997: Arathi et al, 1996), S. alternifolium (Sha Vlli Khan et al., 1995), S. javanicum (Ikeda, 1979), and S. jambolana (Ladhar and Gill, 1991) have been shown to be polyembryonic. The frequency of polyembryonic seed in Syzygium ranges from infrequent to common. Some authors consider reproduction through sexual mechanisms to be substantially diminished in some species of Syzygium such as S. cuminii and S. jambos to the extent that most seed originate by the development of asexual embryos (Narayanaswami and Roy, 1960). Depending on the species, the origin of the asexual tissues can be integumental or nucellar (Ikeda, 1979; Narayanaswami and Roy, 1960). The asexual plants that arise in polyembryonic species can vary in ploidy. Seedlings, presumably of asexual origin, of S. cuminii were shown to be either, diploid, hexaploid or tetraploid with a decrease in tree growth with an increase in the level of ploidy (Ladhar and Gill, 1991). While not common, riberry has been propagated by seed by some nurserymen. The embryo origin of riberry seedlings might therefore have relevance.

Information on pollination mechanisms is scarce but S. cuminii was shown to be pollinated by wind, insects and gravity (Misra and Bajpai, 1984). There are no reports for riberry.

The S. luehmannii flower possesses a bi-locular ovary with axile placentation (Figure 6A) (Hyland, 1983) similar to that described for S. australe (Belsham and Orlovich, 2003). In Myrtaceae species, the ovary usually contains more embryos than will form seed (Lughadha and Proenca, 1996). Ovule abortion has been studied in S. cuminii and shown to be related to an inhibition of resource up take by the sub-ordinate seeds. Extracts from the dominant seeds, containing predominately indole compounds, were shown to inhibit resource uptake by the sub-ordinate seeds (Krishnamurthy et al, 1997: Arathi et al., 1996). This species usually only develops 1 of 30 ovules to maturity.

The nature of flower biology in riberry therefore has implications for understanding fruit set, seediness and propagation strategies. This study was initiated to better understand flowering, pollination, fruit set and seediness in riberry.
Objectives

The key objectives of this research were to investigate the flower biology of riberry to better understand the influence of pollination on fruit set, fruit retention and seed development; and to better understand cropping behaviour. This was achieved by applying a series of pollination treatments including open pollination, self pollination and nil pollination and pollination by genetically different trees (including other species) and measuring the effect on fruit set and seed development. The effect of cincturing on the flowering response was studied at two sites in one season.
Methodology

This study was conducted over 2 years, 2008 and 2009. In the first year all observations and treatments were conducted on the 7-8 year old cuttings of the ‘Glover’s Seedless’ selection of *S. luehmannii* on the property ‘Galeru’ at Cooroy, west of Gympie. Pollen of the ‘Vic’s Choice’ selection of *S. luehmannii*, also grown on ‘Galeru’, was used in some treatments. The variety, ‘Glover’s Seedless’ was originally selected from a seedling (from a wild tree) in coastal Northern NSW, most likely the Coff’s Harbour Region. It was selected on the basis of a high percentage of seedless fruit. ‘Vic’s Choice’ is an independent selection from Northern NSW, possibly Alstonville. In this study they were considered as genetically different. ‘Glover’s Seedless’ and ‘Vic’s Choice’ are the two main commercial varieties of riberry grown in Australia.

In the second year, ‘Glover’s Seedless’ and ‘Vic’s Choice’ did not flower on the experimental property, Galeru Pty Ltd, so all pollination studies were conducted on three mature seedling trees on Maroochy Research Station. These trees were considered genetically different to each other and ‘Glover’s Seedless’.

Flower development

In 2008, 100 immature buds on one tree of ‘Glover’s Seedless’ were tagged and observed daily for signs of development. These observations were used to determine the stages of development of the flower.

Hand Pollination

2008

Several clusters of approximately 10-50 flowers on each of 2 trees were covered with a glassine paper bag before any opened. The clusters were examined each day for flowers that were ready to be emasculated. Flowers were emasculated when the style had just protruded through the end of the bud but before any stamens had unfolded. Emasculation was achieved using a pair of needle-point forceps and a head magnification lens. The stamens were removed by gently cutting them, the petals and sepals at their base without touching the style. No flowers were allowed to shed pollen within the glassine bags. Those treatments requiring pollination were pollinated daily for at least 7 days using freshly collected pollen. A total of 1,619 flowers were emasculated over 16 days. Table 2 shows the pollination treatments used. Data were analysed by residual maximum likelihood with treatments as a fixed effect and trees and samples within trees as random effects using Genstat 11th edition.
Table 2. Pollination treatments in 2008

<table>
<thead>
<tr>
<th>Treatment code</th>
<th>Protected</th>
<th>Emasc.</th>
<th>Hand pollinated</th>
<th>Pollen source</th>
<th>Number of flowers</th>
</tr>
</thead>
<tbody>
<tr>
<td>PEHV</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Vic’s Choice</td>
<td>488</td>
</tr>
<tr>
<td>PIS</td>
<td>Yes</td>
<td>No</td>
<td>No</td>
<td>Glover’s</td>
<td>565</td>
</tr>
<tr>
<td>UIO</td>
<td>No</td>
<td>No</td>
<td>No</td>
<td>Open</td>
<td>915</td>
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<td>Yes</td>
<td>Yes</td>
<td>Glover’s</td>
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<tr>
<td>PEN</td>
<td>Yes</td>
<td>Yes</td>
<td>No</td>
<td>Nil</td>
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<tr>
<td>UEO</td>
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<td>Yes</td>
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<tr>
<td>UIHV</td>
<td>No</td>
<td>No</td>
<td>Yes</td>
<td>Vic’s Choice</td>
<td>774</td>
</tr>
</tbody>
</table>

Treatment code; P=protected, U=unprotected, E=emasculated, I=intact, H=hand pollinated, S=self pollinated, N=nil pollination, V=Vic’s Choice pollen, G=Glover’s pollen, O=open pollinated.

2009

The same basic technique for pollination was used as in the previous year. The trees were seedlings of unknown origin and each was considered as genetically different to each other and representative of *S. leuhmannii*. Each flower cluster was considered a replication and, in most cases, 4 clusters were treated on each tree. A total of 3,072 flowers were emasculated in 2009. Table 3 shows the treatments. Some treatments were similar to 2008. Additional treatments included selfing each flower twice, selfing each flower at least 5 times and pollination with a range of different species. Species were chosen that flowered at a similar time to riberry. Other *S. leuhmannii* pollen was pollen from different seedling trees considered not directly related to the tree being pollinated. Pollen was used fresh in most cases but occasionally 2 days old. The pollen of all species was checked by fluorescence microscopy to ensure it was viable at the time of collection.
Table 3. Pollination treatments in 2009

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Protected</th>
<th>Emasc.</th>
<th>Hand pollinated</th>
<th>Pollen source</th>
<th>Number of flowers</th>
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<tr>
<td>UEO</td>
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<td>Open</td>
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<tr>
<td>PEHR</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Beach Cherry</td>
<td>241</td>
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<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>(S. reinwardtiana)</td>
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</tr>
<tr>
<td>PEHS</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Self</td>
<td>458</td>
</tr>
<tr>
<td>PEHS</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes, repeated</td>
<td>Self</td>
<td>145</td>
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<tr>
<td>PEHL</td>
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<td>Yes</td>
<td>Yes</td>
<td>Other tree of S. luehmannii</td>
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<tr>
<td>PEHW</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Powderpuff Lilly Pilly (S. wilsonii)</td>
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<td>Yes</td>
<td>Raincherry, Fibrous Satinash (S. fibrosum)</td>
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<tr>
<td>PEHAu</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Scrub Cherry</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>(S. australis)</td>
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<tr>
<td>PEHJ</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>Rose Apple</td>
<td>241</td>
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<td></td>
<td></td>
<td></td>
<td></td>
<td>(S. jambos)</td>
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<td>No</td>
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<td>PEHAn</td>
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<td>Yes</td>
<td>Yes</td>
<td>Aniseed Myrtle (S. anisatum)</td>
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<td>PEHC</td>
<td>Yes</td>
<td>Yes</td>
<td>Yes</td>
<td>‘Cascade’**</td>
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<tr>
<td><strong>Total</strong></td>
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<td></td>
<td></td>
<td>3,902</td>
<td></td>
</tr>
</tbody>
</table>


*’Cascade’ is a PBR Variety (Syzygium wilsonii subsp. wilsonii x Syzygium leuhmanii)

**Stigma receptivity**

Stigma receptivity was studied in 2008. To test for stigma receptivity the flowers were emasculated and hand pollinated then left on the plant for approximately 24 hrs to allow pollen germination and pollen tube growth then collected for examination by fluorescence microscopy. This was done on several separate occasions to minimise environmental effects on any one day.

The flowers were collected into FFA fixative (1 formalin: 1 acetic acid: 8 80% ethanol) and for convenience, stored overnight in a refrigerator. The styles were excised from the flower, soaked in stain for approx 1 minute, mounted in stain (decolourised 0.1% aniline blue in 0.1N K2PO4:H2O) and squashed by gently pressing on the cover slide (Currier, 1957). An Olympus BH2-RFCA microscope with a BP440 filter and HBO 103 w/2 lamp was used. Pollen grains with a pollen tube penetrating the style were counted.

Softening of styles in 1N and 0.1N NaOH at 24°C or 60°C for various periods of a few minutes to one hour to aid staining and squashing resulted in rapid blackening of the styles within a few minutes and was consequently omitted. A small number were softened by autoclaving for 20 min at 140 kPa and 125°C in 5% sodium sulphite, and while this worked well, it was found this step was not essential.
**Pollen viability**

Pollen viability was studied in 2008. Stamens were collected from flowers one day before anthesis (before style emergence), at anthesis (style emergence), and one (stamen emergence) and two days after anthesis. At two days after anthesis the stamens had been shed into the glassine bags. On each occasion the stamens were placed in a solution of sucrose (15 g/L) and boric acid (0.2 g/L) and incubated at 30°C overnight. Fresh solution was used on each occasion. For examination, a drop of germinating solution containing pollen was placed on a microscope slide in aniline blue solution as used for stigma receptivity, then observed using fluorescence microscopy. 200-400 pollen grains were observed on each occasion. Stamens collected before anthesis had not released their pollen and consequently an accurate count could not be achieved at that stage. These pollen grains were however highly fertile with prolific pollen tube growth through the anther walls (Figure 3A).

**Seediness**

All fruit produced in the pollination experiments in 2008 and 2009 were collected at maturity, weighed and checked for the presence of seed. Fruit and seed were counted and weighed. In 2008, fruit from an additional 12 *S. luehmannii* trees growing either on the experimental property or in public places, usually road verges and town streets around Nambour, Noosa and Cooroy in south east Queensland, were also collected and examined for seed. A total of 2,178 fruit were examined. Because these additional trees were of different ages and in different locations, it is assumed they are most likely from a different source and hence genetically different.

**Embryony**

All seed collected from the experimental and additional trees in 2008 were planted in seedling trays, one seed per cell and the number of plants produced per seed counted. An additional sample of 180 seed from a seedy tree were collected in 2009, examined under a binocular microscope for the presence of polyembryony and planted.

**Cincturing**

Cinctures were applied to the main limbs of four trees each month from July to September inclusive at two sites, Cooroy SE Qld and Woolgoolga, NSW in 2009. Four trees were left untreated at each site. In June, the cincture was applied with a 2 mm wide hacksaw blade. In July, a 2 mm wide cincture was applied with a double bladed tool to four trees and a single deep cincture applied with a wheeled tool to four trees at both sites. In August and September, only single deep cinctures were applied.

Flowering and fruiting response were assessed by taking digital images of an interrow side of each tree at budding and again at fruiting using a Canon 20D digital camera. A 1 m scale was included in each image. The images were imported into the digital image analysis software program SigmScan Pro Ver 5.0 for measurement of the 2-dimensional area of flower buds and fruit as a percentage of the tree area. These measurements were used as ratings of flowering and fruiting to compare treatments.

Rainfall, and mean minimum and mean maximum temperatures for both districts in 2008 and 2009 are shown in figures 8 and 9. Cooroy rainfall data was recorded by D Haviland at the Cooroy trial site. This was corroborated with a nearby Bureau of Meterology (BOM) weather station (Station 040059). Cooroy temperature data was taken from the BOM weather station at Nambour (Station 040988). Woolgoolga rainfall data was taken from the BOM weather station at Woolgoolga Clear Place (Station 059039). Woolgoolga temperature data was taken from the BOM weather station at Coffs Harbour (Station 059040).
Results

Flower development

The first flower buds were noticed in the first week of October. The hermaphrodite flowers were borne in groups of two, rarely three, in clusters of 50-100 buds. The flower clusters originate in the terminal 2-4 leaf axil pairs. Anthesis occurred from 20 October to 21 November. There is substantial overlap in developmental stages between flowers within each cluster and throughout the tree. While only a single flowering was observed in this study, anecdotal evidence suggests a second minor flowering, in November, can occur in some years.

Flower development is somewhat similar to that reported for the clove, *S. aromaticum* (Poole and Bermawie, 1986). Buds first developed a whitish cap before a small aperture appeared at the domed apex. The style, bent at 90° to the bud, then emerged and straightened within one day (Figure 1). The stamens emerged the following day. Usually by their second day, the anthers had turned brownish and released their pollen (Figure 1). Nectar was usually present at this time (second day of anthesis). Stamens were shed on the third day of anthesis (Figure 2). Anthesis, from style emergence to stamen shedding was therefore approx four days. The styles were shed on about day nine. The typical flower cycle is shown in Table 4 although there was some variation to this. Observation in 2009 indicated the flower cycle could proceed quicker under unseasonally warm conditions. Flowers could proceed from the small aperture stage to full anthesis in one day.

![Figure 1](image-url)  
A cluster of flowers (flowers are in pairs) of *S. luehmannii* showing flowers at different stages.  
(The dehisced anthers are a slight brown colour)
Table 4. Flower development

<table>
<thead>
<tr>
<th>Day of Anthesis</th>
<th>Stage</th>
<th>Comments</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 1</td>
<td>Style emerges</td>
<td>Initially the style is bent at 90° but straightens over a day.</td>
</tr>
<tr>
<td>Day 2</td>
<td>Stamens emerge</td>
<td></td>
</tr>
<tr>
<td>Day 3</td>
<td>Nectar secreted and pollen released</td>
<td>The anthers turn brown as they dehisce and release pollen.</td>
</tr>
<tr>
<td>Day 5</td>
<td>Stamens shed</td>
<td></td>
</tr>
<tr>
<td>Day 9</td>
<td>Style shed</td>
<td></td>
</tr>
</tbody>
</table>

Pollen viability

Data for pollen germination is shown in Table 5. Peak pollen germination and hence viability occurred on day four of anthesis. This approximately coincided with two days after stamen emergence, that is, when anthers dehisced to release pollen or the day after. Figure 3 shows germinating pollen tubes.

Table 5. Pollen germination

<table>
<thead>
<tr>
<th>Time of collection</th>
<th>Germination (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Day 2 of anthesis (stamens not dehisced)</td>
<td>very viable</td>
</tr>
<tr>
<td>Day 3 of anthesis</td>
<td>32.1</td>
</tr>
<tr>
<td>Day 4 of anthesis</td>
<td>69.4</td>
</tr>
<tr>
<td>Day 5 of anthesis (stamens shed)</td>
<td>28.3</td>
</tr>
</tbody>
</table>

Figure 3. Pollen tubes emerging from anthers (A) immersed in a sucrose-boric acid solution overnight and (B) a single germinating pollen tube with brightly fluorescing callose plugs
Stigma receptivity

The mean number of pollen tubes produced on stigmas sampled at different stages of flower development is shown in Table 6 and is a guide to stigma receptivity.

Table 6. Stigma receptivity

<table>
<thead>
<tr>
<th>Stage</th>
<th>Total number of flowers</th>
<th>Mean number of pollen tubes/style</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nil pollination</td>
<td>9</td>
<td>0</td>
</tr>
<tr>
<td>Cap break (Day 0)</td>
<td>6</td>
<td>0</td>
</tr>
<tr>
<td>Style emergence (Day 1)</td>
<td>61</td>
<td>2.1</td>
</tr>
<tr>
<td>Day 2 (Stamen emergence)</td>
<td>12</td>
<td>5.5</td>
</tr>
<tr>
<td>Day 3 (Pollen release)</td>
<td>18</td>
<td>5.0</td>
</tr>
<tr>
<td>Day 5 (Stamen shedding)</td>
<td>7</td>
<td>4.0</td>
</tr>
<tr>
<td>Day 6</td>
<td>16</td>
<td>3.8</td>
</tr>
<tr>
<td>Day 7</td>
<td>5</td>
<td>2.8</td>
</tr>
</tbody>
</table>

The stigma was weakly receptive as soon as the style emerged, even at the bent stage. Maximum stigma receptivity occurred approximately two days after stigma emergence and coincided approximately with stamen emergence and pollen shedding. The stigma remained receptive for at least two days after stamens were shed. Figure 4 shows pollen tubes growing down the stylar canal.

While the technique used was suitable, the collection of styles 24 hr post pollination was only sufficient to allow a small degree of pollen tube development, usually less than 10% of the length of the style. It is suggested that 48-36 hr would be more appropriate. Pollen-style incompatibility, if it exists, may not manifest until further down the style. Longer pollen tubes might therefore give a better indication of pollen compatibility. It would be preferable to see pollen tube growth the full length of the style.

Figure 4. Fluorescing pollen tubes of *S. luehmannii* growing down the stylar canal
The effect of bagging, emasculation and pollination on fruit set and seediness of Glover’s Seedless

Experimental data for experiments on Glover’s trees conducted in 2008 are shown in Table 7.

Table 7. The effect of bagging, emasculation and pollination on fruit set and seediness in Glover’s Seedless in 2008

<table>
<thead>
<tr>
<th>Treatment</th>
<th>% Fruit set</th>
<th>% seeded fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Mean</td>
<td>SEM</td>
</tr>
<tr>
<td>PEHV</td>
<td>35 a,b,c</td>
<td>5.7</td>
</tr>
<tr>
<td>PIS</td>
<td>18 c,d</td>
<td>7.0</td>
</tr>
<tr>
<td>UIO</td>
<td>15 d</td>
<td>5.3</td>
</tr>
<tr>
<td>PEHS</td>
<td>23 b,c,d</td>
<td>6.0</td>
</tr>
<tr>
<td>PEN</td>
<td>41 a</td>
<td>5.7</td>
</tr>
<tr>
<td>UEO</td>
<td>38 a,b</td>
<td>5.7</td>
</tr>
<tr>
<td>UIHV</td>
<td>15 d</td>
<td>5.7</td>
</tr>
</tbody>
</table>

A different letter within the column denotes a significant difference at the P=0.05 level. SEM=standard error of the mean. Treatment code; P=protected, U=unprotected, E=emasculated, I=intact, H=hand pollinated, S=self pollinated, N=nil pollination, V=Vic’s Choice pollen, G=Glover’s pollen, O=open pollinated. Fruit set of ‘Glover’s Seedless’ under natural conditions (treatment 4) was approximately 15%. That is to say, 15% of buds developed and carried a fruit through to maturity. Eighty-five percent of buds were shed.

Protection and emasculation

Protecting the buds or excluding pollinators with a white glassine bag did not affect fruit set. Emasculation of flowers generally improved fruit set. Fruit set for emasculated, open pollinated fruit (treatment UEO), was 250% higher than that which naturally occurred (treatment UIO). It is possible that part of this response might not be due to the emasculation but rather the fact that some flowers are removed to make emasculating easier. This ‘thinning’ of flower clusters might have led to better retention of the remaining flowers.

Emasculated flowers were capable of setting seed if open pollinated naturally (treatment UEO) but not by hand pollination (treatment PEHS). Emasculation by itself did not reduce seed set but the hand pollination procedure used here appears less effective for fertilisation compared to natural pollination.

Pollination

From Table 6, neither restricting the pollen source to ‘Vic’s Choice’ (treatment PEHV versus PEHS) or assisting open pollination with hand pollination using pollen from ‘Vic’s Choice’ (treatment UIHV versus UIO) affected fruit set.

The use of genetically different pollen (cross pollination) can improve fertilisation and fruit set in many tree crops compared to self pollination. This has been clearly demonstrated in the Australian native macadamia (Wallace et al., 1996). The assumption in this study was that ‘Vic’s Choice’ is genetically different to ‘Glover’s’ and hence their combination would represent cross pollination. It is possible that these two varieties are less genetically different than originally assumed.

Although it was not consistent across all treatments, in one instance, where flowers of Glover’s were emasculated, pollination (treatment PEHS versus PEN) reduced fruit set.
Excluding pollinators did not affect fruit set suggesting most fruit set was probably the result of disturbance by wind or gravity, even though unprotected flowers were regularly visited by a range of potential pollinators, principally honey bees, *Apis mellifera*. This is not surprising as flower stages overlap and the close proximity of flowers would facilitate easy transfer of pollen. Agitation by wind for example would cause flowers to brush against each other. It is also evident that the stigma is approximately in line with the anthers. Studies in *S. cumini* indicated that pollinator activity was necessary in that species (Jai Prakash and Singh, 2007) and that wind, insects and gravity were involved (Misra and Bajpai, 1984).

The hand pollination technique worked well in the early stages as indicated by the stigma receptivity studies where pollen tubes were observed growing down the styles. The development of seed in interspecific crosses in 2008 (data not shown) also indicated hand pollination was effective. Seed did not however occur in emasculated, hand pollinated flowers (treatments PEHV and PEHS) but did in emasculated, open-pollinated flowers (treatment UEO). This suggests hand pollination was not as effective for fertilisation as open pollination. It is suspected that repeated pollination of the same flowers might have disrupted fertilisation or damaged flowers. The hand pollination technique was modified in 2009 to minimise pollination of flowers to a maximum of two times.

**Fruit size**

There were no differences between seeded and seedless fruit in mean fruit pulp weight (data not shown) in ‘Glover’s Seedless’. General observation suggests seeded fruit can be smaller than seedless fruit although data collected in 2008 did not support this hypothesis.

**Seediness**

‘Glover’s Seedless’ appeared to have a lower rate of seediness, as did ‘Vic’s Choice’, compared to many other street trees sampled, although this was not compared experimentally. Under natural conditions within the trial, 60-80% of fruit were seedless. Protecting flowers from pollinators did not affect seed set and it is assumed that most seed was the result of selfing. Self-compatibility was demonstrated with treatment PIS.

The highest percentages of seed set occurred in open and self pollination treatments that allowed pollination but did not include hand pollination. No seed occurred where pollination was prevented.

No seeds were obtained where flowers were emasculated and hand pollinated with *S. luehmannii*. A combination of emasculation and frequent hand pollination (selfing) appeared to substantially inhibit seed set. It is possible that excessive pollination in 2008 was the cause of reduced efficiency of hand pollination compared to natural pollination.

Seed appears to be the result of self fertilisation and not agamospermy because no seeds were obtained where flowers were emasculated but not pollinated. However, apomixis was not directly studied. Apomixis has been reported for *S. jambos*, and in that species was associated with polyploidy. Apomixis has been shown to be enhanced by pollination (Chantaranothai and Parnell, 1994).

A high frequency of fruit with a seed in individual street trees within the region (Table 8) suggests seediness might vary genetically between varieties. There is no indication of the nature of the differences because of the lack of experimental design on this aspect. Given the fact that ‘Glover’s Seedless’ was shown to be self-compatible and pollen tubes were regularly observed growing in the style following selfing by hand pollination, yet seed set did not occur, it is likely that it is unrelated to pollinator activity or proximity of pollinator trees and hence is possibly genetic. It could possibly be related to a pre-fertilisation barrier or ovule abortion.
The presence of 26 ovules per fruit suggests the fruit must abort all but one or two where fertilisation is high. Ovule abortion suggests a selection process for fitness could be occurring. Lughadha and Proença (1996) suggested that only the most vigorous embryos develop in such cases.

‘Glover’s’ appears to be at least partially compatible with some other Syzygium species including *S. wilsonii* (Anon, 2001a), *S. oleosum* (Oliver Carter, personal communication) and in this 2008 study, *Syzygium* hybrid ‘Cascade’, *S. fibrosum* and *S. australe*. The Australian native *S. paniculatum* is reported to be compatible with *S. australe* (Anonymous, 2001b). Several exotic *Syzygium* species have also demonstrated inter-specific compatibility (Ladhar and Gill, 1991) suggesting there might be considerable opportunity for hybridisation between *S. luehmannii* and other *Syzygium* species.

The ability of *S. luehmannii* to both self and out-cross is similar to that described for *S. paniculatum* (Payne, 1997).

Seedling populations of open pollinated *S. luehmannii* appear to segregate into various plant forms and dwarfism appears reasonably common. There are at least 3 dwarf varieties of riberry of seedling origin registered with Plant Breeders Rights in Australia (http://pbr.ipaustralia.plantbreeders.gov.au/search_action.cfm?sort=txtCommonName). In this study there appeared to be approximately 50% vigorous types and 50% smaller types in seedling populations of selfed *S. luehmannii*.

Figure 5. A well formed fruit and seed of *S. luehmannii*.

No seeds were obtained where flowers were emasculated and individually hand pollinated with either ‘Glover’s’ or ‘Vic’s Choice’ pollen. Seed was however obtained where flowers were not emasculated but bagged (treatment PIS) indicating some selfing (autogamous and geitonogamous pollination) occurred. The only possible exception is if apomixis occurred. Apomixis is unlikely because seed did not develop where flowers were emasculated and protected from pollination.

A similar quantity of seed was obtained where flowers were emasculated and open pollinated (treatment UEO) as those that were protected but not emasculated (treatment PIS), indicating cross pollination (allogamous plus geitonogamous pollination) between different flowers was no more effective than selfing (autogamous plus geitonogamous pollination) in fertilisation. It is likely however that open pollination within an orchard of trees of soma-clonal origin represents selfing anyway.
Table 8. Seediness of several different trees of S. luehmannii

<table>
<thead>
<tr>
<th>Selection</th>
<th>Total number of fruit</th>
<th>% of fruit with 1 seed</th>
<th>Number of fruit with 2 seed</th>
<th>Number of fruit with 3 seed</th>
<th>% seeded fruit</th>
<th>Av number of seed in seeded fruit</th>
</tr>
</thead>
<tbody>
<tr>
<td>‘Vic’s Choice’</td>
<td>180</td>
<td>17.2</td>
<td>0</td>
<td>0</td>
<td>17.2</td>
<td>1.00</td>
</tr>
<tr>
<td>‘Glover’s’</td>
<td>243</td>
<td>19.8</td>
<td>0</td>
<td>0</td>
<td>19.8</td>
<td>1.00</td>
</tr>
<tr>
<td>Street Tree 1</td>
<td>114</td>
<td>10.5</td>
<td>0</td>
<td>0</td>
<td>10.5</td>
<td>1.00</td>
</tr>
<tr>
<td>Street Tree 2</td>
<td>211</td>
<td>50.0</td>
<td>0</td>
<td>0</td>
<td>50.0</td>
<td>1.00</td>
</tr>
<tr>
<td>Street Tree 3</td>
<td>57</td>
<td>45.6</td>
<td>19.0</td>
<td>3.5</td>
<td>68.4</td>
<td>1.48</td>
</tr>
<tr>
<td>Street Tree 4</td>
<td>84</td>
<td>57.1</td>
<td>3.6</td>
<td>0</td>
<td>60.7</td>
<td>1.06</td>
</tr>
<tr>
<td>Street Tree 5</td>
<td>400</td>
<td>93.5</td>
<td>6.5</td>
<td>0</td>
<td>99.8</td>
<td>1.07</td>
</tr>
<tr>
<td>Street Tree 6</td>
<td>221</td>
<td>16.7</td>
<td>0</td>
<td>0</td>
<td>16.7</td>
<td>1.00</td>
</tr>
<tr>
<td>Street Tree 7</td>
<td>300</td>
<td>7.7</td>
<td>0.3</td>
<td>0</td>
<td>8.0</td>
<td>1.25</td>
</tr>
<tr>
<td>Street Tree 8</td>
<td>180</td>
<td>85.6</td>
<td>2.8</td>
<td>0</td>
<td>88.3</td>
<td>1.06</td>
</tr>
<tr>
<td>Street Tree 9</td>
<td>120</td>
<td>95.8</td>
<td>1.7</td>
<td>0</td>
<td>97.5</td>
<td>1.02</td>
</tr>
<tr>
<td>Street Tree 10</td>
<td>181</td>
<td>46.4</td>
<td>6.6</td>
<td>0</td>
<td>53.0</td>
<td>1.14</td>
</tr>
</tbody>
</table>

Data (% seeded fruit) in Table 8 indicates a wide range of seediness in S. luehmannii. S. luehmannii is capable of producing 1-2 seeds, usually one (Figure 5). Where two seed were produced they were usually, but not always, in separate locules. In only two instances were three seed found in a fruit.

Examination of fruit without seed usually indicated the presence of 13 ovules per locule (Figure 6) to give 26 ovules per fruit. This is close to the 10-12 ovules per locule reported by Hyland, (1983).

Figure 6. Image A shows a section across a longitudinally cut mature fruit showing two locules with a number of ovules in each locule. Image B shows 13 ovules in one of these locules.

The nature of the low incidence of seed in ‘Glover’s Seedless’ was not studied. It is unclear therefore whether it is a pre-fertilisation barrier, a post-fertilisation barrier or embryo abortion. Pollen viability and stigma receptivity were demonstrated to be good. Given the existence of embryo abortion within the genus and the excessive number of ovules in ‘Glover’s Seedless’, it is likely it is the result of embryo abortion, probably associated with selfing.
The effect of bagging, emasculation and pollination on fruit set and seediness of seedy genotypes

This discussion relates to the seedy types of riberry studied in 2009. Data is shown in Table 9. Fruit set under natural conditions for these seedy types was approximately 40% (UIO), apparently much higher than the 15% recorded for ‘Glover’s Seedless’ in 2008.

Protection and emasculation

There were no differences in % fruit set or % seedy fruit between treatments that were protected or not but otherwise equivalent. As in 2008, protecting flowers with glassine bags did not affect treatments. Emasculation did however reduce fruit set and seediness.

Table 9. The effect of protection, emasculation and pollination on fruit set, seediness and pulp recovery of seedy types in 2009

<table>
<thead>
<tr>
<th>Treatment</th>
<th>% fruit set</th>
<th>% seedy fruit</th>
<th>Pulp Yield/flower</th>
<th>% parthenocarpy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Emasculated &amp; open pollinated (UEO)</td>
<td>18.0 b</td>
<td>71.7</td>
<td>0.03 b</td>
<td>7.1</td>
</tr>
<tr>
<td>Not emasculated but allowed to self (PIS)</td>
<td>25.2 ab</td>
<td>90.7</td>
<td>0.04 b</td>
<td>7.6</td>
</tr>
<tr>
<td>Not emasculated &amp; open pollinated (UIO)</td>
<td>39.7 a</td>
<td>96.2</td>
<td>0.08 a</td>
<td>14.9</td>
</tr>
</tbody>
</table>

* ns * ns

Treatment code; P=protected, U=unprotected, E=emasculated, I=intact, H=hand pollinated, S=self pollinated, N=nil pollination, V=Vic’s Choice pollen, G=Glover’s pollen, O=open pollinated. The inhibitory effect of emasculation on fruit set (UEO versus UIO) is the opposite of that recorded for Glover’s Seedless in 2008. The response seen in 2009 suggests emasculation reduced fertilisation which might then affect fruit retention. It also appears the trees used in 2009 had a lower propensity to set fruit parthenocarpically compared to ‘Glover’s Seedless’ in 2008 (15% versus 60-80%). It follows that highly parthenocarpic trees are expected to retain unfertilised flowers better.

Pollination

The effect of several forms of pollination on emasculated flowers is shown in Table 10.

Xenogamous pollination (cross pollination between different seedling trees) produced the highest fruit set, a 3.4 fold increase over self pollination. Nil pollination of emasculated flowers produced the same % fruit set as open or self pollination. Nil pollination produced no seed whereas all forms of pollination produced the same % seedy fruit.

Parthenocarpic fruit set occurred with self, open, xenogamic and nil pollination. Parthenocarpic fruit set varied from 6-14%. There were no significant differences in the level of parthenocarpic fruit set between any of the treatments.

Self and nil pollination produced greater pulp weight per fruit, but because of the differences in fruit set, there were no differences between treatments in yield of pulp per flower.
Table 10. The effect of various forms of pollination on emasculated flowers of seedy types in 2009

<table>
<thead>
<tr>
<th>Treatment</th>
<th>% fruit set</th>
<th>% parthenocarpy</th>
<th>% seedy fruit</th>
<th>Av Pulp wt / fruit</th>
<th>Pulp yield/flower</th>
</tr>
</thead>
<tbody>
<tr>
<td>Xenogamic (PEHL)</td>
<td>56.5 a</td>
<td>14.0</td>
<td>76.2 a</td>
<td>0.11 c</td>
<td>0.06</td>
</tr>
<tr>
<td>Open (UEO)</td>
<td>18.0 b</td>
<td>6.3</td>
<td>71.7 a</td>
<td>0.14 bc</td>
<td>0.03</td>
</tr>
<tr>
<td>Self (PEHS)</td>
<td>16.7 b</td>
<td>6.8</td>
<td>61.4 a</td>
<td>0.20 ab</td>
<td>0.03</td>
</tr>
<tr>
<td>Nil (PEN)</td>
<td>18.0 b</td>
<td>13.1</td>
<td>0 b</td>
<td>0.23 a</td>
<td>0.04</td>
</tr>
</tbody>
</table>

% parthenocarpy = the % of flowers that set seedless fruit. % seedy fruit = the % of fruit that contains seed.

Pollination with pollen from a genetically different riberry tree substantially improved fruit retention. That is to say, more of the flowers developed into fruit. So while xenogametic pollination stimulated greater fruit set than selfing, the level of seediness was not shown to be different suggesting abortion of fertilised ovules from xenogametic pollination occurred at the same frequency as with self pollination. It was expected, but not demonstrated, that greater abortion, and hence lower seediness, would occur with selfing.

Because all fruit retained without pollination were seedless, apomixis did not occur.

**Inter-specific hybridisation**

The results of inter-specific pollination are shown in Table 11.

Hybrid ‘Cascade’ pollen gave higher fruit set than selfing but similar to xenogametic and S. wilsonii pollen. Nil pollination produced the same % fruit set as selfing. Pollen from all species used was as effective or more effective as self pollen in stimulating fruit set.

‘Cascade’, S. wilsonii, S. anisatum and xenogametic pollen produce a greater % of seedy fruit than S. fibrosum, S. australe, S. reinwardtii, S. jambos and nil pollination.

Pollen from S. wilsonii, ‘Cascade’, S. anisatum and self produced the lowest percentage of parthenocarpic fruit set as most flowers that set fruit were seeded. Apart from S. anisatum, these pollen sources were compatible with riberry. Other pollen sources tended to stimulate fruit set more without seed development.

Nil pollination and pollination with S. jambos gave higher mean pulp weight per fruit than pollination with the hybrid ‘Cascade’ and S. anisatum. However, because of the higher fruit set with the hybrid ‘Cascade’, it produced a higher pulp yield per flower than xenogametic pollen.
Table 11. The effect of inter-specific hybridisation on fruit set, seediness, parthenocarpy and pulp recovery of seedy types in 2009

<table>
<thead>
<tr>
<th>Pollen source</th>
<th>% set</th>
<th>% seedy1</th>
<th>% parthenocarpic fruit set2</th>
<th>Pulp wt/fruit</th>
<th>Pulp yield/flower</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nil pollen</td>
<td>18.0 e</td>
<td>0 c</td>
<td>15.9 abcd</td>
<td>0.21 a</td>
<td>0.04 abc</td>
</tr>
<tr>
<td>S. reinwardtiana</td>
<td>25.3 de</td>
<td>18.8 c</td>
<td>20.0 ab</td>
<td>0.16 ab</td>
<td>0.05 bc</td>
</tr>
<tr>
<td>Self</td>
<td>16.7 e</td>
<td>61.4 b</td>
<td>4.4 de</td>
<td>0.17 ab</td>
<td>0.03 c</td>
</tr>
<tr>
<td>Xenogametic</td>
<td>56.5 bc</td>
<td>76.2 ab</td>
<td>9.0 abcde</td>
<td>0.09 c</td>
<td>0.07 abc</td>
</tr>
<tr>
<td>S. wilsonii</td>
<td>38.9 abc</td>
<td>91.1 a</td>
<td>2.0 e</td>
<td>0.14 abc</td>
<td>0.07 ab</td>
</tr>
<tr>
<td>S. fibrosum</td>
<td>21.4 de</td>
<td>29.9 c</td>
<td>17.5 abc</td>
<td>0.16 ab</td>
<td>0.04 abc</td>
</tr>
<tr>
<td>S. australe</td>
<td>29.5 bcd</td>
<td>20.8 c</td>
<td>21.9 a</td>
<td>0.14 abc</td>
<td>0.05 abc</td>
</tr>
<tr>
<td>S. jambos</td>
<td>29.4 cde</td>
<td>8.2 c</td>
<td>23.4 a</td>
<td>0.19 a</td>
<td>0.07 abc</td>
</tr>
<tr>
<td>S. anisatum</td>
<td>17.5 cde</td>
<td>87.5 ab</td>
<td>6.6 abcde</td>
<td>0.11 bc</td>
<td>0.03 abc</td>
</tr>
<tr>
<td>Hybrid ‘Cascade’</td>
<td>70.0 a</td>
<td>96.5 a</td>
<td>0.2 e</td>
<td>0.12 bc</td>
<td>0.10 a</td>
</tr>
</tbody>
</table>

% seedy = the % of fruit that contains seed. % parthenocarpy = the % of flowers that set seedless fruit.

Embryony

2008

Most seed of S. luehmannii produced only one plant (Table 8). A very small number did however produce a second plant (Figure 7). It is possible however that the two plants which appeared to originate from a single seed actually developed from two seeds that developed in the same locule and weakly adhered to each other. While many other species of Syzygium are known to be polyembryonic (Roy, 1960; Sha Vlli Khan et al., 1995), most Australian Syzygiums are monoembryonic (Hyland, 1983). S. paniculatum is the only recorded polyembryonic Australian Syzygium species. Hyland (1983) describes differences in cotyledon size and shape in polyembryonic seed of S. paniculatum. Cotyledon irregularities were not observed in S. luehmannii.

2009

Closer microscopic examination of seed in 2009 confirmed that seed originating from the same locule can sometimes adhere together and be erroneously considered as a single seed. When seeds weakly adhering together are separated and each planted in a single cell, only single plants are obtained. From 180 seeds, 93 germinated with only single plants obtained. There was no indication of polyembryony in S. luehmannii.
Table 12. Germination and frequency of doubles in *S. luehmannii* seed in 2008

<table>
<thead>
<tr>
<th>Selection</th>
<th>Total number of seed planted</th>
<th>Total number of seed germinated</th>
<th>% germinated</th>
<th>% pots with 2 plants</th>
</tr>
</thead>
<tbody>
<tr>
<td>Vic’s Choice</td>
<td>31</td>
<td>25</td>
<td>80</td>
<td>0</td>
</tr>
<tr>
<td>Glover’s</td>
<td>70</td>
<td>54</td>
<td>77</td>
<td>5.6</td>
</tr>
<tr>
<td>Street tree 1</td>
<td>12</td>
<td>12</td>
<td>100</td>
<td>8.3</td>
</tr>
<tr>
<td>Street tree 2</td>
<td>76</td>
<td>8</td>
<td>11</td>
<td>0</td>
</tr>
<tr>
<td>Street tree 3</td>
<td>51</td>
<td>16</td>
<td>31</td>
<td>6.3</td>
</tr>
<tr>
<td>Street tree 4</td>
<td>54</td>
<td>28</td>
<td>52</td>
<td>0</td>
</tr>
<tr>
<td>Street tree 5</td>
<td>330</td>
<td>292</td>
<td>88</td>
<td>4.5</td>
</tr>
<tr>
<td>Street tree 7</td>
<td>25</td>
<td>12</td>
<td>48</td>
<td>0</td>
</tr>
<tr>
<td>Street tree 8</td>
<td>149</td>
<td>53</td>
<td>36</td>
<td>1.9</td>
</tr>
<tr>
<td>Street tree 9</td>
<td>120</td>
<td>63</td>
<td>53</td>
<td>0</td>
</tr>
<tr>
<td><strong>Total</strong></td>
<td><strong>918</strong></td>
<td><strong>563</strong></td>
<td><strong>61</strong></td>
<td><strong>3.7</strong></td>
</tr>
</tbody>
</table>

The seedlings are phanerocotylar and epigeal with fleshy cotyledons. That is to say, the cotyledons are held at ground level, outside a seed covering, and contain reserves for plant growth. As the seed germinates the short hypocotyl raises the seed to ground level or slightly above and the epicotyl then emerges. The seedling does not emerge from the seed but rather the seed splits into two cotyledons and the seedling grows from embryonic tissue connecting the two cotyledons. This seedling form is similar to that described by Philipson (1989) for *S. maire* but is epigeal rather than semi-hypogeal. Hyland (1983) also described *S. luehmannii* as epigeal.

Figure 7. Two plants apparently growing from two seed fused at the testa
Cincturing

Cinctures applied at both trial sites produced some callusing and a small number of new shoots below cinctures. Trees at both sites were generally dormant during June. A very small number of vegetative shoots appeared in mid June, mainly in the tops of trees at both sites. Bud-break occurred in late August at Cooroy and mid August at Woolgoolga. At Cooroy these were vegetative buds and at Woolgoolga they were a mix of vegetative and floral buds. Anthesis in a few trees outside the trial occurred around early September at Cooroy.

No flowering occurred in any of the trial trees at the Cooroy site in 2009 regardless of cincturing treatment. In contrast, all trial trees at the Woolgoolga site flowered and there were no significant treatment differences. Data is shown in table 13. There were however significant differences between tree rows (blocks) where the two rows higher up the slope cropped heavier than the two lower rows.

Table 13. Crop rating for Woolgoolga cincturing trial

<table>
<thead>
<tr>
<th>Month</th>
<th>Tool</th>
<th>Crop Rating</th>
</tr>
</thead>
<tbody>
<tr>
<td>June</td>
<td>Hacksaw</td>
<td>0.12</td>
</tr>
<tr>
<td>July</td>
<td>Double blade</td>
<td>0.12</td>
</tr>
<tr>
<td>July</td>
<td>Wheels</td>
<td>0.12</td>
</tr>
<tr>
<td>Aug</td>
<td>Wheels</td>
<td>0.13</td>
</tr>
<tr>
<td>Sept</td>
<td>Wheels</td>
<td>0.09</td>
</tr>
<tr>
<td>Control</td>
<td>Nil</td>
<td>0.08</td>
</tr>
<tr>
<td>LSD</td>
<td></td>
<td>0.10</td>
</tr>
</tbody>
</table>

It is impossible for cincturing from August onwards to affect floral bud initiation in riberry as bud initiation must occur prior to this time. It is possible that cincturing earlier than June might have had an effect. Flowering only occurs in the axils of the end 2-4 leaf pairs on shoots produced the previous summer-autumn. It is likely that floral bud differentiation occurs in the May to July period.

While a treatment response was not measured, the general response of trees both in the two trials and generally throughout both districts provides some information. No trees at the Cooroy site flowered yet many trees within the district flowered well. Flowering was strong to moderate in most trees observed from coastal SE Qld to coastal northern NSW. This suggests the general environmental conditions in 2009 were suitable for flowering. It was also observed that new growth at both trial sites occurred mainly in the tops of trees suggesting insufficient light on the lower canopy for growth. At Woolgoolga flowering also occurred mainly in the tops of trees and was almost exclusively on one side of the trees, presumably the side with higher light interception.

Temperature data is shown in figure 8 and rainfall data in Figure 9 for the trial site localities.
There are no obvious major differences between sites or years. The two trial sites were in fact remarkably similar. The mean minimums for June and July were slightly lower in 2008 compared to 2009. The winter period in 2008 exhibited more fluctuation in temperatures.
Rainfall patterns were not consistent with differences between sites and years. It is difficult with the limited and variable nature of the data to draw strong conclusions. Some observations can however be made especially on the patterns over the autumn to spring period when flower initiation is expected to occur. The Cooroy site experienced a dry April and spring in 2008. In 2009 it experienced a wet April but a very dry late winter and spring. Trees did not flower at this site in 2008 or 2009. The Woolgoolga site experienced a much wetter March-June period in 2009 compared to 2008. Trees at Woolgoolga did not flower in 2008 but did in 2009.

Trees at the Woolgoolga site flowered well in 2009. This year was characterised by good rainfall over the February to June period.
Implications

The riberry industry in Australia is only small and will have difficulty attracting new growers and hence achieving a critical mass if production issues are not addressed. Inconsistent cropping is probably the predominant constraint at this stage. While this short study has not provided a clear way to improve cropping it has indicated some potential avenues for further work. Cincturing had no effect on cropping suggesting insufficient starch reserves are probably not implicated in inconsistent flowering although cincturing in April-May is worth testing. The reported response of other Myrtaceae and Syzygium to rainfall and the differences in flowering and rainfall observed between the two trial sites over the two year trial period suggests the effect of water availability on flowering should be examined. The limited data presented here suggests good water availability, possibly in excess of 200 mm/month, in the February-June period is likely to be important for flowering.

This project had demonstrated that yield of seedy types can be increased by cross pollination but with a possible increase in seediness. So while a substantial yield increase has been demonstrated, a practical means of utilising this response is needed. A better understanding of the response of ‘Glover’s Seedless’ to cross pollination with a seedy type might be worthwhile. ‘Glover’s Seedless’, while producing predominately seedless fruit in a clonal orchard, appears to have a low rate of fruit set.

From a different perspective the influence of cross-pollination on Glover’s Seedless might be a factor in seed production where seediness is occurring at a high frequency. While this aspect needs further work to be certain, it would be prudent at this stage to avoid planting other Syzygium species, particularly the hybrid ‘Cascade’ in close proximity to riberry if they are likely to flower at a similar time of year.

Riberry produces a small fruit, much smaller than other horticulturally significance exotic species of Syzygium. Good inter-specific compatibility raises the possibility of inter-specific hybridisation to improve fruit size.

The apparent differences in fruit set efficiency of ‘Glover’s Seedless’ and seedy types suggests there might be advantage in testing several selections in replicated trials for yield attributes. The existence of various forms of riberry from seed raises the possibility of selecting more compact varieties or semi-dwarfing rootstocks.
Recommendations

The RIRDC and ANFIL websites are a logical avenue for disseminating the results of this project to current and intending riberry growers. A peer reviewed technical paper should also be published in the relevant scientific literature to make the results available to other researchers.

Inter-specific compatibility seems high between *Syzygium* species. In addition many of the exotic species produce larger fruits than most Australian species. It might be feasible to explore the genetic potential to either increase fruit size or improve cropping consistency of riberry through inter-specific hybridisation. At the very least it would be worthwhile comparing available selections of riberry in a replicated trial to ensure the industry is established with the best genetic material available. Varieties that should be evaluated for cropping include ‘Glover’s Seedless’, ‘Vic’s Choice’ and at least one seedy type. The existence of a range of plant forms in riberry seedlings also raises the possibility of selecting more compact varieties or semi-dwarfing rootstocks to control tree size. Semi-dwarfing rootstocks are worth including in any varietal comparison.

The effect of unrelated pollen on yield and seediness of ‘Glover’s Seedless’ needs to be confirmed. Early indications are there might be potential for increasing yield but at the expense of increased seediness. Where seedlessness is highly valued, the planting of other varieties and species near commercial orchards of seedless riberry should be avoided.

The effect of irrigation in the March to June period on flowering in riberry should be evaluated.
Glossary

Agamospermy .......... asexual reproduction in which seeds are produced from unfertilised ovules. Equivalent to apomixis. Seed without pollination.
Allogamy .................. cross-fertilisation. Fertilisation between two different trees (two different varieties) of the same species.
Anther ..................... the pollen-bearing part of a stamen.
Anthesis .................... act of expansion in flowers, especially the maturing of the stamens. Flower opening.
Apomixis ..................... reproduction which replaces sexual reproduction, so that all seed are genetically identical. The development of unfertilised seed.
Asexual ...................... no fertilisation.
Bi-locular .................. two compartments in the ovary.
Emasculate .................. to remove the male part. In the case of plants it is removing the anthers which contain the pollen.
Epicotyl ...................... that part of the stem above the cotyledons.
Epigeal ...................... seedlings that have their cotyledons above the surface of the ground.
Fluorescence ................ light emitted by a substance when it is exposed to ultraviolet light. Some plant tissues will fluoresce when treated with specific dyes and then viewed with a microscope using ultra-violet light of a specific wavelength.
Glassine ...................... a glazed, semi-transparent paper with slightly greater tolerance to wetting.
Hermaphrodite ............... a flower having normally both the male and the female organs.
Hypocotyl ..................... that part of the stem below the cotyledons.
Hypogeal ..................... seedlings that have their cotyledons underground.
Integument ................... a skin or covering. Equivalent to the testa of a seed.
Interspecific ................ between two species. Inter-specific compatibility is where two species can be cross pollinated.
Locule ......................... separate cavities in an ovary.
Nucellus ....................... the central cellular mass of the ovule, containing the embryo sac.
Ovule ......................... immature seed.
Parthenocarpy ............... formation of fruit without fertilisation of the ovules by pollination. Parthenocarpic fruit is seedless.
Phanerocotyl ................ the fleshy cotyledons are not held with a seed covering but are exposed.
Ploidy ......................... the number of sets of chromosomes in a cell.
Polyplody ..................... having a chromosome number that is greater than the diploid number.
Polyembryony ............... the production of more than one embryo from one ovule.
Semi-hypogeal ............... seedlings that have their cotyledons at the surface of the ground.
Stamen ......................... the pollen-bearing part of a flower, consisting of the filament and the anther.
Stigma ........................ that part of a pistil which receives the pollen.
Testa ......................... the outer, usually hard, integument or coat of a plant seed.
References


Understanding the Cropping Behaviour of Riberry (*Syzygium luehmannii*)

by G M Sanewski

Publication No. 10/194

Commercial use of *Syzygium* in Australia is confined mainly to the landscape and home garden market but there is growing interest in processing of the fruit, particularly Riberry (*Syzygium luehmannii*).

This study highlights the complexity of flowering biology in *Syzygium* and demonstrates how a basic understanding of a species’ fundamental biology is necessary for successful commercial cultivation. This report brings together useful information from previous international research on *Syzygium* as well as providing a basic understanding of flowering biology, the nature of fruit set and seediness in riberry. Much of these findings have implications for the cultural management of riberry orchards to optimise fruit set and minimise seed set. It raises the possibility of avenues for genetic improvement.

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Cover photo: A cluster of flowers (flowers are in pairs) of *S. luehmannii showing flowers at different stages*